

Alexandrina L. Dumitrescu

# Antibiotics and Antiseptics in Periodontal Therapy



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Springer

*Author*

Dr. Alexandrina L. Dumitrescu  
University of Tromsø  
Institute of Clinical Dentistry  
Department of Periodontology  
9037 Tromsø  
Norway  
[alexandrina.l.dumitrescu@gmail.com](mailto:alexandrina.l.dumitrescu@gmail.com)

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## Preface

Periodontitis is defined as an inflammatory disease of the periodontium characterized by inflammation of the gingival and adjacent attachment apparatus, illustrated by loss of clinical attachment due to destruction of the periodontal ligament and loss of the adjacent supporting bone. Periodontitis represents the major cause of tooth loss in adults leading to long-term disability and increased treatment needs. It is well established that the various periodontal diseases are caused by bacterial infection.

Dental plaque was defined as “matrix-enclosed bacterial populations adherent to each other and/or to surfaces or interfaces”. Dental plaque is a microbial biofilm, a diverse microbial community found on the tooth surface embedded in a matrix of polymers of bacterial and salivary origin. This complex biofilm may offer some protection from the host’s immunologic mechanisms as well as chemotherapeutic agents used for treatment. It is therefore logical to do first the mechanical removal and disruption of the dental plaque biofilm itself.

Antimicrobial agents are used extensively in both medicine and dentistry to eliminate infection. These drugs are also widely used as a prophylaxis to prevent infection in the “at risk” patient. This book is a collection of data from scientific papers and textbooks found to be important for the understanding of the antibiotics and antiseptics used in the periodontal therapy.

This book is addressed to dentists, periodontologists, undergraduate and postgraduate dental students, dental hygienists, and other co-associated professionals.

Alexandrina L. Dumitrescu



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# Antimicrobial Resistance of Dental Plaque Biofilm

Alexandrina L. Dumitrescu and Masaru Ohara

For a decade, many microbiologists have been attracted to new emerging concepts such as polymicrobial diseases, heterogeneous biofilms, and multispecies communities. The recent advent of molecular technologies, namely the 16S rRNA gene clone library, fluorescence *in situ* hybridization, and checkerboard DNA–DNA hybridization, has shed new light on dental biofilm research. We now have a much clearer view of the diversity of oral bacteria present in the human oral cavity. Nevertheless, the available information on dental biofilms remains limited. These technologies have allowed for a fragmented observation of these communities, but a full picture of the bacterial interactions and their functions is still lacking. Furthermore, many bacterial species detected in dental biofilms remain uncultured. To further our understanding, a combination of multiple approaches, ranging from the investigation of pure cultures and *in vitro* biofilm model systems to animal models and human investigation studies, should be undertaken. The development of technologies that enable us to analyze putative functions and metabolisms of a complete dental biofilm may be necessary. Such efforts could contribute to the elucidation of ecological constraints that govern multispecies communities, and help develop novel methods of controlling dental biofilms [42].

Biofilms commonly form in a variety of environments including those relevant to public health. Organisms that grow in a biofilm have distinct advantages, including protection from the effects of antimicrobial agents.

Several mechanisms have been proposed to explain how biofilms convey antimicrobial resistance [19].

## 1.1 Characteristics of Dental Plaque as a Bacterial Biofilm

A biofilm may be defined as a sessile community of cells, characterized by a stable, irreversible union to a substratum, interface, or to each other, embedded in a matrix of extracellular polymeric products which it has synthesized and exhibits a different phenotype with respect to growth rate and gene expression from that of planktonic organisms [78].

Organization of microorganisms within biofilms confers, on the component species, properties which are not evident with the individual species grown independently or as planktonic populations in liquid media [25]. The basic biofilm properties are [66]:

- Cooperating community of various types of microorganisms: Microorganisms are arranged in microcolonies.
- Microcolonies are surrounded by protective matrix.
- Within the microcolonies are differing environments.
- Microorganisms have primitive communication systems.
- Microorganisms in biofilm are resistant to antibiotics, antimicrobials, and host response.

### 1.1.1 Biofilm Formation

At present, we know that **bacteria form biofilms** in essentially the same way, irrespective of the ecosystem

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M. Ohara  
Hiroshima University Hospital, Dental Clinic 1-1-2,  
Kagamiyama, Higashi-Hiroshima 739-0046, Japan  
e-mail: mohara@hiroshima-u.ac.jp

they inhabit. The process of forming a biofilm depends on different variables, such as the species of bacteria, the composition of the surface, environmental factors, and essential gene products, and is regulated, at least in part, by the quorum sensing system. In an oversimplified version, adhesion is mediated, in the **first stage**, by nonspecific interactions, followed, in the second stage by the production of specific molecular interactions (by lectin, adhesin, or ligand). In brief, it is possible to differentiate three phases: primary bacterial adhesion; surface conditioning; and secondary bacterial adhesion. Primary bacterial adhesion comprises a random meeting between a conditioned surface and a planktonic bacterium. This stage is reversible and depends on physicochemical variables. First, the organism must be brought into close approximation with the surface, either propelled randomly by a stream of fluid flowing over a surface, for example, or in directed fashion, via chemotaxis and motility. After that, electrostatic interactions, for example, tend to favor repulsion, because most bacteria and inert surfaces are negatively charged and it seems that this factor is important during primary adhesion. However, primary contact generally occurs between an organism and a conditioned surface and hydrophobic conditions can vary greatly. The **second stage of adhesion** is the anchoring phase, in which binding is mediated by specific molecular adhesins located mainly on pili and fimbriae. At the conclusion of this phase, in the absence of physical and chemical intervention, adhesion becomes irreversible and the organism is firmly

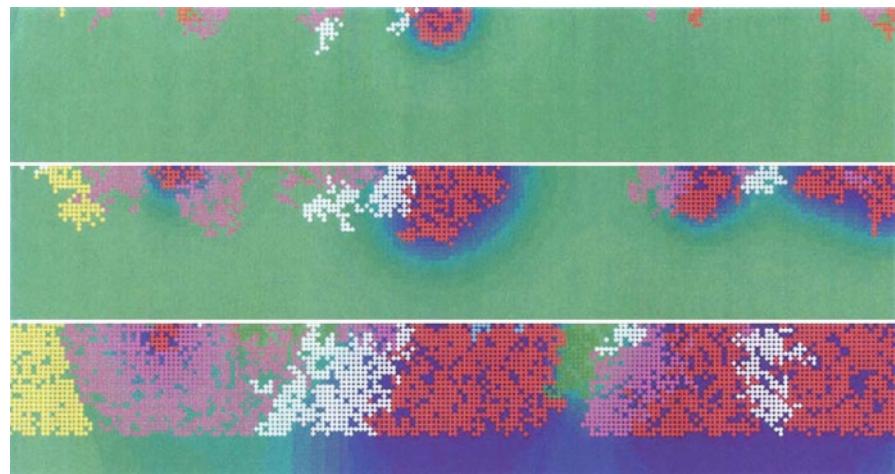
attached to the surface. During the adhesion stage, planktonic bacteria of other species may also be included in the biofilm, forming aggregates on the surface. Biofilm growth and maturation continue to the point where the biofilm reaches a critical mass; dynamic equilibrium is reached when the outermost layer of growth begins to generate planktonic organisms [45, 53, 78].

Many attempts have been made to mathematically model biofilms, both regarding their structure and metabolic processes, and to explain the (lack of) efficacy of antimicrobials. This approach is worthwhile, because a detailed mathematical simulation of biofilm processes could advance our general knowledge and be predictive for choosing antibacterial approaches. In addition, formulating a simulation model pinpoints the questions of interest and is – in general – illustrative in identifying the gaps in one's knowledge. Figure 1.1 shows a snapshot of the simulation of a constant composition film fermentor (CDFF) biofilm, taking into account the growth of various species using stochastic processes and diffusion (mutacins and nutrients) [92].

### 1.1.2 Architecture of Dental Plaque Biofilm

Dental plaque can be defined as “**matrix-enclosed bacterial populations adherent to each other and/or to surfaces or interfaces**” [11, 12].

**Fig. 1.1** Computer modeling of constant composition film fermentor (CDFF) biofilm formation (in cross section), taking into account growth of various species (*expressed in colors*), using stochastic processes and diffusion. Superimposed: mutacin production by red species (*in blue*) [92] (Reprinted with permission of the Society of The Nippon Dental University)



As recently Herrera et al. [37] revealed, dental plaque is a **microbial biofilm** that shares most of the features of other currently known biofilms [6, 13, 15], with antimicrobial resistance being of special relevance [29].

In the subgingival biofilm, four different layers could be distinguished: The first layer of the biofilm is composed of cells displaying little fluorescence relative to cells in the top of the biofilm. Of all the probes tested, only *Actinomyces* sp. gave a positive signal in this layer. The intermediate layer is composed of many spindle-shaped cells of which *Fusobacterium nucleatum*, *Tannerella Forsythia*, and possibly other *Tannerella* sp. The top layer of the biofilm and part of the intermediate layer contain mainly bacteria belonging to the *Cytophaga–Flavobacterium–Bacteroides* cluster (CFB cluster) as detected with probe CFB935 and shown in panel D. CFB935 positive cells are filamentous, rod-shaped, or even coccoid. Samples double-stained with CFB935 and *Tannerella*-specific probes showed that most filamentous bacteria are *Tannerella* sp., while many of the rod-shaped bacteria are *Prevotella* sp. and *Bacteroidetes* sp. as detected with the group-specific probes PREV392 and BAC303, respectively. Besides the presence of bacteria from the CFB cluster, large cigar-like bacteria were in the top layer. These cells belong to the *Synergistetes* group A of bacteria and form a “palisade”-like lining. They were in close contact to eukaryotic cells resembling polymorphonuclear leukocytes (PMNs) according to the presence of polymorph nuclei. Outside the biofilm, a fourth layer without clear organization was observed. *Spirochaetes* were primarily localized in the fourth layer where they are the most dominant species. Bacterial aggregates, called rough and fine test-tube brushes were detected between the *Spirochaetes* [111] (Fig. 1.2).

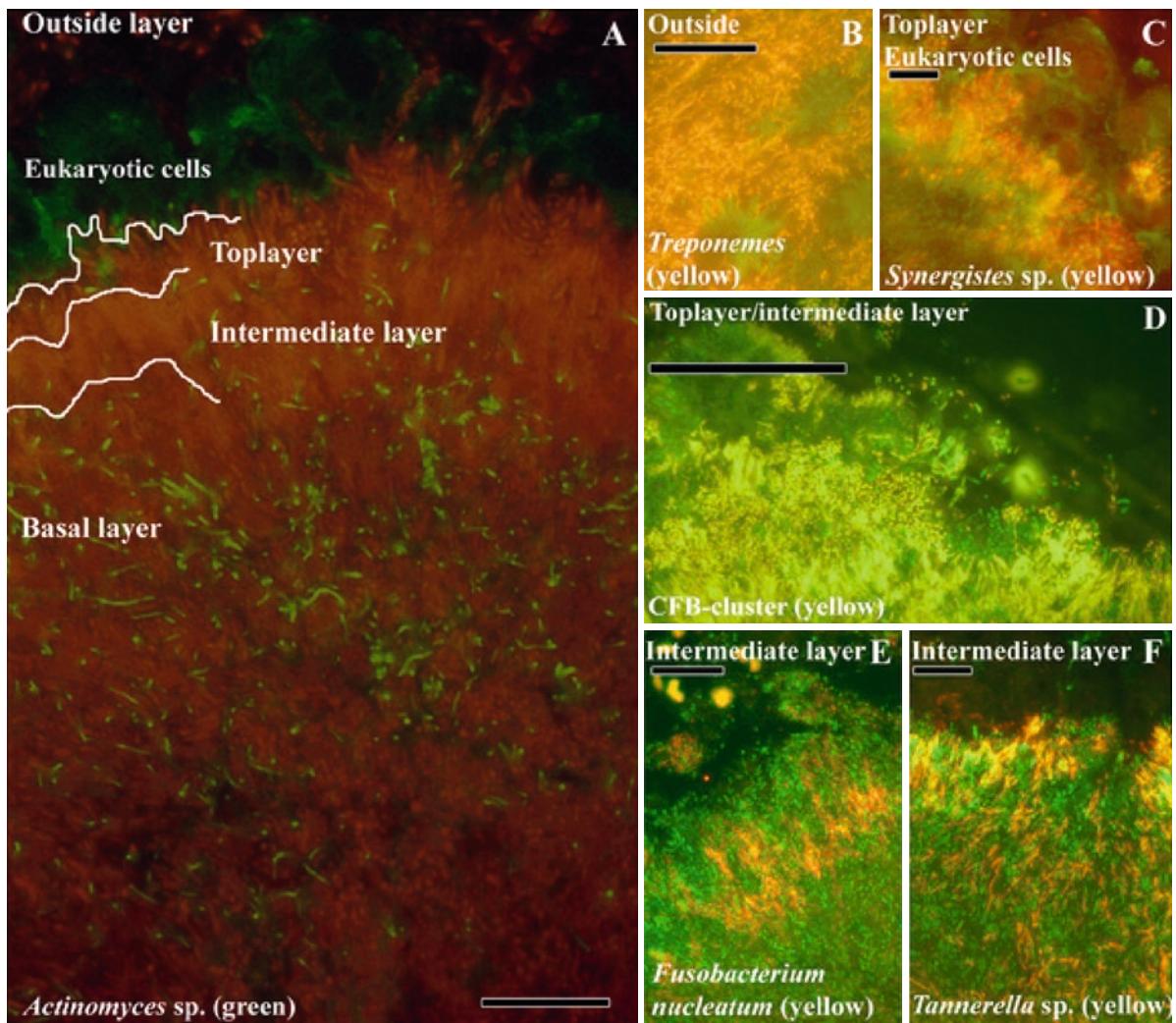
Supragingival biofilms are more heterogeneous in architecture compared to subgingival biofilms. In general, two different layers could be observed. The basal layer adheres to the tooth surface and four different biofilm types were observed. The first is a biofilm composed of only rod-shaped *Actinomyces* cells perpendicularly orientated to the tooth surface. The second type is a mixture of *Actinomyces* sp. and chains of cocci, not identified as streptococci, perpendicularly orientated to the tooth surface. The third type shows a biofilm with filamentous bacteria, streptococci, and yeasts, where streptococci form a distinct colony around yeast cells. The fourth type is a biofilm composed of mainly streptococci growing in close proximity to

*Lactobacillus* sp. that are orientated perpendicularly to the tooth surface [111] (Fig. 1.3).

### 1.1.3 Microbial Composition of Dental Plaque

The colonization pattern and the positive cooperation among **subgingival microbiota** were described by Socransky et al. [86], using DNA probes and checkerboard DNA–DNA hybridization analysis. They reveal that bacteria tend to be grouped in clusters according to nutritional and atmospheric requirements, all, that is, except *Actinomyces viscosus*, *Selenomonas noxia*, and *Aggregatibacter actinomycetemcomitans* (formerly *Actinobacillus actinomycetemcomitans*) serotype b, not belonging to any group [81]. The red cluster consisted of *Porphyromonas gingivalis*, *T. forsythia* (formerly *Bacteroides forsythus*), and *Treponema denticola*. The orange cluster consisted of *F. nucleatum* subsp., *Prevotella intermedia*, and *Prevotella nigrescens*, *Peptostreptococcus micros*, and *Campylobacter rectus*, *Campylobacter showae*, *Campylobacter gracilis*, *Eubacterium nodatum*, and *Streptococcus constellatus*. The three *Capnocytophaga* spp., *Campylobacter concisus*, *Eikenella corrodens*, and *A. actinomycetemcomitans* serotype a, formed the green cluster, while a group of streptococci made up the yellow cluster. *Streptococcus mitis*, *Streptococcus sanguinis*, and *Streptococcus oralis* were most closely related within this group. *Actinomyces odontolyticus* and *Veillonella parvula* formed the purple cluster. *Actinomyces naeslundii* genospecies 2 (formerly *A. viscosus*), *S. noxia*, and *A. actinomycetemcomitans* serotype b did not cluster with other species [20, 86] (Fig. 1.4).

The species within complexes are closely associated to one another: most periodontal sites harbor either all or none of the species belonging to the same complex, while individual species or pairs of species are detected less frequently than expected, reinforcing the hypothesis of the community theory rather than the germ theory. Precise interrelations are established between complexes as well. Microbiota belonging to the red cluster is very seldom detected in the absence of the orange complex, and the higher the detected amounts of orange complex bacteria, the greater is the colonization by red complex members. Yellow and



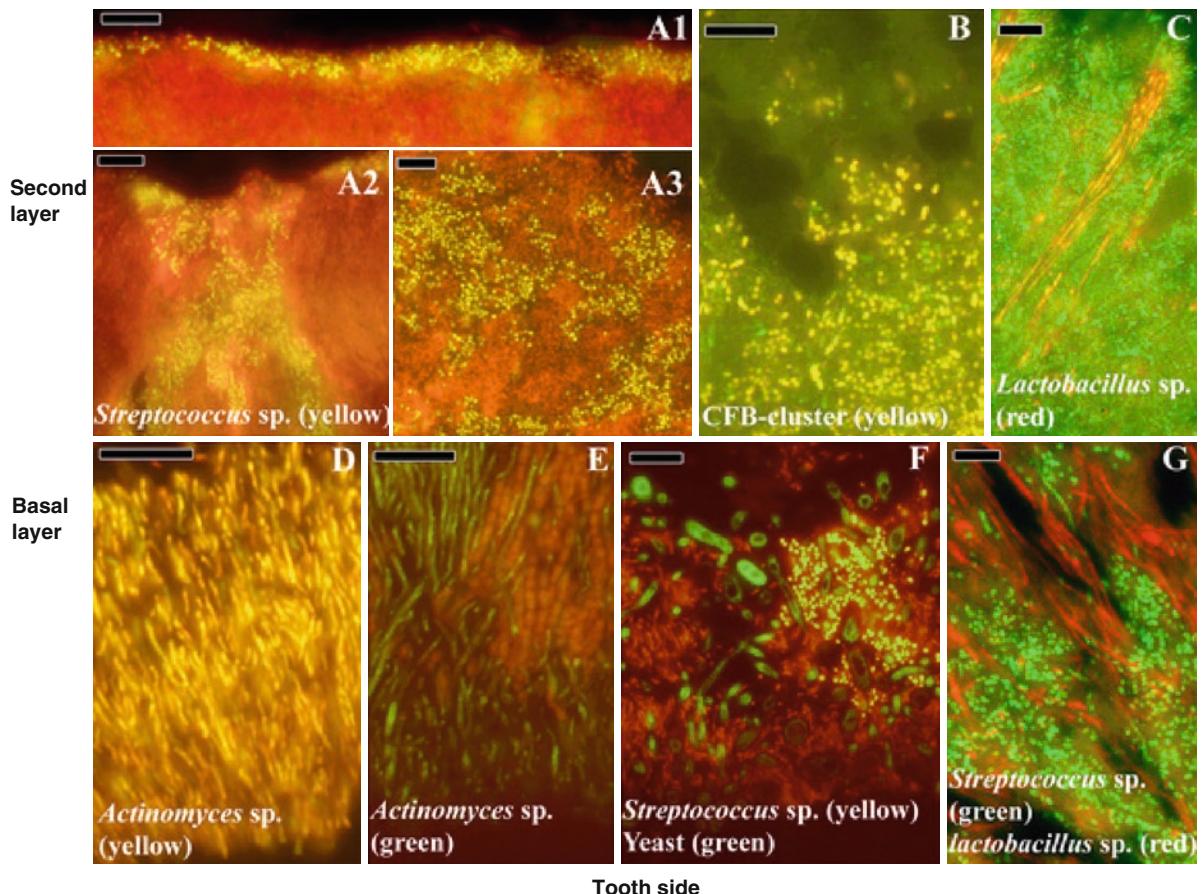
**Fig. 1.2** Localization of the most abundant species in subgingival biofilms. (a) Overview of the subgingival biofilm with *Actinomyces* sp. (green bacteria), bacteria (red), and eukaryotic cells (large green cells on top). (b) *Spirochaetes* (yellow) outside the biofilm. (c) Detail of *Synergistes* (yellow) in the top layer in close proximity to eukaryotic cells (green). (d) CFB cluster (yellow) in the

top and intermediate layers. (e) *Fusobacterium nucleatum* in the intermediate layer. (f) *Tannerella* sp. (yellow) in the intermediate layer. Each panel is double-stained with probe EUB338 labeled with FITC or Cy3. The yellow color results from the simultaneous staining with FITC- and Cy3-labeled probes. Bars are 10 µm.  
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green clusters show a similar preference for each other and a weaker relation with the orange and red complexes, while the purple complex shows loose relations with all the other clusters. Such relations can be explained by mechanisms of antagonism, synergism, and environmental selection [20, 81, 86].

Specific microbial complexes in **supragingival plaque**, similar to those found in subgingival plaque samples with a few minor differences, were recently described [28, 30]. A red complex community was

formed that contained the three species previously identified as the red complex in subgingival plaque, *T. forsythia*, *P. gingivalis*, and *T. denticola*. *E. nodatum* was also part of this complex and *Treponema socranskii* was loosely associated with these four species. A number of species previously identified in subgingival plaque as orange complex species were also detected as part of an orange complex in supragingival plaque. These included *C. showae*, *C. rectus*, *F. nucleatum* subsp. *nucleatum*, *F. nucleatum* subsp.



**Fig. 1.3** Localization of the most abundant species in supragingival biofilms. *Streptococcus* spp. (yellow) form a thin band on top of the biofilm (**a1**), almost engulfing in the biofilm (**a2**) or present as small cells scattered through the top layer of the biofilm (**a3**). (**b**) Cells from the CFB cluster of bacteria in the top layer of the biofilm, without defined structure. (**c**) *Lactobacillus* sp. (red) forming long strings through the top layer. (**d**) *Actinomyces* sp. (yellow) plaque attached to the tooth. (**e**) *Actinomyces* sp. (green)

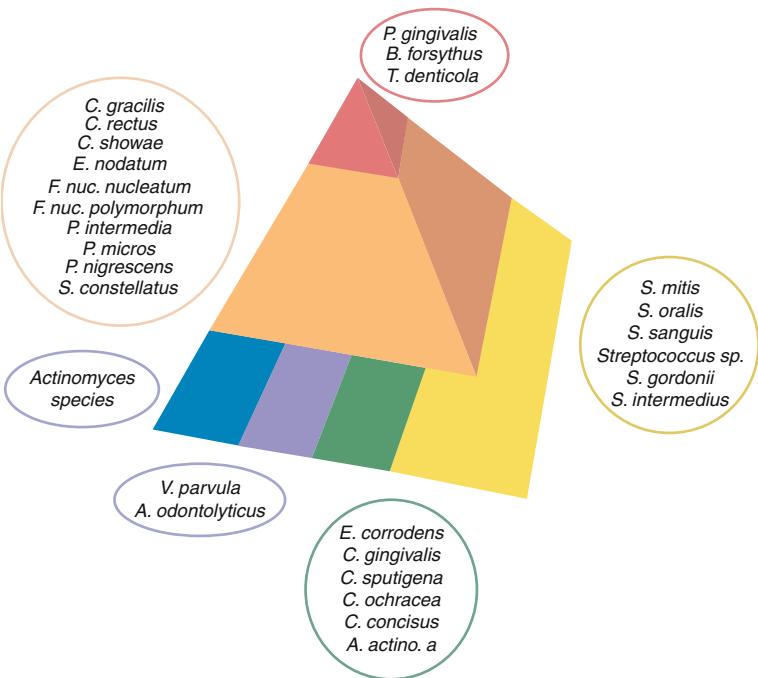
and cocci forming initial plaque. (**f**) Multispecies initial plaque composed of *Streptococcus* sp. (yellow), yeast cells (green), and unidentified bacteria (red). (**g**) *Streptococcus* sp. (green) and *Lactobacillus* sp. (red) forming initial plaque. Black holes might be channels through the biofilm. Panels **a**, **b**, **c**, **e**, **f** are double-stained with probe EUB338 labeled with FITC or Cy3. Bars are 10 µm. doi:10.1371/journal.pone.0009321.g003

*vincentii*, *Fusobacterium periodonticum*, *F. nucleatum* subsp. *polymorphum*, *C. gracilis*, *P. intermedia*, and *P. nigrescens*. These taxa were joined by *Gemella morbillorum*, *Capnocytophaga ochracea*, *S. noxia*, and *Prevotella melaninogenica*. A yellow complex was formed primarily of the *Streptococcus* spp. *S. mitis*, *S. oralis*, *Streptococcus gordonii*, *S. sanguinis*, and, somewhat separately, *Streptococcus anginosus*, *Streptococcus intermedius*, and *S. constellatus*. These species were joined by *Leptotrichia buccalis*, *Propionibacterium acnes*, *Eubacterium saburreum*, *Parvimonas micra* (formerly *Micromonas micra* and *P. micra*), and *A. actinomycetemcomitans*. A tight cluster of *Actinomyces* spp. was formed including *Actinomyces israelii*, *A. naeslundii* 1, *A. odontolyticus*,

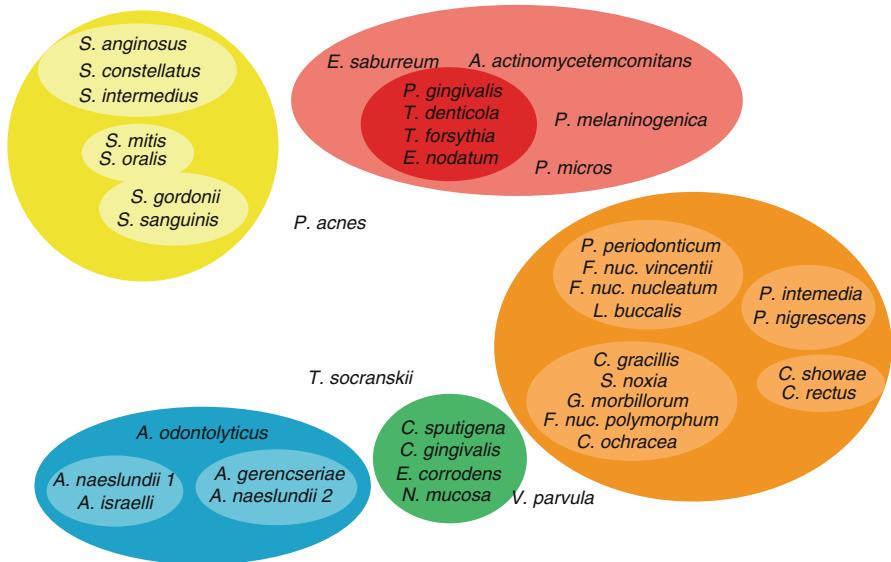
*Actinomyces gerencseriae*, and *A. naeslundii* 2. A green complex consisting of *Capnocytophaga sputigena*, *E. corrodens*, and *Capnocytophaga gingivalis* was formed as well as a loose purple complex consisting of *Neisseria mucosa* and *V. parvula* [28, 30]. It has been recently revealed that while plaque mass was associated with differences in proportions of many species in supragingival biofilms, tooth location also was strongly associated with species proportions [20, 28, 30] (Fig. 1.5).

Combined genomic and proteomic analyses of host–biofilm interactions are beginning to reveal the complex gene–protein interconnected networks present in periodontal health and disease. The concept is

**Fig. 1.4** Diagram of the association among subgingival species. The base of the pyramid is comprised of species thought to colonize the tooth surface and proliferate at an early stage. The orange complex becomes numerically more dominant later and is thought to bridge the early colonizers and the red complex species which become numerically more dominant at late stages in plaque development [87] (Reprinted with permission from John Wiley & Sons)



**Fig. 1.5** Diagrammatic representation of the relationships of species within microbial complexes and between the microbial complexes in supragingival biofilm samples [28] (Reprinted with permission from John Wiley & Sons)



emerging that different bacteria appear to be associated with clinically similar periodontal diseases in different people, and the oral microbiota associated with disease progression may be person-specific ([2] and references therein).

Healthy gingivae have been associated with a very simple supragingival plaque composition: few (1–20)

layers of predominantly gram-positive cocci (*Streptococcus* spp.: *Streptococcus mutans*, *S. mitis*, *S. sanguinis*, *S. oralis*; *Rothia dentocariosa*; *Staphylococcus epidermidis*), followed by some gram-positive rods and filaments (*Actinomyces* spp.: *A. viscosus*, *A. israelii*, *A. gerencseriae*; *Corynebacterium* spp.), and very few gram-negative cocci (*V. parvula*; *Neisseria* spp.). These

latter are aerobic or facultative aerobic bacteria able to adhere to the nonexfoliating hard surfaces; initial adhesion is promoted by surface free energy, roughness, and hydrophilicity, and is mediated by long- and short-range forces [20, 81].

Clinical gingivitis is associated with the development of a more organized dental plaque. Such biofilms are characterized by several cell layers (100–300), with bacteria stratification arranged by metabolism and aerotolerance; besides the gram-positive cocci, rods, and filaments associated with healthy gingivae, the number of gram-negative cocci, rods, and filaments increases and anaerobic bacteria appear (*F. nucleatum*, *C. gracilis*, *T. forsythia*, *Capnocytophaga* spp.). The species involved vary depending on local environmental characteristics, but the colonization pattern is always the same [81]. The severe forms of gingivitis have been associated with subgingival occurrence of the black-pigmented asaccharolytic *P. gingivalis* [103]. In pregnancy gingivitis an association between high levels of *P. intermedia* and elevations in systemic levels of estradiol and progesterone has been observed [47]. Microbial studies in acute necrotizing ulcerative gingivitis (ANUG) indicate high levels of *P. intermedia* and *Treponema pallidum*–related spirochetes (*spirochaete*, in coordination). Spirochetes are found to penetrate necrotic tissue as well as apparently unaffected connective tissue [20, 55, 73].

Comparisons of the microbiology of chronic and generalized aggressive forms of periodontitis are in the early phases. Both conditions appear to be associated with certain cultivable pathogens listed by the 1996 World Workshop in Periodontics, including *P. gingivalis*, *T. forsythia*, *C. rectus*, *Eubacterium* sp., *P. micra*, and *Treponema* sp. Application of culture-independent microbiological methods is beginning to reveal a longer and more diverse list of pathogens than was possible even a few years ago. It is clear that chronic and generalized aggressive periodontitis are not simply gram-negative anaerobic infections, but that gram-positive bacteria and even nonbacterial microbes from the Archaea domain may have an etiological role. Preliminary studies have suggested that individuals with generalized aggressive periodontitis have higher subgingival levels of *Selenomonas* sp. and *T. lecithinolyticum* compared to patients with chronic periodontitis ([2] and references therein).

Shibli et al. [83] compared the supra- and subgingival microbial composition of subjects with and

without peri-implantitis. The microbial profile between supra- and subgingival environments did not differ substantially, especially in the healthy group. Three host-compatible bacterial species (*A. naeslundii* 1, *S. intermedius*, and *S. mitis*) and one putative periodontal pathogen (*F. periodonticum*) were present at higher levels in the supragingival samples compared with the subgingival samples of the healthy implants. The levels of three beneficial microorganisms, *V. parvula*, *S. gordonii*, and *S. intermedius*, as well as *F. periodonticum*, were significantly increased in the supragingival biofilm compared with the subgingival biofilm of the diseased implants. There was a trend toward higher mean counts of some pathogens such as *F. nucleatum* subsp. *nucleatum*, *P. intermedia*, *P. nigrescens*, *T. denticola*, *S. noxia*, and *T. forsythia* in the subgingival biofilm of the peri-implantitis group.

#### 1.1.4 Importance of Biofilms in Human Diseases

Bacteria-forming biofilms are an important cause of persistent infection. It is clear from epidemiological data that biofilms play a role in certain diseases, particularly cystic fibrosis, periodontitis, bloodstream, and urinary tract infections as a result of indwelling medical devices, and it is particularly important in immunocompromised patients [78].

The main pathogenic mechanisms include [78]:

1. The detachment of cells or cell aggregates, including even only a very small number of cells, from a biofilm on an indwelling medical device, is capable of producing a bloodstream or urinary tract infection. Microorganisms detaching from a biofilm on a medical device or another infection can easily escape from the immune system and cause infection.
2. Endotoxin production in gram-negative bacteria in the biofilm of an indwelling medical device may activate an immune response from the patient.
3. The last mechanism is provision of a niche to generate resistant organisms. Bacteria can exchange plasmids, including resistance genes, by conjugation within the biofilm [78].

## 1.2 Mechanisms of Biofilm Resistance Against Antimicrobials

Growth as a biofilm almost always leads to a large increase in resistance to antimicrobial agents compared with cultures grown in suspension (planktonic) in conventional liquid media, with up to 1,000-fold decreases in susceptibility reported [10, 25, 78, 85].

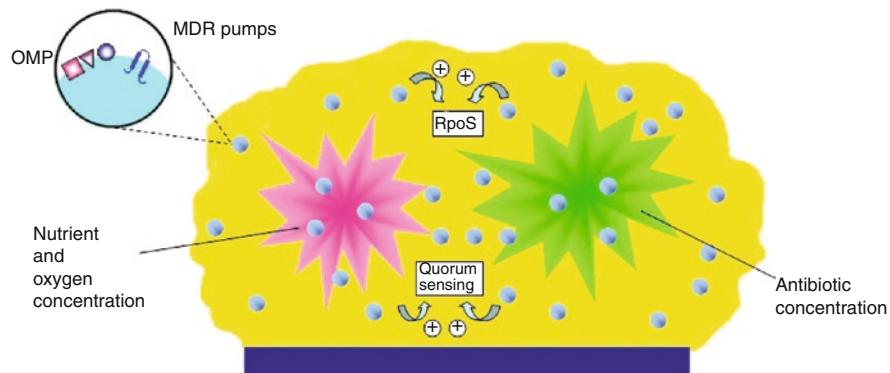
Mechanisms responsible for a high level of resistance in biofilms are due to one or more of the following (Fig. 1.6):

### 1.2.1 Biofilm Impermeability to Antimicrobial Agents

Antimicrobial molecules must reach their target in order to inactivate the enmeshed bacteria. The biofilm glycocalyx protects infecting cells from humoral and cellular host defence systems as well as the diffusion of antimicrobials to cellular targets, acting as a barrier by influencing the rate of transport of molecules into the biofilm interior [78]. More comprehensive reviews in this subject were performed by Lewis et al. [54], Mah and O'Toole [58], and Xu et al. [110].

Communication among the different species within biofilms appears to be the key to understanding how plaque can act as a single unit, and how specific bacteria emerge and impair the balance with the host. **Physical (coaggregation and coadhesion), metabolic, and physiological (gene expression and cell-cell signaling) interactions** yield a positive cooperation among different species within the biofilm: the metabolic products of some organisms may promote the further growth of other bacteria or prevent their survival. A key role in the cooperative processes is played by *F. nucleatum*, able to form the needed “bridge” between early, i.e., *Streptococci* spp., and late colonizers, especially obligate anaerobes. In the absence of *F. nucleatum*, *P. gingivalis* cannot aggregate with the microbiota already present such as the facultative aerobes *A. naeslundii*, *Neisseria subflava*, *S. mutans*, *S. oralis*, and *S. sanguinis* (formerly *S. sanguis*). The presence of *F. nucleatum*, on the other hand, enables anaerobes to grow, even in the aerated environment of the oral cavity. Other microorganisms are also able to link otherwise noncommunicating bacteria (i.e., *S. sanguinis* forms a “corn cob” complex together with *Corynebacterium matruchotii* (formerly *Bacterionema matruchotii*) and *F. nucleatum*), and this may represent the basic event leading to biofilm initiation and development [81].

Sedlacek and Walker [82] utilized an in vitro biofilm model of subgingival plaque to investigate



**Fig. 1.6** Drug resistance in biofilms. A schematic of mechanisms that can contribute to the resistance of biofilm-grown bacteria to antimicrobial agents. The extracellular polysaccharide is represented in yellow and the bacteria as blue ovals. Biofilms are marked by their heterogeneity and this heterogeneity can include gradients of nutrients, waste products and oxygen (illustrated by colored starbursts). Mechanisms of resistance in the biofilm include increased cell density and physical exclusion of the antibiotic. The individual bacteria in a biofilm can also undergo

physiological changes that improve resistance to biocides. Various authors have speculated that the following changes can occur in biofilm-grown bacteria: (1) induction of the general stress response (an *rpoS*-dependent process in Gram-negative bacteria); (2) increasing expression of multiple drug resistance (MDR) pumps; (3) activating quorum-sensing systems; and (4) changing profiles of outer membrane proteins (OMP) [58] (Reprinted with permission from Elsevier)

resistances in subgingival biofilm communities to antibiotics commonly used as adjuncts to periodontal therapy. Biofilms were grown on saliva-coated hydroxyapatite supports in trypticase–soy broth for 4 h to 10 days and then exposed for 48 h to either increasing twofold concentrations of tetracycline, amoxicillin, clindamycin, and erythromycin or therapeutically achievable concentrations of tetracycline, doxycycline, minocycline, amoxicillin, metronidazole, amoxicillin/clavulanate, and amoxicillin/metronidazole. The authors indicated that concentrations necessary to inhibit bacterial strains in steady-state biofilms were up to 250 times greater than the concentrations needed to inhibit the same strains grown planktonically. Resistance appeared to be age-related because biofilms demonstrated progressive antibiotic resistance as they matured with maximum resistance coinciding with the steady-state phase of biofilm growth.

Because subgingival bacteria are organized in biofilms, in principle, they are less susceptible to antimicrobials. The oral plaque biofilm needs to be mechanically removed or disturbed in order for antimicrobials to be effective. To date, the only predictable way to disturb the dental biofilm is by using mechanical means [37, 100].

### 1.2.2 Altered Growth Rates in Biofilm Organisms

An alternative proposed mechanism for resistance of biofilm-associated cells (sessile organisms) to antimicrobials is that the growth rate of these organisms is significantly slower than the growth of planktonic (biofilm free) cells; therefore, the uptake of the antimicrobial molecules is diminished [1, 18, 19, 101].

### 1.2.3 The Biofilm Microenvironment Antagonizing Antimicrobial Activity

In the biofilm microenvironment, various factors can affect activity of antimicrobial agents in vitro, including  $pO_2$ ,  $pCO_2$ , divalent cation concentration, hydration level, pH, or pyrimidine concentration, producing

adverse effects for antimicrobial action deep in the bacterial biofilm where acidic and anaerobic conditions persist [78]. There is also evidence that relatively large amounts of antibiotic-inactivating enzymes such as  $\beta$ -lactamases which accumulate within the glycocalyx produce concentration gradients that can protect underlying cells [85].

Handal et al. [34] assessed the extent of  $\beta$ -lactamase-producing bacteria in subgingival plaque samples obtained from 25 patients with refractory marginal periodontitis in the USA.  $\beta$ -lactamase-producing bacteria were detected in 72% patients. The most prominent  $\beta$ -lactamase-producing organisms belonged to the anaerobic genus *Prevotella*. Other enzyme-producing anaerobic strains were *F. nucleatum*, *P. acnes*, and *Peptostreptococcus* sp. Facultative bacteria, such as *Burkholderia* spp., *Ralstonia pickettii*, *Capnocytophaga* spp., *Bacillus* spp., *Staphylococcus* spp., and *Neisseria* sp., were also detected among the enzyme producers [34].

Antimicrobial resistance and  $\beta$ -lactamase production of clinical isolates of *A. actinomycetemcomitans*, *P. gingivalis*, *F. nucleatum*, *E. corrodens*, and *Prevotella* spp. were studied [5, 7, 23, 34, 35, 46, 51, 56, 57, 62, 67, 99].

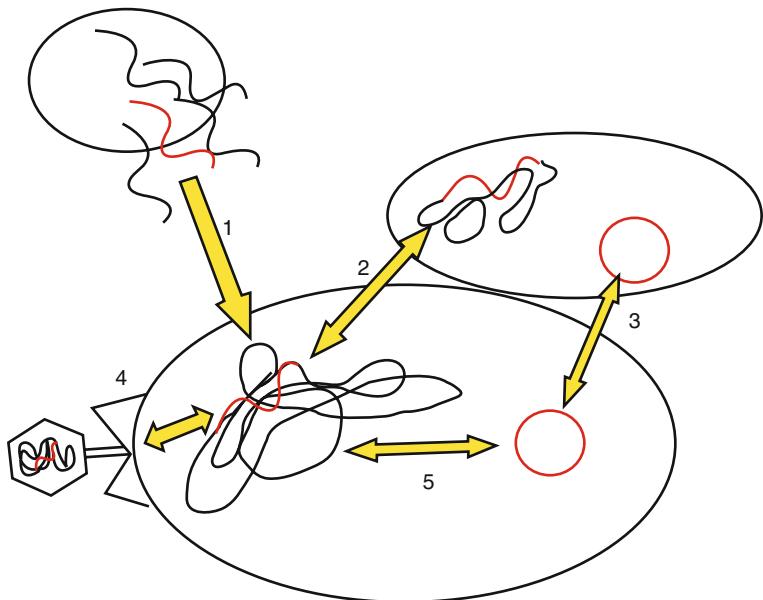
To overcome  $\beta$ -lactamase-mediated resistance, a combination of  $\beta$ -lactam and a  $\beta$ -lactamase inhibitor, which protects the  $\beta$ -lactam antibiotic from the activity of the  $\beta$ -lactamase, has been widely used in the treatment of human infections. Although there are some very successful combinations of  $\beta$ -lactams and  $\beta$ -lactamase inhibitors, most of the inhibitors act against class A  $\beta$ -lactamases and remain ineffective against class B, C, and D  $\beta$ -lactamases [68].

### 1.2.4 The Role of Horizontal Dissemination in the Biofilm

Horizontal gene transfer among bacteria is recognized as a major contributor in the molecular evolution of many bacterial genomes. In addition, it is responsible for the seemingly uncontrollable spread of antibiotic-resistance genes among bacteria in the natural and nosocomial environments [75].

Horizontal transmission of genetic information between bacteria can occur by three gene transfer

**Fig. 1.7** Transfer of genetic information between bacterial cells. Transformation (1), conjugation of a conjugative transposon (2) and a conjugative plasmid (3), and transduction (4) are shown. Also shown is the integration of a plasmid into the chromosome (5). The mobile DNA is shown in red and the chromosomal DNA is shown as a thin red line. Cell membranes are thick black lines [75] (Reprinted with permission from John Wiley & Sons)



mechanisms: conjugation, transduction, and transformation (Fig. 1.7). The mechanisms are distinct, and all have specific physical and biological requirements. **Conjugation** requires that the donor cell have a conjugative element, usually a plasmid or a transposon, and that physical contact be made between donor and recipient cells to initiate transfer of the DNA molecule. Both cells must also be metabolically active to initiate the process. Genetic exchange via **transduction** also requires that the participant cells be metabolically stable. This process involves transducing bacteriophage particles that harbor the foreign DNA. In this case, the host and donor can be physically separated, since the phages are able to exist independently of the bacteria for the time span necessary for gene transfer to occur. The third mechanism, gene transfer by **transformation**, does not require a living donor cell, since free DNA released during cell death and lysis is the principal source of the donor DNA. The persistence and dissemination of the DNA are the major factors influencing the maximal time and distance that DNA and host cells can be separated. The recipient cell must, however, be metabolically active to transport and incorporate the foreign DNA [14].

The three fundamental mechanisms of horizontal gene transfer all play a part in the dissemination of genetic information throughout the oral cavity, and these three processes include plasmids, conjugative

transposons (CTn), and bacteriophages, which have been demonstrated to be mobile [75].

#### 1.2.4.1 Plasmids

Plasmids usually exist as independently replicating units; however, on some occasions they will integrate into the bacterial chromosome. Plasmids are grouped into incompatibility (Inc) groups, based on their inability to coexist in the same cell. Plasmids from the same Inc group usually have identical or similar replication/partition systems. Only one plasmid from one Inc group can exist in a cell at one time; if another plasmid belonging to the same Inc group enters the cell, one plasmid will eventually be lost during cell division owing to mutual interference of the replication process by the other plasmid, leading to an unequal amount of the two plasmids in the dividing cell [75].

Plasmids are common in both gram-positive and gram-negative organisms isolated from the oral cavity. Among the most important plasmids in mediating broad host-range gene transfer are those of the **IncP group** (usually found in gram-negative bacteria), as they are the most stable low-copy-number plasmids known to date [75].

The first example of conjugal transfer of DNA from *Escherichia coli* to the periodontal pathogen

*A. actinomycetemcomitans* was presented by Goncharoff et al. [26]. The 60-kb **IncP plasmid RK2** confers resistance to ampicillin (Ap), kanamycin (Kin), and tetracycline (Tc), is self-transmissible to a wide variety of bacteria, and replicates in diverse gram-negative bacterial hosts. Plasmids pRK2525 and pRK212.2 are both Tc<sup>r</sup> derivatives of RK2. Both plasmids contain an insertion at the *Sall* site located within the Tc<sup>r</sup> determinant at approximately kb 14 on the RK2 map. Derivatives of the incompatibility group P (IncP) plasmid RK2 successfully transferred from an *E. coli* donor to an *A. actinomycetemcomitans* recipient. The resulting *A. actinomycetemcomitans* transconjugants transferred the plasmids back to *E. coli* recipients. The IncP transfer functions were also used in *trans* to mobilize the IncQ plasmid pBKI from *E. coli* to *A. actinomycetemcomitans*. The **IncP and IncQ plasmids** both transferred into *A. actinomycetemcomitans* at high frequencies (0.3–0.5 transconjugants per donor) and showed no gross deletions, insertions, or rearrangements. Determinations of minimum inhibitory concentrations (MICs) of various antibiotics for the *A. actinomycetemcomitans* transconjugant strains demonstrated the expression of ampicillin, chloramphenicol, and kanamycin resistance determinants ([26] and the references therein).

A specific cell wall antigen of high molecular weight which appears to be unique to virulent strains of *A. viscosus* and *A. naeslundii* is composed of two parts: a polysaccharide moiety containing 6-DOT as the major sugar and determinant of serologic specificity, and a small peptide bearing some resemblance to the peptidoglycan. A positive correlation between the presence of this antigen and an extrachromosomal piece of DNA having most of the properties of a bacterial plasmid was revealed [32].

When antibiotic susceptibility of *A. actinomycetemcomitans* isolated from periodontally diseased sites was tested, 82% of the *A. actinomycetemcomitans* isolates hybridized with the *tetB* determinant. The *tetB* determinant was transferable between *A. actinomycetemcomitans* isolates and a *Haemophilus influenzae* recipient, and appears to be associated with conjugative plasmids [79].

It has been also shown that oral streptococci might exchange genetic information in the oral cavity. Chromosomal and plasmid-borne antibiotic resistance markers could be readily transferred from *S. mutans* GS-5 to *S. milleri* NCTC 10707 or *S. sanguis* Challis during mixed growth [49].

*F. nucleatum* is a gram-negative anaerobic rod found in dental plaque biofilms, and is an opportunistic pathogen implicated in periodontitis as well as a wide range of systemic abscesses and infections. Genomic analyses of *F. nucleatum* indicate considerable genetic diversity and a prominent role for horizontal gene transfer in the evolution of the species. Several plasmids isolated from *F. nucleatum*, including pFN1, harbor relaxase gene homologs that may function in plasmid mobilization [9].

#### 1.2.4.2 Conjugative Transposons

Transposons are borne both by plasmids and the chromosome and have an enormous variation in their genetic organization, the genes responsible for their insertion and excision, and in the accessory or passenger genes they carry. Transposable elements are also able to interact, by recombination between elements and/or by transposition into other elements, forming novel chimeric elements [74].

Warburton et al. [102] demonstrated the transfer of antimicrobial resistance (doxycycline) carried on a conjugative transposon between oral bacteria during systemic antimicrobial treatment of periodontitis in humans. Streptococci were cultured before and after treatment from the subgingival plaque of two patients with periodontitis, genotyped and investigated for the presence of antimicrobial resistance determinants and conjugative transposons. In one subject, a strain of *S. sanguinis* resistant to doxycycline was a minor component of the pretreatment streptococcal flora but dominated post treatment. In a second subject, a strain of *Streptococcus cristatus*, which was sensitive to doxycycline before treatment, was found to have acquired a novel conjugative transposon during treatment, rendering it resistant to doxycycline and erythromycin. The novel transposon, named **CTn6002**, was sequenced and found to be a complex element derived in part from Tn916, and an unknown element which included the erythromycin resistance gene *erm(B)*. A strain of *S. oralis* isolated from this subject pretreatment was found to harbor CTn6002 and was therefore implicated as the donor [102].

Tetracycline-resistant streptococci are frequently isolated from the oral cavity of humans, and resistance is most commonly conferred by *tetM*, a ribosomal

protection protein often associated with the conjugative transposon (cTn) Tn916. **Tn916** belongs to a family of cTns that are composed of functional modules involved in conjugation, antibiotic resistance, regulation, and integration and excision. The finding of *tetS* in the same relative position as *tetM* in a broad-host-range Tn916-related element supports the view that conjugative transposons are composed of modules that are able to exchange with modules from other elements, possibly by homologous recombination. It now seems apparent that not only is Tn916 involved in the dissemination of *tetM*, but it is also involved in the dissemination of *tetS* [52, 72].

The presence of erythromycin resistance genes in oral streptococci is important because viridans group streptococci have been shown to cause systemic diseases and they can disseminate the erythromycin resistance genes to other more pathogenic bacteria, such as *Streptococcus pneumoniae*. Villedieu et al. [98] identified 12 isolates that, as well as being resistant to erythromycin, were also resistant to tetracycline. The filter-mating study showed that 4 of 12 isolates were able to transfer genes encoding resistance to both erythromycin [*ermB*] and tetracycline [*tetM*] to an *E. faecalis* recipient. These two genes have previously been found on the same conjugative transposon, Tn1545, which belongs to a larger class of conjugative transposons that includes the well-studied element Tn916. It was revealed that there is variation in the restriction pattern of the Tn1545-like elements and that these elements are widespread in the oral cavity and, more particularly, in oral streptococci. Moreover, it was demonstrated that these elements are capable of intergenic transfer [98].

Intergeneric transfer of a conjugative transposon in a mixed-species biofilm demonstrating the ability of conjugative transposons to disseminate antibiotic resistance genes in a mixed-species environment was reported by Roberts et al. [76]. Tn5397 is a conjugative transposon originally isolated from *Clostridium difficile* strain 630, which confers tetracycline resistance (Tcr) upon its host via the *tetM* gene. Tn5397 can be transferred to, and from, *Bacillus subtilis* and *C. difficile*. Tn5397 is closely related to the well-studied conjugative transposon Tn916 in the regions concerned with conjugation [76].

The complete genome sequence of *S. mutans* enables a better understanding of the complexity and genetic specificity of this organism. Strain **UA159**

contains one probable conjugative transposon (**TnSmu1**) that is related to but distinct from the well-known Tn916 from *Enterococcus faecalis*. Prominent in the genome is a large transposon-like region (TnSmu2) containing the genes similar to gramicidin and bacitracin synthetases; these genes include some of the largest open reading frames (ORFs) found in the genome (1 over 8 kb). This large region, over 40 kb, is flanked by remnants of transposases of the ISL3 family, arranged in inverted orientation relative to each other, whose reading frames are disrupted by frameshifts, and contain several gene fragments from other insertion sequence (IS) element families. The nucleotide composition of this region markedly diverges from the genome average, having a %G+C of 28.9%. Such deviations may point to foreign origins of these elements. Although the transposons, IS elements, and fragments are distributed over the entire genome, there are several regions containing clusters of IS elements or remnants, suggesting that these regions may represent “hot spots” for integration [102].

The complete 2,343,479-bp genome sequence of the gram-negative, pathogenic oral bacterium *P. gingivalis* strain W83, a major contributor to periodontal disease, was also determined. The transposable elements include IS elements and miniature inverted-repeat transposable elements (MITEs) and large stretches of genes that resemble remnants of conjugable and mobilizable transposons based on sequence similarity to elements previously described in *Bacteroides* spp. Although there are 96 complete or partial copies of IS elements and MITEs present in strain W83 that occupy more than 94 kb of the genome, the transposable elements are rarely found in a functional gene. Instead, these elements have inserted almost exclusively into intergenic regions and other copies of transposable elements, except for one insertion into a putative outer membrane protein (PG0176/PG0178) that is intact in at least four other strains of *P. gingivalis* (accession numbers AB069977 to AB069980) [63].

The gram-negative oral bacterium *A. actinomycetemcomitans* has been implicated as a causative agent of several forms of periodontal diseases. The conjugative tetracycline resistance transposon Tn916 was transduced to *A. actinomycetemcomitans* recipients as a unit. Transfer by transformation or conjugation was experimentally excluded. Tn916 integrated at different sites within the *A. actinomycetemcomitans* genome,

suggesting an integration by transposition rather than by homologous recombination of flanking sequences [105, 106].

*E. corrodens* is found in dental plaque and periodontal lesions and is implicated in the initiation and progression of certain destructive periodontal disease syndromes. Although most *E. corrodens* strains are susceptible to  $\beta$ -lactam antibiotics, some are resistant. A sequence similar to a portion of transposon Tn3 has been identified in DNA from *E. corrodens* EC-38 [51].

#### 1.2.4.3 Bacteriophage

Bacteriophage can contribute to horizontal gene transfer by transduction (a process in which bacterial DNA becomes erroneously packaged into phage heads) or lysogenic conversion (where the phage genome enters the bacterial genome and results in a phenotypic change, depending on which determinants are present on the bacteriophage genome or which host genes are inactivated as a result of insertion of the phage genome into the bacterial DNA). Bacteriophages are responsible for the lysogenic conversion of many different nonpathogenic bacteria (including *E. coli*, *Vibrio cholerae*, *Listeria* spp., and *Streptococcus* spp.) to pathogens [75].

There are a limited number of reports in the literature on the isolation of bacteriophages from the oral cavity [40]. A range of bacteriophages specific for species of *Veillonella* spp. [39], *Actinomyces* spp. [16, 93], *S. mutans* [17], *Enterococcus faecalis* [4], and *A. actinomycetemcomitans* [36, 43, 69, 80, 88, 105–107] have been described in dental plaque samples or in saliva.

#### 1.2.5 Communications Systems (Quorum Sensing)

The regulation of bacterial gene expression in response to changes in cell density is known as quorum sensing. Quorum-sensing bacteria synthesize and secrete extracellular signaling molecules called autoinducers, which accumulate in the environment as the population increases [65]. Gram-positive bacteria generally communicate via small diffusible peptides, while many gram-negative bacteria secrete acyl homoserine lactones (AHLs) [104], the structure of which varies

depending on the species of bacteria that produce them. AHLs are involved in quorum sensing whereby cells are able to modulate gene expression in response to increases in cell density. Another system involves the synthesis of autoinducer-2 (AI-2), the structure of which is unknown, but a gene product, LuxS, is required [21, 108]. This system may be involved in cross-communication among both gram-positive and gram-negative bacteria, as homologues of LuxS are widespread within the microbial world [60]. Several strains of *P. intermedia*, *F. nucleatum*, and *P. gingivalis* (formerly *Bacteroides gingivalis*) were found to produce such activities [24, 109]. It was also revealed that the signals produced by subgingival bacteria induce both intra- and interspecies responses in the mixed-species microbial communities that exist in the oral cavity [65].

#### 1.2.6 Antibiotic Susceptibility of Bacterial Species Residing in Biofilms

Although it has been shown that bacterial species residing in biofilms are much more resistant to antibiotics than the same species in a planktonic state, antibiotics have been used frequently in the treatment of periodontal infections [91]. van Winkelhoff et al. [97] and Slots and Ting [84] revealed that systemically administered antibiotics provided a clear clinical benefit in terms of mean periodontal attachment level “gain” post therapy when compared with groups not receiving these agents. Meta-analyses performed by Herrera et al. [38] and Haffajee et al. [27] indicated that adjunctive systemically administered antibiotics can provide a clinical benefit in the treatment of periodontal infections. However, it must be pointed out that not every study found that adjunctive systemically administered antibiotics provided a benefit to the subject in terms of clinical or microbial outcomes beyond control mechanical debridement therapies [91].

The emergence of resistant pathogens is of concern not only in medicine, but also in dentistry as it may be one reason for treatment failure [48]. Several studies have evaluated the antibacterial susceptibility and resistance development of dental plaque bacteria [8, 22, 33, 34, 41, 50, 61, 71, 77].

The level of resistance varies between countries, which can be attributed to the different use of antibiotics [48]. It was demonstrated that bacterial resistance in subgingival plaque samples taken from adult periodontitis patients against a number of common antibiotics was higher in Spain than in the Netherlands. A higher level of resistance in Spain was found for penicillin, amoxicillin, metronidazole, clindamycin, and tetracycline [94, 96]. Also several Spanish bacterial species isolated from periodontal lesions demonstrated higher MIC values when compared with Dutch isolates. Differences were observed for the  $\beta$ -lactam antibiotics, such as penicillin and amoxicillin, against *P. intermedia*, *F. nucleatum*, and *A. actinomycetemcomitans* strains [95]. Kulik et al. [48] evaluated the resistance profiles of *A. (Actinobacillus) actinomycetemcomitans*, *P. gingivalis*, and *P. intermedia/P. nigrescens* to detect possible changes in antibiotic resistance over the time period of 1991–2005 in Switzerland, a country with the lowest antibiotic consumption among European countries. No antibiotic resistance was detected in *P. gingivalis*, whereas a few isolates of *P. intermedia* were not susceptible to clindamycin (0.9%), phenoxy-methylpenicillin (13.5%), or tetracycline (12.6%). Amoxicillin/clavulanic acid, tetracycline, and metronidazole were the most effective antibiotics against *A. actinomycetemcomitans* with 0%, 0.8%, and 20.8% nonsusceptible isolates, respectively. However, 88% of the *A. actinomycetemcomitans* isolates were nonsusceptible to phenoxy-methylpenicillin and 88% to clindamycin. When strains isolated in the years 1991–1994 were compared with those isolated in the years 2001–2004, there was no statistically significant difference in the percentage of *A. actinomycetemcomitans* strains nonsusceptible to clindamycin, metronidazole, or phenoxy-methylpenicillin, or in the percentage of *P. intermedia* strains nonsusceptible to phenoxy-methylpenicillin or tetracycline ( $P>0.4$  each).

The tetracyclines, metronidazole, and  $\beta$ -lactams are among the most widely used agents for treating periodontal conditions. Mechanisms of bacterial resistance to these antibiotics have been extensively described and attributed to resistance genes [44]. Many genes for bacterial resistance to tetracycline have been identified and characterized. These include *tet A, B, C, D, E, G, H, I, K, L, M, O, Q*, and *X* associated with gram-negative bacteria, and *tet K, L, M, O, P, Q, S, Otr A, B, and C* (oxytetracycline resistance determinants) associated with gram-positive bacteria.

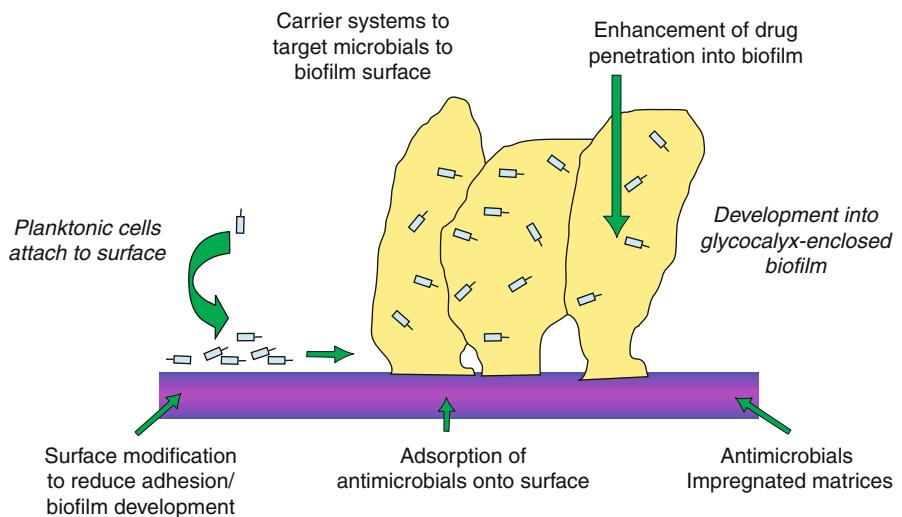
Various tetracycline resistance determinants have been associated with putative periodontal pathogenic bacteria. For example, *Bacteroides* has been shown to exhibit *tetM*, *tetQ*, and *tetX*; *Veillonella* is often associated with *tetM* and *tetQ*; and *Streptococcus* and *Peptostreptococcus* are reported to express *tetO* and *tetK*. Other *tet*-resistant genes and their association with the genera of purported periodontal pathogens include: *tetB* and *Treponema*; *tetM* and *Eikenella*, *Fusobacterium*, and *Prevotella*; *tetO* and *Campylobacter*; and *tetQ*, which has been associated with the genera *Porphyromonas* and *Capnocytophaga* ([59] and the references therein). The genetic determinants of bacterial resistance to metronidazole have not been extensively investigated in the oral environment. Nitroimidazole resistance genes *nim* (A–D) carrying this property are found in plasmids or the chromosome and their proposed mechanism of action is the encoding of a reductase that cannot convert the pro-drug into active nitroimidazoles, thus preventing the formation of toxic radicals required for antimicrobial activity [31, 44, 89].

### 1.2.7 Role for Drug Delivery Systems for Combating Bacterial Resistance of Biofilm

A number of the key elements in the infectious process and formation of biofilm have been proposed for application of novel technologies and drug delivery systems – prevention of colonization and biofilm formation, accumulation at the biofilm surface, and drug penetration into the biofilm (Fig. 1.8). Given the increasing use of relatively invasive medical and surgical procedures, the material properties of medical devices have received much attention as have strategies to target antimicrobials to device-related infections. Dental plaque and oral hygiene is another common therapeutic target and there is now a considerable body of work using carrier systems to target antibiotics against intracellular infections [85].

As ordinary antiseptics are difficult to maintain at therapeutic concentrations in the oral cavity and can be rendered ineffective by resistance development in target organisms, a unique alternative antimicrobial approach was described [70]. A novel approach, **photodynamic therapy**, could be a solution to these problems. Lethal

**Fig. 1.8** Anti-biofilm strategies [85] (Reprinted with permission from Elsevier)



photosensitization of many bacteria, both gram-positive and gram-negative, was found in many studies. The advantage of this new approach includes rapid bacterial elimination, minimal chance of resistance development, and safety of adjacent host tissue and normal microflora. However, in a recent meta-analysis, Azarpazhooh et al. [3] showed that photodynamic therapy as an independent treatment or as an adjunct to scaling and root planning (SRP) versus a control group of SRP did not demonstrate statistically or clinically significant advantages. Combined therapy of photodynamic therapy + SRP indicated a probable efficacy in CAL gain (MD: 0.34 mm; 95% confidence interval: 0.05–0.63 mm) or probing depth reduction (MD: 0.25 mm; 95% confidence interval: 0.04–0.45 mm). As a conclusion, the routine use of photodynamic therapy for clinical management of periodontitis was not recommended.

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## The Prophylactic Use of Antibiotics in Periodontal Therapy

“Antibiotic prophylaxis” is defined in the MeSH (Index Medicus: Medical Subject Headings) browser as “use of antibiotics before, during, or after a diagnostic, therapeutic, or surgical procedure to prevent infectious complications” (<http://www.ncbi.nlm.nih.gov> [45]).

“Dental procedure” is defined as a procedure performed by an oral health caregiver, for example, dental assistant, dental hygienist, or dentist. Toothbrushing was not included in this definition [45].

It should also be noted that there are several important issues related to antibiotic prophylaxis:

1. The range of potential side effects from the administration of antibiotics is vast, largely with a hypersensitive etiology, but some direct toxic effects may also occur. All four types of hypersensitivity reaction have been reported with the use of penicillins including the most severe reaction, anaphylactic shock, and other type I reactions including allergic bronchial obstruction, allergic rhinitis, and angio-edema; hemolytic anemia, type II, has been recorded; drug fever, a type III reaction; and the delayed type hypersensitivity (type IV) of allergic dermatitis summarized in *Meyler's side effects of drugs* [11, 119, 145].
2. The development of resistant organisms is also a real problem. Recent indiscriminate use of antibiotics has resulted in an increase in the prevalence of penicillin-resistant *streptococcus viridans* in blood cultures. Overall, however, the contribution of inappropriate dental prescription is significantly less than the medical contribution and, in turn, this is vastly less than issues relating to the use of antibiotics in agricultural animals [145]. In the 1990s, we were introduced to health-care-acquired methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-resistant *Enterococcus*, and drug-resistant

*Pseudomonas aeruginosa* in hospitals and a global threat of penicillin-resistant *Streptococcus pneumoniae*. We are now seeing additional drug-resistant organisms circulating outside health care institutions, such as community-acquired MRSA (a dramatically virulent organism), coliforms producing extended-spectrum β-lactamases (enzymes that render them resistant to all penicillins and cephalosporins), and a new aggressive strain of *Clostridium difficile* [112].

As there is often confusion concerning the indications and scientific basis for the use of antibiotics in conjunction with dental procedures [44, 48, 86, 122, 127, 144, 170], the present chapter reviews the literature regarding the scientific rationale for antibiotic prophylaxis and summarizes a series of practice guidelines used in making clinical decisions. This presents a dilemma for the dentist because he or she may feel obligated to use antibiotic prophylaxis in inappropriate or unnecessary scenarios [156].

Recently, Lockhart et al. [91] reviewed the medical and dental literature for scientific evidence regarding the use of antibiotics to prevent local and systemic infections associated with dental treatment. Situations commonly considered by dentists for potential use of prophylactic antibiotics were reviewed to determine current evidence with regard to use of antimicrobial agents. This included prevention of distant spread of oral organisms to susceptible sites elsewhere in the body and the reduction of local infections associated with oral procedures. The authors concluded that there are relatively few situations in which antibiotic prophylaxis is indicated. Aside from the clearly defined instances of endocarditis and late prosthetic joint infections, there is no consensus among experts on the need for prophylaxis. There is wide variation in

recommended protocols, but little scientific basis for the recommendations. The emerging trend seems to be to avoid the prophylactic use of antibiotics in conjunction with dental treatment unless there is a clear indication. The risk of inappropriate use of antibiotics and widespread antibiotic resistance appear to be far more important than any possible perceived benefit [91].

## 2.1 Levels of Bacteremia Associated with Interventional Procedures and Everyday Activities

The basis for many of the decisions that have been made regarding which procedures merit antibiotic prophylaxis is the assumption that the bacteremia that arises following interventional procedures is a key part of the causative process in the development of infective endocarditis (IE) (NICE 2008).

### 2.1.1 Frequency of Bacteremia Associated with Dental Procedures and Oral Activities

Transient bacteremia is common with manipulation of the teeth and periodontal tissues, and there is a wide variation in reported frequencies of bacteremia in patients resulting from dental procedures: tooth extraction (10–100%), periodontal surgery (36–88%), scaling and root planning (8–80%), teeth cleaning (up to 40%), rubber dam matrix/wedge placement (9–32%), and endodontic procedures (up to 20%). Transient bacteremia also occurs frequently during routine daily activities unrelated to a dental procedure, such as tooth-brushing and flossing (20–68%), use of wooden toothpicks (20–40%), use of water irrigation devices (7–50%), and chewing food (7–51%) [168] (Tables 2.1–2.5)

It should be noted that there is a wide range of values between different studies. This may be a result of different analytical methods and sampling procedures, and these results should be interpreted with caution [123, 152]. If anaerobic techniques are added then a

wider range and greater extent of bacteremia is demonstrated. The time at which blood is taken for analysis and the frequency in a single patient are also key values to examine when reviewing bacteremia studies [123, 145]. The incidence after other types of medical procedures is even less well-established [152].

### 2.1.2 Nature of Bacteremia Associated with Dental Procedures and Oral Activities

As evident from Tables 2.1–2.6, bacteria of potential oral origin detected in odontogenic bacteremias show some degree of diversity, but to a much lesser extent than that of the oral cavity. It is probable that this lack of diversity of blood-borne bacterial species is due to (1) their virulence attributes that facilitate entry into the bloodstream, (2) the rapid clearance of the bacteria by host defenses, and (3) the low threshold of the current detection methods used in clinical laboratories. However, as in the oral niche, the predominant bacterial genus isolated from the blood is *Streptococcus*. More than half of systemic affections are of streptococcal etiology, indicating that the predominance of streptococci in the oral niche could be a factor in determining their entry into the bloodstream [123].

### 2.1.3 Magnitude of Bacteremia Associated with Dental Procedures and Oral Activities

Few published studies exist on the magnitude of bacteremia after a dental procedure or from routine daily activities [26, 50, 52, 99–101, 133, 137]. There are no published data that demonstrate that a greater magnitude of bacteremia, compared with a lower magnitude, is more likely to cause IE in humans. The magnitude of bacteremia resulting from a dental procedure is relatively low ( $<10^4$  colony-forming units of bacteria per milliliter), similar to that resulting from routine daily activities, and is less than that used to cause experimental IE in animals ( $10^6$  to  $10^8$  colony-forming units of bacteria per milliliter) [42, 168].

**Table 2.1** Summary of studies on bacteremia associated with oral hygiene measures

| Reference          | Subjects  | Dental procedure or oral activities evaluated  | Sampling time                                      | Identified species  | Outcomes   |
|--------------------|---|--|--|---|--|
| Crasta et al. [29] | 30 individuals with chronic periodontitis (29–75 years) and 30 with periodontal health (28–71 years)        | Dental flossing  | Baseline, 30 s and 10 min after flossing cessation | <i>Viridans streptococci</i> were isolated from 19% of positive subjects and accounted for 35% of microbial isolates.   | 40% of periodontitis subjects and 41% of periodontally healthy subjects tested positive for bacteremia following flossing. 20% of subjects had a detectable bacteremia at 10 min post-flossing. No patient or clinical factors were significantly associated with post-flossing bacteremia   |
| Lucas et al. [99]  | 141 children and adolescents, aged between 3 and 17 years, having dental treatment under general anesthesia | Toothbrushing (baseline) with (1) Oral B 30 toothbrush or (2) Braun or (3) Sonicare electric toothbrush or (4) dental handpiece and rubber cup | Before and 30 s after toothbrushing                | A wide variety of bacteria were isolated. Of these, oral <i>Streptococcus</i> spp. comprised 2% and 15% at baseline and 30 s after toothbrushing, respectively. Coagulase Negative <i>Staphylococcus</i> spp. comprised 12 and 24% at baseline and after toothbrushing, respectively. Other bacteria included <i>Lactobacillus</i> spp., <i>Actinomyces</i> spp., <i>Neisseria</i> spp. and <i>Micrococcus</i> spp. No obligate anaerobes were detected | There was a significantly greater prevalence of bacteremia following the use of the dental handpiece and rubber cup only, compared with baseline (37% vs 15%) ( $P = 0.02$ ). There was a significantly greater aerobic intensity of bacteremia per milliliter of blood following brushing with the Sonicare compared with baseline ( $P = 0.03$ ). There was also a significantly greater aerobic ( $P = 0.001$ ) and anaerobic ( $P = 0.005$ ) bacteremia following the slow handpiece and rubber cup compared with baseline |
| Bhanji et al. [20] | 50 children receiving dental treatment under general anesthesia   | Manual or Sonicare toothbrushing   | After induction and 30 s after brushing            | Mainly <i>Streptococcus</i> spp.  | The Sonicare induced significantly more bacteremias than manual toothbrushing. The frequency of bacteremia was 46% with manual brushing; 18% aerobic, 9% anaerobic, and 73% both. This differed significantly ( $P < 0.05$ ) with 78% bacteremias in the Sonicare group; 22% aerobic, 22% anaerobic, and 56% both  |

(continued)

**Table 2.1** (continued)

| Reference            | Subjects   | Dental procedure or oral activities evaluated   | Sampling time   | Identified species   | Outcomes  |
|----------------------|--|---|---|--|---|
| Forner et al. [54]   | 20 healthy individuals, 20 patients with gingivitis, 20 patients with periodontitis                    | Three experimental procedures: (1) Chewing on three pieces of chewing gum base performed for 10 min. (2). Supervised toothbrushing performed for 2 min. (3). Full-mouth scaling performed according to individual need by a combination of hand instruments and airscaler | Baseline and at 0.5, 10, and 30 min   | A total of 163 isolates were collected from 29 bacteremic episodes in 23 of the 60 participants. The isolates were from two healthy individuals, four gingivitis patients, and 17 periodontitis patients after chewing, toothbrushing, or scaling. The bacterial isolates included a range of <i>Streptococcus</i> , <i>Enterococcus</i> , <i>Actinomyces</i> , <i>Lactobacillus</i> , <i>Corynebacterium</i> , <i>Porphyromonas</i> , <i>Prevotella</i> , and <i>Fusobacterium</i> species in addition to one isolate of <i>Candida</i> . The most predominant <i>Streptococcus</i> species were <i>S. mitis</i> , <i>S. oralis</i> , and <i>S. sanguis</i> | Following chewing four (20%) of the periodontitis patients were bacteremic whereas none of the periodontally healthy individuals or gingivitis patients showed evidence of bacteremia. None of the periodontally healthy individuals or gingivitis patients was bacteremic after toothbrushing. However, one (5%) of the periodontitis patients was bacteremic immediately after toothbrushing. After 30 min, bacteremia was found in another patient from whom no bacteria were recovered in the blood samples collected at 0.5 or 10 min after toothbrushing. Bacteremia after scaling occurred in two (10%) of the healthy participants, in four (20%) of the gingivitis patients, and in 15 (75%) of the periodontitis patients. The magnitude decreased considerably within the 30 min |
| Hartzell et al. [68] | 30 military beneficiaries  | Routine toothbrushing for 1 min using a standardized soft-bristle toothbrush  | 3 different time points: at baseline and 30 s and 20 min after brushing   | Three of 180 blood cultures were positive for <i>Propionibacterium acnes</i> (a known contaminant)   | The rate of true bacteremia in this study was zero, which is much lower than previous studies. Bacteremia after tooth-brushing in a healthy population is a rare occurrence   |
| Lockhart et al. [94] | 290 subjects presented to urgent care service with the need for extraction of at least 1 erupted tooth | (1) toothbrushing, (2) single-tooth extraction with amoxicillin prophylaxis, or (3) single-tooth extraction with identical placebo  | 1.5 min and at 5 min after the initiation of surgery or brushing. Additional blood samples were drawn 20, 40, and 60 min after the end of the procedure | 98 different bacterial species, the most common of which belonged to the genera <i>Streptococcus</i> (49%), <i>Prevotella</i> (9%), <i>Actinomyces</i> (5%), and <i>Fusobacterium</i> (5%)   | Cumulative incidence of endocarditis-related bacteria from all 6 blood draws was 23%, 33%, and 60% for the toothbrushing, extraction-amoxicillin, and extraction-placebo groups, respectively ( $P < 0.0001$ ). The vast majority of bacteremic subjects (93%) had a brief duration of bacteremia (<20 min). There was a significant drop in the incidence of positive cultures at 20 min in all 3 groups (all $P < 0.0001$ ), and this continued at 40 and 60 min, with little difference between the brushing and extraction-amoxicillin groups at draws 4 to 6. Five percent of subjects in the extraction-placebo group and 2% of the brushing group were still bacteremic at 60 min  |

|                      |  |   |   |   |   |
|----------------------|--|---|---|---|---|
| Lockhart et al. [95] | 290 subjects presented to urgent care service with the need for extraction of at least 1 erupted tooth | (1) Toothbrushing, (2) single-tooth extraction with placebo                 | During and immediately after toothbrushing or extraction (draws 2 and 3); and at 20, 40, and 60 min after toothbrushing or extraction | 43.8% of the 32 IE-associated oral bacterial species were viridans streptococci, 13 of 27 (48.1%) cultures positive for bacteria in the toothbrushing group contained viridans streptococci, compared with 106 of 152 (69.7%) cultures positive for bacteria in the extraction group ( $P < 0.05$ ) | Cumulative incidence of IE-related bacteremia was 22.5% and 60.4% for the toothbrushing and extraction groups. The risk of developing bacteremia increased 6% for each additional year of age. In addition, all measures of oral hygiene (mean plaque score, plaque score of $\geq 2$ , mean calculus score, calculus score of $\geq 2$ ) and one measure of gingival bleeding (generalized bleeding with toothbrushing) were significantly associated with IE-related bacteremia. Participants with mean plaque or calculus scores of 2 or greater had 3.78- and 4.43-fold increased risks, respectively, of developing an IE-related bacteremia after toothbrushing ( $P < 0.01$ ). In addition, the presence of generalized bleeding after toothbrushing was associated with an almost eightfold risk of developing bacteremia caused by oral bacterial species implicated in IE |
| Lucas & Roberts [98] | 155 children receiving dental treatment under general anesthesia                                       | (1) toothbrushing, (2) professional cleaning with a rubber cup, (3) scaling | Baseline and 30 s after cleaning  | <i>S. mitis</i> , <i>S. sanguis</i> , and Coagulase-negative <i>staphylococci</i>   | There was no significant difference in the prevalence of positive or intensity of bacteremia between the three groups (Brushing, 39%, cleaning 25%, scaling, 40%)   |

**Table 2.2** Summary of studies on bacteremia associated with periodontal procedures

| Reference          | Subjects  | Dental procedure or oral activities evaluated   | Sampling time  | Identified species  | Outcomes  |
|--------------------|---|---|--|---|---|
| Allison et al. [4] | 12 untreated dental patients                              | Ultrasonic scaling and root planning associated pre- and intraoperatively with an irrigant containing 0.12% CHX versus placebo solution | Preoperatively, and postoperative at 1 min after completing the scaling of each quadrant and then 10 min after scaling the second quadrant | 36 positive cultures, 13 of which were viridans group streptococci  | 9/12 in Placebo group (75%), 3/12 (25%) in CHX group exhibited bacteremia. The results show that there was no difference in the distribution or presentation of periodontal disease between the experimental and control quadrants  |
| Assaf et al. [13]  | 22 plaque-induced generalized chronic gingivitis patients | Use of diode lasers (DLs) to reduce bacteremia associated with ultrasonic scaling (controls US only, cases: US + DL)                    | Controls: just before and 3 min after initiation of US; Cases: 30 min after DL, just before and 3 min after initiation of US               | Seven of the 23 positive cultures contained more than one species; polymicrobial bacteremia containing both aerobic and anaerobic bacteria was also detected. The identified microorganisms were: <i>Streptococcus</i> spp., <i>Prevotella intermedia</i> and <i>P. nigrescens</i> , <i>Prevotella melaninogenica</i> , <i>Capnocytophaga</i> spp., <i>Haemophilus</i> spp., <i>Bacteroides</i> spp., and <i>Fusobacterium</i> spp. Among all samples, <i>Streptococcus</i> spp. were the most commonly detected (61%). <i>Streptococcus mitis</i> , <i>S. salivarius</i> , and <i>S. sanguis</i> were frequently recovered from the bloodstream throughout the study | Among the 22 patients, 15 (68%) had detectable bacteremia after US alone, and 8 patients (36%) had detectable bacteremia following DL + US. All patients with negative blood cultures after DL + US as well; this was the case for a total of seven patients who showed no bacteremia at any step throughout the whole experiment. Thus, eight patients showed positive blood cultures associated with both, US and DL + US. However, seven patients had negative blood cultures after DL + US, though they showed a positive result after US alone. When compared to US alone, DL + US showed a significant reduction in the prevalence of odontogenic bacteremia ( $P < 0.05$ ) |

|                    |  |   |  |   |  |
|--------------------|--|---|--|---|--|
| Cherry et al. [26] | 60 patients with gingivitis              | 0.9% saline and 7.5% povidone-iodine rinses for 2 min before ultrasonic scaling | Before and after 30 s and 2 min of scaling | Bacterial isolates were recovered from the baseline blood samples of four subjects (three NaCl; one POV-1) and consisted of coagulase-negative staphylococci, one mixed colony of <i>S. epidermidis/Corynebacterium</i> spp. and a yeast. No microorganisms of oral origin were found in any of the baseline blood samples.     | Bacteremia occurred in 33.3% of the saline group and 10% of the povidone-iodine group. Regression analysis showed that rinsing with povidone-iodine was approximately 80% more effective than rinsing with saline in reducing the occurrence of bacteremia, with a statistically significant odds ratio of 0.189 (95% CI: 0.043–0.827). Bacteremia magnitude was 0.1 colony-forming units/ml in the povidone-iodine subjects and 0.1–0.7 CFU/ml. in the saline group. Age was the only factor significantly predictive for the development of a post-scaling bacteremia ( $P<0.05$ , OR=1.40, 95% CI: 1.00–1.97). The OR for the recovery of a bacteremia following scaling increased by 1.4 for every 10-year increase in age of the subjects. No other patient characteristics or clinical parameters were found to have a significant association with the occurrence of bacteremia |
| Daly et al. [33]   | 30 patients with untreated periodontitis | Periodontal probing using a standard periodontal probe                          | Before and immediately after probing       | Aerobic/facultative isolates ( <i>Viridans streptococcus</i> spp. 45%, <i>Corynebacterium</i> spp. 15%, <i>Coagulase negative staphylococci</i> 10%, <i>Gemella</i> spp. 5%, <i>Streptococcus milleri</i> 5%), <i>Anaerobic isolates (Bacteroides</i> spp. 10%, <i>Desulfomonas</i> spp. 5%, <i>Peptostreptococcus</i> spp. 5%) | 13 patients (43%) exhibited bacteremia of oral origin. No association was found between the severity of periodontitis, as determined by the deepest pockets recorded, and the occurrence of bacteremia. The results indicate that periodontal probing can cause bacteremia in patients with periodontitis.   |

(continued)

**Table 2.2** (continued)

| Reference        | Subjects  | Dental procedure or oral activities evaluated  | Sampling time  | Identified species   | Outcomes   |
|------------------|---|--|--|--|--|
| Daly et al. [35] | 20 patients with adult periodontitis and 20 with chronic gingivitis | Periodontal probing using a standard periodontal probe   | Prior to and immediately following periodontal probing | <i>Streptococcus</i> spp. were the most common isolates in both groups | Probing caused bacteremia of oral origin in eight (40%) of the periodontitis patients and two (10%) of the gingivitis patients. Compared with the gingivitis group the odds ratio (OR) for bacteremia in the periodontitis group was 5.993 (95% CI 1.081 to 33.215). Bleeding on probing (OR 1.025, 95% CI 1.004 to 1.047) and mean probing depth per tooth (OR 1.444, 95% CI 1.055 to 1.977) were significantly associated with bacteremia. No significant correlations were found between bacteremia and age, number of teeth probed, smoking status, PI, or total probing depth   |
| Fine et al. [52] | 18 healthy subjects with measurable bacteremia                      | 5 min ultrasonic subgingival scaling and subgingival irrigation with an antiseptic mouth rinse (cases) or a 5% hydroalcohol (controls) | Baseline and postoperatively                           | Not given  | Aerobic and anaerobic microbiological counts were detected for all 18 subjects who completed the study. Aerobic counts were 2.44 CFU/mL for the antiseptic mouth rinse and 3.22 CFU/mL for the control mouth rinse. Mean posttreatment aerobic counts were 4.67 CFU/mL for the antiseptic mouthrinse and 38.72 CFU/mL for the control mouth rinse. The mean pretreatment anaerobic colony counts were 0.50 CFU/mL for the antiseptic mouth rinse and 1.78 CFU/mL for the control mouth rinse. Mean posttreatment anaerobic colony counts were 1.61 CFU/mL for the antiseptic mouth rinse and 14.89 CFU/mL for the control mouth rinse. Overall, the antiseptic mouth rinse resulted in aerobic counts that were 92.3% lower than those with the control mouth rinse and anaerobic counts that were 87.8% lower. These differences were statistically significant ( $P < 0.0001$ ). |

|                       |   |  |  |  |  |
|-----------------------|---|--|--|--|--|
| Forner et al.<br>[54] | 20 healthy individuals, 20 patients with gingivitis, 20 patients with periodontitis | 3 experimental procedures: (1) Chewing on three pieces of chewing gum base performed for 10 min (2). Supervised toothbrushing performed for 2 min (3) Full-mouth scaling performed according to individual need by a combination of hand instruments and airscaler | Baseline and at 0.5, 10, and 30 min  | A total of 163 isolates were collected from 29 bacteremic episodes in 23 of the 60 participants. The isolates were from two healthy individuals, four gingivitis patients, and 17 periodontitis patients after chewing, toothbrushing, or scaling. The bacterial isolates included a range of <i>Streptococcus</i> , <i>Enterococcus</i> , <i>Actinomyces</i> , <i>Lactobacillus</i> , <i>Corynebacterium</i> , <i>Porphyromonas</i> , <i>Prevotella</i> , and <i>Fusobacterium</i> species in addition to one isolate of <i>Candida</i> . The most predominant <i>Streptococcus</i> species were <i>S. mitis</i> , <i>S. oralis</i> , and <i>S. sanguis</i> . | Following chewing four (20%) of the periodontitis patients were bacteremic whereas none of the periodontally healthy individuals or gingivitis patients showed evidence of bacteremia. None of the periodontally healthy individuals or gingivitis patients was bacteremic after toothbrushing. However, one (5%) of the periodontitis patients was bacteremic immediately after tooth brushing. After 30 min, bacteremia was found in another patient from whom no bacteria were recovered in the blood samples collected at 0.5 or 10 min after toothbrushing. Bacteremia after scaling occurred in two (10%) of the healthy participants, in four (20%) of the gingivitis patients, and in 15 (75%) of the periodontitis patients. The magnitude decreased considerably within the 30 min |
| Kinane et al.<br>[80] | 30 volunteers with untreated periodontal disease                                    | Periodontal probing, supervised 2 min toothbrushing, full-mouth ultrasonic scaling   | Sample 1: at baseline, Sample 2: following full-mouth periodontal probing depth (range 30 s to 1 min), Sample 3: at the second visit, 2 weeks later, Sample 4: after supervised 2 min toothbrushing (never more than 3 min), Sample 5: immediately following full-mouth ultrasonic scaling | Several organisms were detected: <i>Anaerobic Streptococci</i> , <i>Propionobacterium acnes</i> , <i>Neisseria pharyngis</i> , <i>Streptococcus viridans</i> , <i>Micrococcus</i> , <i>Staphylococcus albus</i> , <i>Hemophilus aphrophilus</i> , <i>Coag-ve Staphylococci</i> <i>Micrococcus</i> , <i>Prevotella intermedia</i> , <i>Actinomyces naeslundii</i> , <i>Gamella haemolysins</i> , <i>Streptococcus parasanguis</i> , <i>A. naeslundii</i> , <i>Eubacterium sp.</i> , <i>Eubacterium limosum</i> , <i>A. Naeslundii</i> , <i>Streptococcus parasanguis</i> , <i>Propionobacterium acnes</i>   | Using culture methods, the incidence of bacteremias was as follows: following ultrasonic scaling (13%), periodontal probing (20%), and toothbrushing (3%). PCR analysis revealed bacteremia incidences following ultrasonic scaling, periodontal probing, and toothbrushing of 23%, 16%, and 13%, respectively   |

(continued)

**Table 2.2** (continued)

| Reference            | Subjects  | Dental procedure or oral activities evaluated | Sampling time  | Identified species  | Outcomes  |
|----------------------|---|---|--|---|---|
| Lafaurie et al. [83] | 27 patients with generalized severe chronic periodontitis (GChP) and 15 patients with generalized aggressive periodontitis (GAgP) | Scaling and root planning                     | Immediately before the SRP procedure (T1), immediately after treatment (T2), 15 min after treatment (T3) and 30 min after treatment (T4) | The periodontopathic microorganisms more frequently found in peripheral blood were <i>P. gingivalis</i> , and <i>M. micros</i> . <i>Campylobacter</i> spp., <i>E. corrodens</i> , <i>T. forsythensis</i> , <i>Fusobacterium</i> spp., and <i>P. intermedius</i> were isolated less frequently. <i>Actinomyces</i> spp. Was isolated frequently and <i>Capnocytophaga</i> spp., <i>Prevotella melanogena</i> , <i>Propionibacterium acnes</i> , <i>Bifidobacterium</i> spp., <i>Eubacterium aerofaciens</i> and <i>Gemella morbillorum</i> were also isolated but in lower proportion. ( $P < 0.05$ ). In 73.8% (31/42) of the patients, bacteria were isolated in blood immediately after treatment (T2). In 38% (16/42) of the patients, bacteremia was evident after 15 min (T3), and 19% (8/42) still had positive cultures after 30 min (T4). One patient (2.4%) presented a positive culture before SRP (T1) <i>P. gingivalis</i> , <i>Actinomyces</i> spp., <i>T. forsythensis</i> , and <i>P. intermedius</i> / <i>nigrescens</i> were isolated more frequently from the blood of GAgP patients than GChP patients but the differences were significant only for <i>T. forsythensis</i> ( $P < 0.05$ ) | Microorganisms growing under anaerobic conditions were identified in the peripheral blood of 80.9% (34/42) of the patients after SRP. Bacteremia was found in 93.7% (14/15) of the GAgP patients and 74.1% (20/27) of the GChP patients without significant differences between the two groups of periodontitis ( $P < 0.05$ ). |
| Maestre et al. [103] | 13 patients with generalized chronic periodontitis  | Scaling and root planning                     | Pre-treatment and immediately after odontology treatment (full-mouth scaling).   | The anaerobic bacteria ( <i>Prevotella</i> spp., <i>Micromonas micros</i> and <i>Fusobacterium nucleatum</i> ) were the most predominant microorganism.   | None of the 13 patients had bacteremia before the procedures. Bacteremia after scaling occurred in 10/13 (76.9%) of periodontitis patients.   |

|                             |   |   |   |
|-----------------------------|---|---|---|
| Pérez Chaparro et al. [126] | 16 unrelated, systemically healthy adults with either GCP (generalized chronic periodontitis) or GAP (generalized aggressive periodontitis). Patients must have had at least ten pockets with probing depth $\geq 7$ mm requiring periodontal surgery after scaling and root planning | Periodontal surgery after (H1) prior to the SRP; scaling and root planning (H2) immediately at the end of the procedure; (H3) 15 min and (H4) 30 min after the end of SRP was assessed during 10 min (10 sites, 1 min per site)   | <i>P. gingivalis</i><br>Seven patients showed positive <i>P. gingivalis</i> bacteremia. The most frequent <i>fmA</i> type II, followed by Ib, III, and IV. In blood strains, type II was followed by IV, Ib, and III  |
| Reinhardt et al. [133]      | 30 healthy human volunteers   | Sterile and nonsterile water irrigation during ultrasonic scaling   | Baseline and immediately postoperative Gram-positive aerobic and anaerobic cocci<br>The difference in the bacteremia incidence after scaling with sterile water (50%) versus scaling with tap water (53.3%) was not significant. The degree of the bacteremias (less than 1 colony forming unit/mL) was similar between groups. Therefore tap water irrigation used in ultrasonic scaling did not appear to be a significant causative agent in postoperative bacteremias |
| Waki et al. [165]           | 60 periodontal maintenance patients   | 4 groups: (1) subgingival irrigation, with 0.12% CHX and daily marginal irrigation with 0.04% CHX; (2) subgingival irrigation with 0.12% CHX and daily marginal irrigation with water; (3) subgingival and daily marginal irrigation with water; (4) Non-irrigation (control) | <i>Eubacterium lentum</i> ,<br><i>Propionibacterium acnes</i> ,<br><i>Streptococcus species</i> , <i>Neisseria species</i> , <i>Candida albicans</i> ,<br><i>Staphylococcus species</i> , and un-identified Gram-negative rods<br>Bacteremia was detected prior to Sc/RP in 2 patients (3.33%) and after Sc/RP in 10 patients (16.67%). No significant effect by treatment regimens on post-Sc/RP bacteremia could be detected  |

**Table 2.3** Summary of studies on bacteremia associated with surgical procedures

| Reference                   | Subjects   | Dental procedure or oral activities evaluated   | Sampling time   | Identified species  | Outcomes   |
|-----------------------------|--|---|---|---|--|
| Bahrani-Mougeot et al. [15] | 290 healthy adults requiring the extraction of a tooth and having at least ten remaining teeth   | 3 groups: (i) 2 min of toothbrushing, (ii) single-tooth extraction with the American Heart Association recommended dose of oral amoxicillin administered 1 h prior to the procedure, and (iii) tooth extraction with a placebo (6).   | Draw 1, baseline before the procedure; draw 2, 90 s into the procedure; draw 3, 3.5 min after draw 2; draws 4, 5, and 6, 20, 40, and 60 min, respectively, following the completion of the dental procedure | Sequence analysis of 16S rRNA genes identified 98 different bacterial species recovered from 151 bacteremic subjects. Of interest, 48 of the isolates represented 19 novel species of <i>Prevotella</i> , <i>Fusobacterium</i> , <i>Streptococcus</i> , <i>Actinomyces</i> , <i>Capnocytophaga</i> , <i>Selenomonas</i> , and <i>Veillonella</i>  | 151 bacteremic subjects (52.07%), 20% were still bacteremic at 20 min, 9% at 40 min, and 6% at 60 min. Antibiotic prophylaxis reduced the incidence of bacteremia from dental extraction. It also resulted in bacteremia with fewer bacterial species, which were cleared from the blood in a shorter time (i.e., mostly within 20 min). Although antibiotic prophylaxis reduced the bacteremia of several species of streptococci, as expected, it does not seem to affect species of proteobacteria (e.g., <i>Escherichia coli</i> ) and <i>Prevotella</i> |
| Diz Dios et al. [41]        | 221 patients who, for behavioral reasons (autism, learning disabilities, phobias, etc.), underwent dental extractions under general anesthesia | (1) 53 control patients who did not receive any type of prophylaxis before extraction; (2) 56 patients in the AMX group: 2 g of Amoxicillin orally 1–2 h before extraction; (3) 54 patients received clindamycin 600 mg orally 1–2 h before extraction; (3) 58 patients received 400 mg Moxifloxacin orally 1–2 h before extraction | Baseline and 30 s, 15 min, and 1 h after the dental extractions   | The most frequent bacterial genus in the positive blood cultures in the control group was <i>Streptococcus</i> (63.1%), particularly the viridans group, followed by the genera <i>Staphylococcus</i> (11.3%) and <i>Neisseria</i> (7.5%). In the AMX group, the most frequent bacterial isolates were viridans group streptococci (44.4%), all of which belonged to the <i>S. mitis</i> group, followed by obligate anaerobes, such as <i>Pepostreptococcus</i> spp. (11.1%) and <i>Prevotella</i> spp. (11.1%). In the CLJ group, the most frequent bacterial genus was <i>Streptococcus</i> (58.5%), particularly the viridans group, followed by the genera <i>Neisseria</i> (14.8%) and <i>Prevotella</i> (5.9%). In the MXF group, the most frequent isolates were viridans group streptococci (67.7%), followed by <i>Staphylococcus</i> spp. (9.7%) and obligately anaerobic bacteria (9.7%). |  |

|                  |                     |  |   |  |  |
|------------------|---------------------|--|---|--|--|
| Hall et al. [67] | 38 healthy patients | Erythromycin (1 g) or clindamycin (0.6 g) orally 1.5 h prior to dental extraction  | Before, during and again 10 min after dental extraction | Anaerobic bacteria dominated the findings of postextraction bacteremia and Gram-positive strains greatly outnumbered Gram-negative bacteria. Aerobic bacteria other than viridans streptococci were recovered infrequently   | The incidence of bacteremia found during dental extraction was 79% in the erythromycin group and 84% in the clindamycin group. Ten minutes after extraction the incidence had decreased to 58% and 53% respectively. The overall incidence of viridans streptococcal bacteremia was 79% in the erythromycin group and 74% in the clindamycin group (two patients in the clindamycin group developed bacteremia 10 min after extraction only). Strains of <i>Streptococcus intermedius</i> were most frequently isolated followed by strains of <i>Streptococcus mitis</i> and <i>Streptococcus sanguis</i> in both prophylaxis groups. The incidence of anaerobic bacteremia during dental extraction was 58% in the erythromycin group and 74% in the clindamycin group. Species of <i>Actinomyces</i> , <i>Eubacterium</i> , and <i>Lactobacillus</i> were most commonly recovered while <i>Propionibacterium acnes</i> and <i>Bacteroides</i> species were isolated from single patients only |
| Hall et al. [66] | 39 healthy patients | 1 g cefaclor (19 patients) or placebo (20 patients) 1 h prior to dental extraction | Before, during and again 10 min after dental extraction | Aerobic bacteremia: Strains of <i>Streptococcus intermedius</i> were most frequently isolated, followed by strains of <i>Streptococcus mitis</i> in both patient groups. Anaerobic bacteremia: <i>Actinomyces</i> spp. were the most commonly recovered strains while Gram-negative strains of <i>Veillonella</i> and <i>Prevotella</i> were only isolated from single patients. | The incidence of positive blood cultures during dental extraction was 79% in the cefaclor group and 85% in the placebo group. Ten minutes after extraction the incidence had decreased to 53% and 47%, respectively. The incidence of bacteremia with viridans streptococci during extraction was 79% in the cefaclor group and 50% in the placebo group. Ten minutes after extraction the incidence was 26% and 30%, respectively. The incidence of anaerobic bacteremia during and 10 min after dental extraction was 74% and 47%, respectively, in the cefaclor group, and 75% and 35%, respectively, in the placebo group. No difference in the incidence or magnitude of bacteremia was observed when the two patient groups were compared.   |

(continued)

**Table 2.3** (continued)

| Reference            | Subjects   | Dental procedure or oral activities evaluated  | Sampling time  | Identified species  | Outcomes   |
|----------------------|--|--|--|---|--|
| Lockhart et al. [93] | 100 children enrolled (mean age, 3.5 years) requiring dental treatment in the operating room settings  | Amoxicillin elixir (50 mg/kg) or placebo 1 h before nasotracheal intubation and dental procedures                                  | At 8 specific time points after intubation, dental restorative and cleaning procedures, and before, during, and after dental extraction(s), to include blood drawings up to 45 min after the last extraction | The majority of the 152 positive cultures and of the 29 different bacteria identified were Gram-positive cocci. There was a >5-fold difference in the number of positive blood cultures recovered from the placebo ( $n=128$ ) versus the amoxicillin ( $n=24$ ) groups. <i>Viridans streptococci</i> made up 45% ( $n=57$ ) of the total bacteria cultured in the placebo group versus 33% ( $n=8$ ) of the amoxicillin group. | The overall incidence of positive cultures from all 8 draws was greater in the placebo group (84%) than the amoxicillin group (33%) ( $P<0.0001$ ). The highest incidence of positive cultures at a single time point occurred 1.5 min after completion of tooth extraction(s) in the placebo group (76%) versus the amoxicillin group (15%) ( $P<0.0001$ ). The incidence of bacteraemia after intubation and restorative and cleaning procedures was 18% and 20% in the placebo groups and 4% and 6% in the amoxicillin groups ( $P=0.05$ and 0.07, respectively). Subjects in the placebo group had a bacteraemia incidence of 18% at 15 min, 16% at 30 min, and 14% at 45 min. By contrast, only 1 subject in the amoxicillin group had bacteraemia at 15 min after extraction(s). |
| Lockhart et al. [94] | 290 subjects presented to urgent care service with the need for extraction of at least 1 erupted tooth | (1) toothbrushing, (2) single-tooth extraction with amoxicillin prophylaxis, or (3) single-tooth extraction with identical placebo | 1.5 min and at 5 min after the initiation of surgery or brushing. Additional blood samples were drawn 20, 40, and 60 min after the end of the procedure  | 98 different bacterial species, the most common of which belonged to the genera <i>Streptococcus</i> (49%), <i>Prevotella</i> (9%), <i>Actinomyces</i> (5%), and <i>Fusobacterium</i> (5%).   | Cumulative incidence of endocarditis-related bacteria from all 6 blood draws was 23%, 33%, and 60% for the toothbrushing, extraction-amoxicillin, and extraction-placebo groups, respectively ( $P<0.0001$ ). The vast majority of bacteremic subjects (93%) had a brief duration of bacteraemia (<20 min). There was a significant drop in the incidence of positive cultures at 20 min in all 3 groups (all $P<0.0001$ ), and this continued at 40 and 60 min, with little difference between the brushing and extraction-amoxicillin groups at draws 4 to 6. 5% of subjects in the extraction-placebo group and 2% of the brushing group were still bacteremic at 60 min.   |

|                      |  |  |  |  |   |
|----------------------|--|--|--|--|---|
| Lockhart et al. [95] | 290 subjects presented to urgent care service with the need for extraction of at least 1 erupted tooth                                   | (1) toothbrushing, (2) single-tooth extraction with placebo  | During and immediately after toothbrushing or extraction (draws 2 and 3); and at 20, 40 and 60 min after toothbrushing or extraction | 43.8% of the 32 IE associated oral bacterial species were viridans streptococci. 13 of 27 (48.1%) cultures positive for bacteria in the toothbrushing group contained viridans streptococci, compared with 106 of 152 (69.7%) cultures positive for bacteria in the extraction group ( $P < 0.05$ ). | Cumulative incidence of IE-related bacteremia was 22.5% and 60.4% for the toothbrushing and extraction groups. The risk of developing bacteremia increased 6% for each additional year of age. In addition, all measures of oral hygiene (mean plaque score, plaque score of $\geq 2$ , mean calculus score, calculus score of $\geq 2$ ) and one measure of gingival bleeding (generalized bleeding with toothbrushing) were significantly associated with IE-related bacteremia. Participants with mean plaque or calculus scores of 2 or greater had 3.75- and 4.43-fold increased risks, respectively, of developing an IE-related bacteremia after toothbrushing ( $P < 0.01$ ). In addition, the presence of generalized bleeding after toothbrushing was associated with an almost eightfold risk of developing bacteremia caused by oral bacterial species implicated in IE |
| Rahn et al. [130]    | 120 patients who were scheduled for dental treatment involving either an intraligamentary injection or an elective extraction of a molar | Comparison of the efficacy of 10% povidone-iodine solution (40 cases), 0.2% CHX (40 cases), sterile water (40 controls) applied into the sulcus of the affected tooth in preventing posttreatment bacteremia | Before the dentist administered the antiseptic and 2, 4, and 6 min after the dental procedure was finished                           | <i>Viridans streptococci</i> were detected in 13 cultures of the control group, four of the povidone-iodine group and 14 of the chlorhexidine group  | Bacteremia was detected in 21 control subjects (52.5%), 11 povidone iodine subjects (27.5%), and 18 chlorhexidine subjects (45.0%). 206 organisms were identified: 87 in the control group, 42 in the povidone-iodine group and 77 in the chlorhexidine group. The difference between the incidence of bacteremia (aerobes and anaerobes) in the povidone-iodine group and the control group was statistically significant ( $P < 0.05$ ). There was also a statistically significant difference between the incidence of <i>viridans streptococci</i> bacteremia in the povidone-iodine group and the chlorhexidine group with respect to the control group ( $P < 0.01$ ), but not between the chlorhexidine group alone and the control group  |

(continued)

**Table 2.3** (continued)

| Reference            | Subjects   | Dental procedure or oral activities evaluated  | Sampling time  | Identified species   | Outcomes  |
|----------------------|--|--|--|--|---|
| Rajasuo et al. [131] | 15 generally healthy young conscripts serving in the Finnish Defence Forces referred for surgical extraction of a mandibular third molar tooth | Surgical extraction of a mandibular third molar tooth after extraction   | 1 min after incision, 1, 5, 10, 15, and 30 min after extraction                            | On average, $3.9 \pm 2.6$ bacterial species per bacteremia-positive subject were detected in the blood. A total of 31 different bacterial species or groups was isolated from the blood cultures: 8 aerobic and 23 anaerobic. Anaerobic <i>Prevotella</i> , <i>Eubacterium</i> , and <i>Peptostreptococcus</i> species predominated, whereas viridans-group streptococci and <i>Streptococcus milleri</i> group were the most common aerobic species. All but one bacteremia-positive subject (93%) had at least one species detected both in the blood and in the pericoronal pocket samples. 43% of the cases had at least one and the same species detected simultaneously in the extraction socket sample and in the blood | Of the 16 subjects, 14 (88%) had detectable bacteremia. Anaerobic species were found in every case, and five subjects were also positive for aerobic species. Half the subjects showed bacteremia immediately after the incision, whereas 44% were culture-positive at 1 and 10 min after extraction. Two subjects still had positive results 30 min after surgery  |
| Roberts et al. [137] | 207 children   | Four groups: a baseline with no surgical intervention (group I), after a single-tooth extraction (group II), multiple-tooth extraction (group III), and mucoperiosteal flap elevation (group IV) | After anesthesia (baseline), during the procedure, and after the procedure                 | 113 different isolates, 57% of which were streptococci   | The broth culture was positive for group I 111% of the time, group II for 43%, group III for 54%, and group IV for 43%. The Pediatric Isolator system was found to be a poor method for detecting bacteremia, having only one quarter the sensitivity of the broth culture technique. When organisms were isolated, the intensity of bacteremia ranged from 1 to 3,400 colony forming units per milliliter (cfu/mL) |
| Roberts et al. [136] | 500 children attending the Eastman Dental Hospital for treatment under general anesthesia (mean age 7.6 years)                                 | Dental extraction  | 10 s, 30 s, 1 min, 2 min, 4 min, 7.5 min, 15 min, 30 min, 45 min, and 1 h after extraction | The genera most often detected were <i>Streptococcus</i> , <i>Actinomyces</i> , and <i>Staphylococcus</i>  | Bacteremia was highest during and up to 7.5 min and then diminished rapidly as most of the bacteremia had been removed by the immune defense systems within 1.5 min   |

|                       |   |                         |   |
|-----------------------|---|-------------------------|---|
| Tomás et al.<br>[154] | 53 patients undergoing dental extractions under general anesthesia for behavioral reasons (autism, cerebral palsy, learning disabilities, hyperactivity, phobias, etc.) | Dental extractions      | <p>Baseline and at 30 s, 15 min and 1 h after the dental extractions</p> <p>A total of 133 bacterial strains were isolated of which 7.5% were aerobes, 82.7% were facultative anaerobes and 9.8% were obligate anaerobes. With respect to their Gram-stain pattern and morphology, 70.9% were Gram-positive cocci, 8.3% Gram-negative cocci, 4.5% Gram-positive bacilli, and 8.3% Gram-negative bacilli. The most frequent were <i>Streptococcus</i> spp. (63.8%), particularly <i>S. viridans</i>, followed by <i>Staphylococcus</i> spp. (11.2%) and <i>Neisseria</i> spp. (7.5%). Of the 73 isolates of <i>S. viridans</i>, 60% belonged to the <i>mitis</i> group, 29% to the <i>anginosus</i> group, 5.5% to the <i>salivarius</i> group, 4.1% to the <i>bovis</i> group and 1.4% to the <i>mutans</i> group. The most prevalent obligate anaerobes were <i>Fusobacterium</i> spp., <i>Peptostreptococcus</i> spp., <i>Bacteroides</i> spp., and <i>Prevotella</i> spp.</p> <p>A total of 131 bacteria were isolated from third molar postextraction blood cultures: 87.9% were <i>Streptococcus viridans</i>, 4.6% <i>Neisseria</i> spp., 1.5% <i>Staphylococcus</i> spp., 1.5% <i>Corynebacterium</i> spp., 1.5% <i>Leuconostoc</i> spp., 1.5% <i>Rothia dentocariosa</i>, and 1.5% other bacteria</p> |
| Tomás et al.<br>[155] | 100 patients undergoing third molar extractions under general anesthesia  | Third molar extractions | <p>Baseline, 30 s and 15 min after completing the final extraction</p> <p>The prevalence of bacteremia was 62% at 30 s after completing the first extraction of a mandibular third molar and 67% at 15 min after finishing the final extraction</p>   |

(continued)

**Table 2.3** (continued)

| Reference             | Subjects   | Dental procedure or oral activities evaluated  | Sampling time   | Identified species  | Outcomes  |
|-----------------------|--|--|---|---|---|
| Vergis et al. [162]   | 36 outpatients in a dental clinic (21–79 years) without underlying cardiac valvular disease or prosthetic cardiac valves | Topical antibiotic prophylaxis for bacteremia after dental extractions: (1) 2 mouthwash applications of 60-mL amoxicillin each retained 1 to 2 min, experimental prophylaxis, (2) six 500-mg amoxicillin capsules, standard prophylaxis, or (3) no prophylaxis | After dental extraction                                 | Twenty-three species of bacteria were isolated from 18 patients: viridans streptococci, 61% (14 of 23 species); <i>Fusobacterium</i> , 9% (2 of 23 species); and 1 species each for nonhemolytic streptococci, <i>Veillonella</i> , <i>Actinomyces viscosus</i> , and coagulase-negative staphylococci. Four patients had multiple organisms isolated from the blood cultures                                     | Breakthrough bacteremia after dental extraction was observed in 60% (6 of 10 patients) who received topical amoxicillin and in 89% (8 of 9 patients) who received no prophylaxis ( $P=0.30$ ). By comparison, breakthrough bacteremia after dental extraction was observed in 10% (1 of 10 patients) who received standard prophylaxis with oral amoxicillin (60% vs. 10%; $P=0.05$ ). Longer rinse times (2 min vs. 1 min) had no discernible effect. A 2-min rinse time resulted in a 55% incidence (6 of 11 patients) of bacteremia, whereas a 1-minute rinse time resulted in 50% incidence (2 of 4 patients) |
| Wahlmann et al. [164] | 59 patients with multiple-tooth extraction in preparation for radiotherapy of oral cancer                                | 1.5 g cefuroxime was administered intravenously 10 before start of multiple-tooth extractions  | At the start of the surgical procedure and 30 min later | Gram-positive cocci, mostly streptococci, were the predominant organisms, followed by Gram-negative rods; growth was mainly anaerobic. A total of 54 different strains of Gram-positive cocci were studied; 39 (74%) of these grew under aerobic and 14 (26%) under anaerobic conditions. Gram-negative bacteria were isolated from nine blood cultures   | There was no statistically significant difference in the occurrence of bacteremia and oral hygiene or periodontal status. It should be noticed that without antibiotic prophylaxis, patients with poor oral hygiene had a bacteremia rate of 100%, whereas bacteremia occurred in only 80% of the patients with adequate oral hygiene. This difference was not significant. The duration of the surgical procedure had no significant influence on bacteremia rates. The only other factor which had a significant influence on the rate of bacteremia was the number of extracted teeth                          |
| Brown et al. [24]     | 61 participants, aged 15–35 years, requiring the removal of third molars   | Intraoral suture removal 7 days after the extraction; 0.12% CHX preprocedural rinse (tests) or not (controls)  | Baseline and 90 s after suture removal                  | Facultative organisms, predominantly <i>Streptococcus</i> , were present in all specimens. Two samples yielded anaerobic growth of either <i>Prevotella</i> or <i>Peptostreptococcus</i>  | 10.9% of the patients developed bacteremia after the removal of sutures; the use of a preprocedural chlorhexidine rinse did not significantly reduce the incidence  |
| King et al. [81]      | 20 healthy patients who required extractions of at least five erupted teeth and placement of several sutures             | Removal of intraoral sutures Preoperatively, immediately after the extractions, before suture removal, and immediately following removal of the intraoral silk sutures   | Not known   | Fourteen of 16 patients yielded positive blood cultures following tooth extractions. One of 20 patients yielded a positive blood culture following suture removal. Even though the incidence of bacteremia following intraoral suture removal is relatively low (5%), this study suggests that intraoral suture removal is not a benign procedure for those persons who are considered high-risk cardiac patients |   |

**Table 2.4** Summary of studies on bacteremia associated with endodontic procedures

| Reference             | Subjects  | Dental procedure or oral activities evaluated  | Sampling time   | Identified species  | Outcomes  |
|-----------------------|---|--|---|---|---|
| Bender et al. [18]    | 50 patients (19–78 year) divided into two groups: Group A (26), in which endodontic manipulation was done only within the confines of the root canal, and Group B (24), in which the endodontic manipulation was purposely done beyond the confines of the root canal. Both groups were classified as to vital and nonvital pulps | Endodontic manipulation within or beyond the confines of the root canal  | Before, immediately after, and 10 min after endodontic manipulation   | The bacterium that was recovered in four instances was the <i>Streptococcus viridans</i> . Twice the <i>S. viridans</i> was isolated alone, and twice it was recovered mixed with diphtheroids  | Blood samples taken immediately after manipulation were positive in 6 out of 50 cases, an incidence of 12%. Blood samples taken 10 min later exhibited no growth. In Group A, in which instrumentation was done within the confines of the root canal, only one blood culture out of 26 cases was positive. In Group B, in which manipulation was performed beyond the apex of the root canal, six out of 24 cases, or 25%, were positive for growth in the blood culture flasks. They were positive in both vital and nonvital cases (three vital and three nonvital). The same organisms were isolated from root canals |
| Debelian et al. [39]  | 26 single-rooted teeth in 26 patients   | Endodontic therapy of teeth with asymptomatic apical periodontitis (Group 1: reamers were used to a level 2 mm beyond the apical foramen, Group 2: instrumentation ended inside the root canal 1 mm short of the apical foramen) | During the instrumentation and 10 min after the treatment was completed   | <i>Propionibacterium acnes</i> , <i>Pepostreptococcus prevoti</i> , <i>Fusobacterium nucleatum</i> , <i>Prevotella intermedia</i> , and <i>Saccharomyces cerevisiae</i> , <i>P. intermedia</i> , <i>Actinomyces israelii</i> , <i>Streptococcus intermedius</i> , and <i>Streptococcus sanguis</i>                          | In 7/13 (53.84%) patients of Group 1 and in 4/13 (30.77%) patients of Group 2 several species were recovered from the blood.  |
| Savarrio et al. [141] | 30 patients, mean age of 46 years   | Nonsurgical root-canal therapy   | Preoperatively, 5 min after access had been gained to the root-canal system, during instrumentation and 5 min after completion of the treatment | In 7 of 30 patients (23.3%), the same species of organism [ <i>Propionibacterium acnes</i> (2), <i>Streptococcus parasanguis</i> (2), <i>Actinomyces naeslundii</i> (1), <i>Prevotella buccae</i> (1) and <i>Streptococcus sanguis</i> (1)] was identified in the blood stream and in the sample from the root-canal system | Overall, a detectable bacteremia was present in 9 of 30 (30%) patients who had no positive preoperative control.  |

**Table 2.5** Summary of studies on bacteremia associated with orthodontic procedures

| Reference           | Subjects   | Dental procedure or oral activities evaluated  | Sampling time                         | Identified species   | Outcomes  |
|---------------------|--|--|---------------------------------------|--|---|
| Erverdi et al. [51] | 30 patients treated using the Edgewise technique   | Orthodontic debanding and debonding  | Before and after the removal of bands | <i>Streptococcus salivarius</i> and <i>S. sanguis</i> II-2 were identified in the preoperative blood samples, and <i>Streptococcus sanguis</i> I-3 and <i>S. mitis</i> I were identified in the postoperative samples. The numbers of <i>S. salivarius</i> and <i>S. sanguis</i> II-2 per mL of blood isolated from the preoperative samples were 3 colony forming units (CFU)/mL and 8 CFU/mL, respectively. The numbers of <i>S. sanguis</i> I-3 and <i>S. mitis</i> I per mL of blood isolated from the postoperative samples were 12 CFU/mL and 6 CFU/mL, respectively | A 6.6% bacteremia prevalence was observed in both preoperative and postoperative blood samples  |
| Erverdi et al. [50] | 80 young adult patients (banding group: 40 patients asked to rinse their mouth with CHX for 60 s just prior to fitting of the bands; debanding group: 40 patients asked to use the mouthwash immediately before removal of bands and brackets) | Orthodontic banding and debanding, following the application of a 0.2% CHX mouthwash | Pretreatment and posttreatment        | In the banding group, the microorganism isolated from the posttreatment blood sample was <i>Bacteroides oralis</i> with 4 colony forming units per mL of blood (CFU/mL). In the debanding group, <i>Staphylococcus aureus</i> was the species identified in the pretreatment blood sample. Posttreatment <i>Streptococcus sanguis</i> I-2 was identified as the causative agent with a quantity of 2 CFU/mL  | In the banding group, no bacteremia was detected in the pretreatment sample and 2.5% posttreatment bacteremia was detected in the posttreatment sample. In the debanding group, 2.5% bacteremia was found in both the pre- and posttreatment samples. The application of CHX mouthwash resulted in a decrease in the prevalence of bacteremia after banding and debanding, but the decrease was not statistically significant |

|                    |  |  |  |   |  |
|--------------------|--|--|--|---|--|
| Gürel et al. [65]  | 25 subjects (mean age, 14.4 years; range, 12.2–16.6 years) with Class I skeletal and dental relationships and bilateral crossbite who underwent rapid maxillary expansion  | Removal of a modified bonded rapid maxillary expansion appliance   | Baseline and the second 3 min after removal of the appliance | The bacteria isolated were <i>Streptococcus sanguinis</i> , <i>Streptococcus mutans</i> , <i>Streptococcus oralis</i> , <i>Staphylococcus hominis</i> , <i>Staphylococcus aureus</i> , <i>Kocuria rosea</i> , and <i>Micrococcus luteus</i> | Overt soft-tissue bleeding was observed in 11 of the 25 patients during appliance removal, and 8 of 25 patients showed bacteremia after appliance removal. No statistically significant relationship was found between overt bleeding and bacteremia incidence ( $P = 0.054$ )   |
| Lucas et al. [101] | 81 children undergoing general anesthesia (GA) for dento-alveolar surgery related to their orthodontic treatment were randomly allocated to the impression or separator group. A further 61 children, receiving orthodontic treatment allocated to the banding or archwire adjustment groups | Orthodontic treatment procedures: upper alginate impression, separator placement, band placement, and adjustment of an archwire on a fixed appliance | Before treatment and 30 s after procedure                    | 161 discrete colonies of 15 different species. <i>S.gordonii</i> , <i>S.sanguis</i> , <i>S.salivarius</i> , <i>S.vestibularis</i> , and coagulase-negative staphylococci were the most frequent isolates (69.9% of the colonies)            | There was no significant difference in the number of positive blood cultures between baseline (23%), and following an upper alginate impression (31%); between baseline (27%), and placement of a separator (36%); between baseline (36%), and fitting or placement of a band (44%); or between baseline (33%), and archwire adjustment (19.4%). For the separator group only the mean total number of aerobic and anaerobic bacteria combined, isolated from the blood samples (cfu of bacteria per ml of blood), was significantly greater following the placement of a separator ( $2.2 \pm 9.1$ ), compared with baseline ( $0.9 \pm 0.2$ ; $P < 0.02$ ). This investigation demonstrates that the only orthodontic treatment procedure that causes a significant bacteremia is the placement of a separator |

(continued)

**Table 2.5** (continued)

| Reference          | Subjects                         | Dental procedure or oral activities evaluated | Sampling time   | Identified species  | Outcomes  |
|--------------------|----------------------------------|---|---|---|---|
| Lucas et al. [100] | 49 children, mean age 15.4 years | Deband and gold chain adjustment              | Before treatment and 30 s after either upper dband or gold chain adjustment | The bacteria isolated following debanding and gold chain adjustment were similar to those following placement of separators and alginate impressions. These were coagulase negative staphylococci, <i>Micrococcus</i> spp., <i>Aerococcus</i> spp., and <i>Serratococcus mucilaginosus</i> . Bacteria isolated from the baseline blood samples included <i>Streptococcus</i> spp., <i>Corynebacterium</i> spp., coagulase-negative staphylococci, and <i>Micrococcus</i> spp. | There was no significant difference ( $P>0.05$ ) in the prevalence of bacteremia between baseline (19%) and following upper dband (26%) or between baseline (57%) and gold chain adjustment (57%). There was also no significant difference ( $P>0.05$ ) in the intensity of the anaerobic bacteremia between baseline and following deband or gold chain adjustment. Although the number of subjects undergoing gold chain adjustment was small, the findings demonstrate that neither upper debanding nor gold chain adjustment is associated with a significant bacteremia |
| Rosa et al. [139]  | 8 patients (18.5±3.9 years old)  | Removal of Haas palatal expanders             | Baseline and 3 min after removal of the apparatus                           | All strains isolated were <i>Streptococcus oralis</i> (negative for aesculin/arginine hydrolyses and amygdalin/arbutin/inulin/mannitol/sorbitol fermentation, and produced extracellular polysaccharide when exposed to glucose)  | Bottles containing blood taken before apparatus removal did not show bacterial growth. However, four of the eight postremoval blood samples showed turbidity after 5 days, and bacterioscopy analysis showed Gram-positive cocci  |

**Table 2.6** Summary of studies on bacteremia associated with miscellaneous oral procedures

| Reference          | Subjects   | Dental procedure or oral activities evaluated  | Sampling time   | Identified species                               | Outcomes   |
|--------------------|--|--|---|--|--|
| Flood et al. [53]  | 25 patients. In 13 patients, the abscesses were aspirated with a needle prior to incision and drainage, while needle aspiration was omitted in the remaining 12.   | Routine incision and drainage of dento-alveolar abscesses  | Immediately before the surgical procedures and at 1-min intervals for a period of 5 min after surgery | All three isolates were <i>Streptococcus</i> spp | Needle aspiration of pus resulted in a significant reduction ( $P < 0.05$ ) in the bacteremic episodes (0 out of 13) during subsequent surgery as compared with incision and drainage, without aspiration (3 out of 12). Bacteremia appeared to be transient, although, in one case it was detected at 5. It is concluded that bacteremic episodes occur during incision and drainage of dento-alveolar abscesses and this may be reduced by aspiration of the abscess contents prior to incision and drainage                   |
| Geerts et al. [57] | 67 subjects were periodontally examined and grouped according to their periodontal status. This classification was based on an original index of severity of periodontal disease (periodontal index for risk of infectiousness, PIRI) aimed at reflecting the individual risk of systemic injury from the periodontal niches. Thus, the patients were classified into 3 risk groups: low, PIRI=0, $n = 25$ ; moderate, $1 \leq \text{PIRI} \leq 5$ , $n = 27$ ; and high $6 \leq \text{PIRI} \leq 10$ , $n = 15$ . | Gentle mastication   | Before and 5 to 10 min after a standardized session of gentle mastication                             | Not given  | Overall, blood levels of endotoxin after mastication were found to be significantly higher than before mastication ( $0.89 \pm 3.3$ pg/ml vs. $3.0 \pm 5.8$ pg/ml; $P = 0.0002$ ). Likewise, the incidence of positive endotoxemia rose from 6% before mastication to 24% after mastication ( $P = 0.001$ ). When accounting for the PIRI index, endotoxin levels and positive endotoxemia proved to be significantly higher in patients with severe periodontal disease than in the subjects with low or moderate periodontitis |
| Hunter et al. [72] | 40 patients  | Dental prophylaxis by use of a commercially available air polishing device (cases) and by means of the conventional rubber cup and paste method (controls) | Before and 5 min after the procedure  | Mainly viridans group <i>streptococci</i>        | The likelihood of a bacteremia resulting from air polishing, in the absence of gingivitis, was less (15%) than in the group undergoing conventional prophylaxis (35%). Although a higher number of bacteremias was seen in the control group, the difference between the two groups was not statistically significant  |

(continued)

**Table 2.6** (continued)

| Reference            | Subjects   | Dental procedure or oral activities evaluated   | Sampling time  | Identified species  | Outcomes  |
|----------------------|--|---|--|---|---|
| Hurwitz et al. [73]  | 32 patients in good health and without signs or symptoms of systemic disease. Nineteen of the patients had generalized marginal gingival inflammation, with or without accompanying caries | Routine prophylaxis and fluoride application  | Within 6 min of completion of the prophylactic procedure | Not necessary   | All of the blood cultures obtained proved to be sterile   |
| Murphy et al. [113]  | 21 patients with untreated chronic periodontitis (32–75 years) and 20 with plaque-induced gingivitis (26–54 years)   | Chewing a standard wax medium for 4 min and 5-min post-chewing  | Before, during, and 5-min post-chewing                   | Skin contaminants ( <i>Staphylococcus epidermidis</i> , <i>Propionibacterium</i> spp.) were detected in blood samples from three patients (two periodontitis; one gingivitis) | No bacteremia of oral origin was detected in any patient  |
| Roberts et al. [135] | 735 anesthetized children aged 2–16 years  | Oral manipulative procedures; baseline dental and dental examination; toothbrushing, polishing, and scaling; intraligamental injection and nasotracheal tube; rubber dam placement, slow drill, fast drill, and matrix band placement; single extractions, multiple extractions, and mucoperiosteal flaps | 30 s after each of 13 dental operative procedures        | A total of 365 organisms were isolated. Among them, 212 (58%) were viridans streptococci  | All procedures were associated with a bacteremia. Percentage of positive blood cultures (%): Baseline 9.4, Dental examination 17.0, Toothbrushing 38.5, Polishing teeth 24.5, Scaling teeth 40.0, Intraligamental injection 96.6, Nasotracheal tube 9.7, Rubber dam placement 29.4, Slow drill 12.8, Fast drill 4.3, Matrix band placement 32.1, Single extraction 38.7, Multiple extractions 50.9 Mucoperiosteal flap 39.2, Cardiac patients 10.2. The procedure with the highest association was intraligamental injection (96.6%), and that with the lowest association was the fast drill 4.3%. The percentage of positive samples in nine of the groups was significantly greater than that in the baseline group. These nine groups were toothbrushing, polishing teeth, scaling teeth, intraligamental injection, rubber dam placement, matrix band placement, single extraction, multiple extractions, and the raising of a mucoperiosteal flap |

|                        |  |  |   |   |  |
|------------------------|--|--|---|---|--|
| Sonbol et al.<br>[147] | 205 children and adolescents undergoing general anesthesia for dental treatment                        | Conservative dental procedures; placement of rubber dam, use of the fast drill, use of the slow drill and placement of a matrix band and wedge | Baseline and 30 s after a single conservative procedure   | The most frequently isolated bacteria were <i>Streptococcus</i> spp. (56%), <i>Actinomyces</i> spp. (15%) and coagulase-negative <i>Staphylococcus</i> spp. (15%) | There was a significantly greater prevalence of bacteremia following placement of rubber dam (29% pre 54% post, $P = 0.01$ ) and placement of matrix band and wedge compared with baseline (32% pre 66% post, $P = 0.001$ ). A significantly greater number of CFU/ml were isolated anaerobically following both placement of rubber dam ( $P = 0.001$ ) and matrix band and wedge compared with baseline ( $P = 0.0001$ )                                     |
| Brennan et al.<br>[21] | 100 children (aged 1–8 years) receiving placebo or amoxicillin (50 mg/kg) 1 h before dental procedures | Dental restorative treatment   | 2 min after intubation (draw 1); after dental restorations, pulp therapy and cleaning (draw 2); 10 min later (draw 3); and five draws during and after dental extractions (draws 4–8) | Not given   | The incidence of bacteremia from draw 2 was 20% in the placebo group and 6% in the amoxicillin group ( $P = 0.07$ ). The incidence of bacteremia from draw 3 was 16% in the placebo group and 0% in the amoxicillin group ( $P = 0.03$ ). Subjects with higher gingival scores were significantly more likely to have a bacteremia for draw 2 ( $P = 0.01$ ). No other dental disease parameters had a statistically significant association with a bacteremia |

### **2.1.4 Duration of Bacteremia Associated with Dental Procedures and Oral Activities**

Bacteremia peaks during the first 2 min following tooth extraction or invasive dental procedure, and falls in time due to innate and adaptive defense mechanisms [104, 25, 123]. However, several studies have reported persistent bacteremia up to 60 min [15, 93, 136, 154]. From the cumulative data, it can be deduced that the bacteremic incidence peaks within the first few minutes and then gradually declines after 10–20 min. Furthermore, depending on the threshold sensitivity of the detection method, the total reduction in the bacteremic load may vary from 10% to 90% at 30 s and from 2% to 20% at 60 min. It is important, however, that in spite of an initial steep fall, a few bacteria survive in the circulation after a bacteremic challenge from the oral cavity. The role of these persisters and how they survive host defenses need to be evaluated further, as they may well be the ones that evade the initial host immune burst and have the propensity to seed target organs and cause systemic and distant infections [123].

### **2.1.5 Impact of Dental Disease on Bacteremia**

Although it is generally believed that there is a relationship between the risk of infective endocarditis and the extent of dental disease in the anatomical region of a dental procedure, the literature is controversial on this issue [97]. In a double-blind, randomized placebo-controlled study of 70 patients, the status of dental disease was compared with the incidence and nature of aerobic and anaerobic bacteremias following a single-tooth extraction and the antibacterial effect of rinses with chlorhexidine hydrochloride [96]. Although there was a wide range of severity of odontogenic disease, this did not correlate with results of blood cultures. Similar controversial results were also obtained related to the correlation between the gingival inflammation/periodontal status and bacteremia induced by chewing [54, 57, 113], toothbrushing [54], dental flossing [29], periodontal probing using a standard periodontal

probe [33, 35], full-mouth scaling, and root planning [54, 83, 103]. However, significant dental bacteremia occurs whether or not there is discernible bleeding at the site of the operative procedure. Bleeding is a poor predictor of odontogenic bacteremia [138].

### **2.1.6 Impact of Oral Hygiene on Bacteremia**

Although it has long been assumed that poor oral hygiene and dental disease are important risk factors for IE, [95] was the first to show a relationship between oral hygiene and gingival disease parameters and the risk of developing an IE-associated bacteremia after a routine daily event such as toothbrushing. One hundred and ninety-four participants in the study were in either a toothbrushing group or a single-tooth extraction with placebo group. The authors assessed the participants' oral hygiene, gingivitis, and periodontitis statuses. They assayed blood samples obtained before, during, and after the toothbrushing or extraction interventions for IE-associated bacteria. The authors found that all measures of oral hygiene (mean plaque score, plaque score of  $\geq 2$ , mean calculus score, calculus score of  $\geq 2$ ) and one measure of gingival bleeding (generalized bleeding with toothbrushing) were significantly associated with IE-related bacteremia. Participants with mean plaque or calculus scores of 2 or greater had 3.78- and 4.43-fold increased risks, respectively, of developing an IE-related bacteremia after toothbrushing ( $P < 0.01$ ). In addition, the presence of generalized bleeding after toothbrushing was associated with an almost eightfold risk of developing bacteremia caused by oral bacterial species implicated in IE. The authors further evaluated the impact of poor oral hygiene on the incidence of bacteremia by assessing the outcomes of participants in the toothbrushing group whose plaque and calculus scores were in the highest quartile. Nineteen of 98 participants in the toothbrushing group had both plaque and calculus scores that were in the highest quartile. The incidence of bacteremia after toothbrushing in this group was 42% ( $n = 8$ ), which was significantly greater than the incidence among participants whose scores were in the lower three quartiles (18%,  $P = 0.02$ ) [95].

We now have scientific evidence that good oral hygiene and gingival health are associated with a reduced risk of developing bacteremia, which may translate into a reduced risk of developing IE.

### **2.1.7 Impact of Type of Dental Procedure on Bacteremia**

It is generally felt that bacteremia is influenced by the invasiveness and the duration of a procedure and that dental extractions are the most likely of dental procedures to cause bacteremia, ranging from 10% to 94% in different studies (Tables 2.1–2.6). However, many studies are poorly controlled for invasiveness, the sequence of the surgical procedure, or the timing of the blood drawings in relation to the onset or completion of surgery. The invasiveness of procedures as a cause of distant site infection is therefore unclear, and little is known about the relative incidence, nature, magnitude, and duration of bacteremia from a wide variety of dental procedures [97].

### **2.1.8 Impact of Antibiotic/Antiseptic Therapy on Bacteremia**

Elimination or reduction of organisms associated with invasive dental procedures can occur either at the source (that is, the mouth) or after they enter the systemic circulation [97]. The efficacy of topical [26, 52, 130, 162, 165] or systemic antimicrobial prophylaxis was evaluated by several researchers [4, 21, 41, 66, 67, 93, 94].

Results are contradictory with regard to the efficacy of the use of topical antiseptics in reducing the frequency of bacteremia associated with dental procedures, but the preponderance of evidence suggests that there is no clear benefit. Topical antiseptic rinses do not penetrate beyond 3 mm into the periodontal pocket and therefore do not reach areas of ulcerated tissue where bacteria most often gain entrance to the circulation. On the basis of these data, it is unlikely that topical antiseptics are effective to significantly reduce the frequency, magnitude, and duration of bacteremia associated with a dental procedure [168].

### **2.1.9 Cumulative Risk over Time of Bacteremias from Routine Daily Activities Compared with the Bacteremia from a Dental Procedure**

Transient bacteremia is reported to occur frequently in the context of daily routine activities such as tooth-brushing, flossing, or chewing [15, 20, 29, 54, 68, 80, 94, 95, 98, 99, 123]. It therefore appears plausible that a large proportion of IE-causing bacteremia may derive from these daily routine activities [150]. In addition, in patients with poor dental health, bacteremia can be observed independently of dental procedures, and rates of post-procedural bacteremia are higher in this group [97, 152].

## **2.2 Prophylaxis of Infective Endocarditis**

IE is a rare but severe disease, with 100% mortality in the pre-antibiotic era. However, neither the incidence nor the mortality of the disease has decreased in the last 30 years. Despite major advances in both diagnostic and therapeutic procedures, this disease still carries a poor prognosis and a high mortality [110, 152]. The incidence of IE ranges from one country to another with 10,000 to 20,000 new cases diagnosed in the USA each year, while in France, infective endocarditis ranges from 25 to 30 cases/million inhabitants/year (about 1,500 cases/year) [36, 43].

IE is not a uniform disease, but presents in a variety of different forms, varying according to the initial clinical manifestation, the underlying cardiac disease (if any), the microorganism involved, the presence or absence of complications, and underlying patient characteristics. For this reason, IE requires a collaborative approach, involving primary care physicians, cardiologists, surgeons, microbiologists, infectious disease specialists, and frequently others, including neurologists, neurosurgeons, radiologists, and pathologists [152]. The epidemiology of infective endocarditis is complex to assess because diagnosis is difficult and referral bias has a large impact on the clinical characteristics of the population studied in different clinical settings [43].

### 2.2.1 Pathogenesis of IE

Infective endocarditis is a microbial infection of the endocardial surfaces usually involving the heart valves. Infection on a compromised endocardial surface (especially damaged or prosthetic heart valves) gives rise to the formation of vegetations. These vegetations proliferate and eventually can destroy the valves. Embolism of fragments of the vegetations can damage organs and tissues including the brain, lung, and coronary arteries [143].

The organisms most frequently responsible for infective endocarditis are those that have the greatest ability to adhere to damaged valves [109, 110]. Together, *Staph aureus*, *Streptococcus* spp, and enterococci are responsible for more than 80% of all instances of disease. These organisms have surface adhesins that mediate attachment to the vegetation. These adhesins are referred to as MSCRAMMs or microbial surface component reacting with adhesive matrix molecules [110, 124].

Most organ systems can be involved and the fact that IE may present to doctors in a variety of specialties means that they must be made aware of IE as a potential diagnosis warranting prompt specialist investigation and treatment. Guidelines for diagnosis and treatment are therefore important and worthy of widespread dissemination, such as those produced by the European Society of Cardiology: [www.escardio.org/knowledge/guidelines/Guidelines\\_Infective\\_Endocarditis.htm](http://www.escardio.org/knowledge/guidelines/Guidelines_Infective_Endocarditis.htm) [132].

### 2.2.2 Clinical Features of IE

Clinical features of IE are [132]:

- *Systemic features*: high remitting pyrexia, rigors, anorexia, weight loss, arthralgia, and fatigue [132]
- *Cardiac manifestations*: new or worsening cardiac murmurs – typically due to valvular regurgitation; or the development of cardiac failure. Abscesses of the heart and fistulous connections between cardiac structures are serious complications [132]
- *Extracardiac manifestations* consist of *embolic* as well as *vasculitic* phenomena. All major vessels may be the recipient of infected emboli from valve vegetations. Renal, splenic, and neurological complications may be particularly serious. Right-sided IE results in pulmonary infarcts and abscesses and

is often associated with i.v. drug abuse, infected pacemakers, or central i.v. lines. Culture-negative IE, including fungal IE, often has specific clinical features [132].

- The Duke criteria form the basis of the diagnosis of which blood cultures, echocardiography, and sometimes serology are most important [132].

#### Laboratory Findings

- Leukocytosis with neutrophilia
- Increased erythrocyte sedimentation rate
- Positive C-reactive protein
- Increased levels of serum immunoglobulins
- Blood cultures positive for pathogen
- Positive rheumatoid factor

#### Electrocardiogram Findings

- Prolonged PR interval (caused by abscess)
- Silent myocardial infarction (caused by emboli in coronary artery)

#### Echocardiography

- Vegetations
- Valvular perforations
- Other abnormalities (e.g., abscesses, pericarditis) [84]

### 2.2.3 Identification of At-Risk Patients for Occurrence or Mortality from Endocarditis

To reduce the risk of IE following dental procedures, prophylactic measures have been developed by experts in the fields of microbiology, epidemiology, cardiology, and dentistry. The principle preventive measure recommended is the use of prophylactic antibiotics before certain dental procedures in patients identified as at-risk (Table 2.7).

### 2.2.4 Identification of At-Risk Dental Procedures

The identification of at-risk procedures for infective endocarditis includes the consideration of different characteristics: (1) the number of identified

**Table 2.7** Guidelines for the identification of patients at “risk” who may require prophylaxis for infective endocarditis before dental procedures (Reprinted from [36] with permission from BMJ Publishing Group Ltd.; Reprinted from [59] with permission from Oxford University Press; Reprinted from [76] with permission from Elsevier; Reprinted from [116] with permission from NHS; Reprinted from [152] with permission from Oxford University Press; Reprinted and adapted from [168] with permission of the American Dental Association. (©2007 American Dental Association. Excerpted by JADA by permission of Circulation. Circulation 2007 ©2007 American Heart Association)

| Association   | Predisposing cardiac conditions   |
|---|---|
| French recommendations 2002   | <p><b>High risk (Group A)</b></p> <ul style="list-style-type: none"> <li>• Valvar prostheses (mechanical, homograft, or bioprostheses)</li> <li>• Nonoperated cyanotic congenital heart disease and pulmonary–systemic shunts</li> <li>• History of IE</li> </ul> <p><b>Lower risk of IE (Group B)</b></p> <ul style="list-style-type: none"> <li>• Valvar diseases: aortic insufficiency, mitral insufficiency, aortic stenosis</li> <li>• Mitral valve prolapse with mitral insufficiency or valve thickening</li> <li>• History of IE   Bicuspid aortic valve</li> <li>• Congenital non-cyanotic heart diseases except for atrial septal defect (cardiac condition without risk)</li> <li>• Obstructive hypertrophic cardiomyopathy (with murmur on auscultation)</li> </ul>   |
| British Society for Antimicrobial Chemotherapy recommendations 2006   | <ul style="list-style-type: none"> <li>• Previous infective endocarditis</li> <li>• Cardiac valve replacement surgery, i.e., mechanical or biological prosthetic valves</li> <li>• Surgically constructed systemic or pulmonary shunt or conduit</li> </ul>   |
| American Heart Association recommendations 2007, 2008   | <ul style="list-style-type: none"> <li>• Previous IE</li> <li>• Congenital heart disease (CHD)<sup>a</sup></li> <li>• Unrepaired cyanotic CHD, including palliative shunts and conduits</li> <li>• Completely repaired congenital heart defect with prosthetic material or device, whether placed by surgery or by catheter intervention, during the first 6 months after the procedure<sup>b</sup></li> <li>• Repaired CHD with residual defects at the site or adjacent to the site of a prosthetic patch or prosthetic device (which inhibit endothelialization)</li> <li>• Cardiac transplantation recipients who develop cardiac valvulopathy</li> <li>• Patients with prosthetic cardiac valves or prosthetic material used for cardiac valve repair</li> </ul>   |
| Australasian Society of Cardiac and Thoracic Surgeons and the Cardiac Society of Australia and New Zealand recommendations 2008 | <ul style="list-style-type: none"> <li>• Prosthetic cardiac valve or prosthetic material used for cardiac valve repair</li> <li>• Previous infective endocarditis</li> <li>• Congenital heart disease <i>but</i> only if it involves: <ul style="list-style-type: none"> <li>– Unrepaired cyanotic defects, including palliative shunts and conduits</li> <li>– Completely repaired defects with prosthetic material or devices, whether placed by surgery or catheter intervention, during the first 6 months after the procedure (after which the prosthetic material is likely to have been endothelialized)</li> <li>– Repaired defects with residual defects at or adjacent to the site of a prosthetic patch or device (which inhibit endothelialization)</li> </ul> </li> <li>• Cardiac transplantation with the subsequent development of cardiac valvulopathy</li> <li>• Rheumatic heart disease in Indigenous Australians only</li> </ul> |
| National Institute for Health and Clinical Excellence recommendations 2008  | <ul style="list-style-type: none"> <li>• Previous IE</li> <li>• Acquired valvular heart disease with stenosis or regurgitation</li> <li>• Valve replacement</li> <li>• Structural congenital heart disease, including surgically corrected or palliated structural conditions, but excluding isolated atrial septal defect, fully repaired ventricular septal defect or fully repaired patent ductus arteriosus, and closure devices that are judged to be endothelialized</li> <li>• Previous infective endocarditis</li> <li>• Hypertrophic cardiomyopathy</li> </ul>   |

(continued)

**Table 2.7** (continued)

| Association   | Predisposing cardiac conditions  |
|---|--|
| Task Force on the Prevention, Diagnosis, and Treatment of Infective Endocarditis of the European Society of Cardiology (ESC) recommendations 2009 | <p>Patients with the <b>highest risk</b> of infective endocarditis include three categories:</p> <ol style="list-style-type: none"> <li>1. Patients with a prosthetic valve or a prosthetic material used for cardiac valve repair</li> <li>2. Patients with previous IE</li> <li>3. Patients with congenital heart disease <ul style="list-style-type: none"> <li>(a) Cyanotic heart diseases, without surgical repair or with residual defects, palliative shunts, conduits</li> <li>(b) Congenital heart disease with complete repair with prosthetic material whether placed by surgery or by percutaneous technique, up to 6 months after the procedure</li> <li>(c) When a residual defect persists at the site of implantation or a prosthetic material or device by cardiac surgery or percutaneous technique</li> </ul> </li> </ol> <p>Antibiotic prophylaxis is no longer recommended in other forms of valvular or congenital heart disease</p> |

<sup>a</sup>Except for the conditions listed above, antibiotic prophylaxis is no longer recommended for any other form of CHD

<sup>b</sup>Prophylaxis is reasonable because endothelialization of prosthetic material occurs within 6 months after the procedure

infective-endocarditis-inducing pathogens (inoculum) colonizing the area that could enter the bloodstream in case of an invasive procedure; (2) the amount of the bleeding induced by the procedure; (3) the proportion of bacteria-positive blood cultures after a given procedure; (4) the magnitude of bacteremia after the procedure; (5) the duration of the bacteremia; and (6) the number of reported cases of infective endocarditis after a given procedure. However, there is wide variation in reported frequencies of these characteristics after procedures (Table 2.9) [43].

In general, prophylaxis is recommended for procedures associated with significant bleeding from hard or soft tissues, including periodontal surgery, scaling, and professional teeth cleaning, procedures with a high incidence of bacteremia (may occur in 70% or more patients) (Table 2.8). Dental procedures with a moderate incidence of bacteremia (may occur in 30% or more patients) should be considered for prophylaxis depending on the circumstances of the procedure and the periodontal condition (Table 2.10). Thus, for example, periodontal probing on a single healthy tooth would not justify antibiotic prophylaxis whereas full-mouth periodontal probing on a patient with periodontitis would.

**Infective Endocarditis Prophylaxis Expert Group. Prevention of endocarditis. 2008 update from Therapeutic Guidelines: Antibiotic version 13, and Therapeutic Guidelines: Oral and Dental version 1. Melbourne: Therapeutic Guidelines Limited, 2008.**

Prophylaxis is not recommended for procedures with a low incidence of bacteremia. Patient-performed oral hygiene activities, such as toothbrushing, flossing, or use of oral irrigators, can produce similar incidences of bacteremia as that caused by most dental procedures (excluding extractions and subgingival scaling/root planning). As these activities are performed more frequently, they have the potential to produce regular episodes of bacteremia, particularly in patients with gingival inflammation. It is considered that the cumulative effect of repeated episodes of bacteremia caused by oral hygiene activities is very likely to be a more important risk factor for infective endocarditis than isolated episodes of bacteremia occurring during dental visits, especially in patients with poor oral health and hygiene [74].

Good oral hygiene is probably the most important factor in reducing the risk of endocarditis in susceptible individuals, and access to high-quality dental care should be facilitated [36, 55, 152]. Once a patient is found to have a cardiac anomaly putting him or her at a risk of endocarditis, the patient should be referred to have their dental hygiene optimized. Similarly, a patient who has received an intracardiac prosthesis (valve, conduit, aortic graft) should be referred for dental assessment. Interventions ideally should be performed at least 14 days prior to surgery to allow mucosal healing. Those patients who undergo urgent or emergency valve replacement should have a dental assessment performed as soon as practicable after surgery, and a risk assessment performed to determine the

**Table 2.8** Dental procedures for which endocarditis prophylaxis is recommended for patients in Table 2.7. (Reprinted from [55] with permission from Elsevier; Reprinted from [59, 132] with permission from Oxford University Press; Reprinted from [76] with permission from Elsevier; Reprinted from [152] with permission from Oxford University Press; Reprinted from [168] with permission of the American Dental Association (©2007 American Dental Association. Excerpted by JADA by permission of Circulation. Circulation 2007. ©2007 American Heart Association.); Reprinted from [116], [www.nice.org.uk/CG64](http://www.nice.org.uk/CG64) with NHS permission

|   | Dental procedures  |
|---|--|
| French Society of Cardiology (FSC) recommendations 2002   | <p><b>In patients at high risk:</b> Prophylaxis is recommended in all invasive dental procedures</p> <p><b>In patients at low risk:</b> Prophylaxis is optional in invasive dental procedures, based on composite clinical assessment of the patient and procedural risk. Post hoc prophylaxis can be given in the event of unexpected procedural complexity</p> <p>Invasive dental procedures are:</p> <ul style="list-style-type: none"> <li><b>Setting up a sterile operating area</b></li> <li><b>Nonsurgical periodontal care</b> <ul style="list-style-type: none"> <li>• Scaling with or without polishing</li> <li>• Probing</li> </ul> </li> <li><b>Endodontic care: treating teeth with living pulpa</b></li> <li><b>Prosthetic procedures with a risk of bleeding</b></li> <li><b>Surgical procedures</b> <ul style="list-style-type: none"> <li>• Dental avulsion</li> <li>• Healthy tooth</li> <li>• Alveolectomy</li> <li>• Separation of roots</li> <li>• Impacted tooth or disimpaction</li> <li>• Germectomy</li> <li>• Frenectomy</li> <li>• Biopsy of accessory salivary glands</li> <li>• Bone surgery</li> </ul> </li> <li><b>Dentofacial orthopedics</b></li> <li><b>Inserting braces</b></li> </ul> |
| British Society for Antimicrobial Chemotherapy recommendations 2006   | All dental procedures involving dento-gingival manipulation  |
| American Heart Association (AHA) recommendations 2007, 2008   | Endocarditis prophylaxis is reasonable for patients with the highest risk of adverse outcomes who undergo dental procedures that involve manipulation of either gingival tissue or the periapical region of teeth or perforation of the oral mucosa (excluding local anesthetic injection through noninfected tissue)  |
| Australasian Society of Cardiac and Thoracic Surgeons and the Cardiac Society of Australia and New Zealand recommendations 2008 | <p><b>Prophylaxis Always Required</b></p> <ul style="list-style-type: none"> <li>• Extraction</li> <li>• Periodontal procedures, including surgery, subgingival scaling and root planning</li> <li>• Replanting avulsed teeth</li> <li>• Other surgical procedures (e.g., implant placement, apicoectomy)</li> </ul> <p><b>Prophylaxis required in some circumstances</b></p> <p><i>Consider prophylaxis for the following procedures if multiple procedures are being conducted, the procedure is prolonged, or periodontal disease is present:</i></p> <ul style="list-style-type: none"> <li>• Full periodontal probing for patients with periodontitis</li> <li>• Intraligamentary and intraosseous local anesthetic injection</li> <li>• Supragingival calculus removal/cleaning</li> <li>• Rubber dam placement with clamps (where risk of damaging gingiva)</li> </ul>  |

(continued)

**Table 2.8** (continued)

| Dental procedures   |  |
|---|--|
| National Institute for Health and Clinical Excellence recommendations 2008  | <ul style="list-style-type: none"> <li>• Restorative matrix band/strip placement</li> <li>• Endodontics beyond the apical foramen</li> <li>• Placement of orthodontic bands</li> <li>• Placement of interdental wedges</li> <li>• Subgingival placement of retraction cords, antibiotic fibers or antibiotic strips</li> </ul> |
| Task Force on the Prevention, Diagnosis, and Treatment of Infective Endocarditis of the European Society of Cardiology (ESC) recommendations 2009 | None   |

most appropriate plan for any remedial dental treatment. All elective dental procedures should ideally be delayed for at least 3 months postsurgery [59].

### **2.2.5 Recommended Prophylactic Regimens for Dental Procedures**

Providing prophylaxis for bacterial infective endocarditis (IE) prior to dental procedures has long been a topic of controversy because the disease has a low incidence but a relatively high mortality rate. Understanding which patients need prophylaxis and which antibiotics are appropriate is important for health care providers so that high-risk patients are treated appropriately [12, 41, 58, 86, 91, 106, 151].

The rationale for the use of antibiotic prophylaxis for surgical, including dental, manipulations is that the procedures cause bacteremia and the bacteremia may cause endocarditis. As a result, the antibiotics should be given to susceptible patients before the bacteremia is generated. These steps have been demonstrated in animal studies but it remains an unanswered question as to whether or not this reflects what occurs in humans [145].

Antibiotics may prevent endocarditis either by killing bacteria or by damaging them to an extent that the host defenses can then destroy them. Therefore, the antibiotic may work before the bacteria enter the bloodstream, after they enter the bloodstream, or on colonies of bacteria. The primary mechanism by which antibiotic prophylaxis could occur has not been established but a number of studies show that

bacteremia is reduced both in quantity and time in the presence of antibiotics. However, it is most likely that, as they may prevent bacterial adherence, antibiotics primarily work on bacterial colonies within the endocardium [145].

The AHA first introduced prophylaxis guidelines in 1955 and has updated them regularly since then. As randomized controlled human studies in this area have not been ethical until now, not to mention the difficulty involved in designing such a complex study, guidelines to date had to be empirical [129].

The AHA introduced these guidelines in 1955 because

- Prevention of IE is better than cure
- Certain cardiac conditions predispose to IE
- Bacteremia with IE-causing microorganisms occurs with dental procedures
- Antibiotic prophylaxis has been effective in preventing experimental IE in animals
- Antibiotic prophylaxis was thought to be effective in humans
- Case reports link dental procedures with the onset of IE
- The risk of adverse reactions with antibiotic use is low and
- Morbidity and mortality associated with IE is significant [129]

The American Heart Association (AHA) has proposed and updated guidelines in this area since 1955. The list of cardiac conditions, dental procedures, and antibiotic regimes has expanded since then. It is worth noting that in 1990 the AHA suggested that their recommendations

were to serve as guidelines and not as established standard of care. This was due to the potential medicolegal risks associated with IE prophylaxis. The 1997 guidelines also acknowledged that most cases of IE are not attributable to an invasive procedure but rather are the result of randomly occurring bacteremias from routine daily activities [129].

Numerous different guidelines on prophylactic regimes for infective endocarditis (IE) have been proposed, including those from the European Infectious Diseases Society consensus [88], New Zealand National Heart Foundation [46], Swiss Working Group for Endocarditis Prophylaxis [111], French Society of Cardiology recommendations 2002 [36], Netherlands Heart Foundation [159, 160], British Society for Antimicrobial Chemotherapy (BSAC) [59], German recommendations of the Deutschen Gesellschaft für Kardiologie und der Paul Ehrlich Gesellschaft für Chemotherapie [114], American Heart Association (AHA) [168], Australian Prevention of Endocarditis Guidelines, Australasian Society of Cardiac and Thoracic Surgeons and the Cardiac Society of Australia and New Zealand [76], National Institute for Clinical Excellence (NICE) [116], Antibiotique therapy group, University of Leuven [38] and European Society of Cardiology [152]. A total of 17/20 Swedish counties had recommendations developed by the pharmaceutical committees for the use of antibiotic prophylaxis. Three counties did not have recommendations concerning the use of antibiotic prophylaxis in oral healthcare. Thirteen counties used a joint recommendation and four counties had their own recommendation, which made a total of five documents that were evaluated in this study. The recommendations were current during 2008. Of these, two were updated in 2008, one in 2007, one in 2005 and one in 1999 [45].

### 2.2.5.1 2002: French Society of Cardiology (Société Française De Cardiologie SFC) Recommendations

The French guidelines published in 2002 challenged accepted practice, emphasizing the importance of general and oral hygiene in populations at risk and suggesting restriction of prophylaxis to patients with the highest ratio of benefit to individual and collective risk. The full recommendations are available at [www.infectiologie.com](http://www.infectiologie.com) [40, 128].

The French working group stated that general hygiene measures are the most important. They aim at decreasing the risk of bacteraemia. They include oral, dental, and skin hygiene to prevent any rupture of the skin or mucosal barriers, disinfection of wounds, curative antibiotic treatment of infection, and strict compliance with asepsis when performing procedures at risk of infection. A systematic surveillance of the oral and dental state is mandatory at least twice a year for patients with heart diseases. In patients at risk (both high and low), it is recommended that local chlorhexidine-based antiseptics be used as mouthwash for 30 s before dental procedures and that dental care be provided in as few sessions as possible. If several sessions are required, and if the practitioner uses antibiotic prophylaxis, the sessions must be scheduled at least 10 days apart if possible [36].

The working group suggested:

- to maintain the principle of antibiotic prophylaxis when performing procedures in patients at risk with cardiac conditions, but
- to limit its indications to cases that have the highest ratio of individual benefit to individual and collective risk [36, 55].

In patients with high risk, using antibiotic prophylaxis according to the rules described in Table 2.11 is recommended for non-contraindicated invasive oral or dental procedures. In patients with cardiac conditions conferring lower risk of IE, antibiotic prophylaxis is optional. Antibiotic prophylaxis must be chosen by health-care professionals, taking into account the nature of the procedures and the patient's general condition (Tables 2.9 and 2.10). Whatever the choice, it must be made after informing the patient and obtaining his or her consent to the proposed strategy. Each patient should be given a follow-up leaflet where the proposed strategy has to be reported. Patients should know that in case of fever or symptoms, especially in the month following dental procedures, they must consult a physician as soon as possible before starting any drug and inform the physician of their dental history so that blood cultures can be made before initiating any antibiotic [36].

In both groups of cardiac patients, with low and high risk of infective endocarditis, some procedures are contraindicated, such as:

- Intraligamentous local anesthesia
- Endodontic care: treatment of teeth with devitalized pulp, including further surgical pulp canal treatment

**Table 2.9** Dental procedures not requiring prophylaxis for any cardiac category (Reprinted from [55] with permission from Elsevier; Reprinted from [59, 132] with permission from Oxford University Press; Reprinted from [76] with permission from Elsevier; Reprinted from [152] with permission from Oxford University Press; Reprinted from [168] with permission of the American Dental Association (©2007 American Dental Association. Excerpted by JADA by permission of Circulation. Circulation 2007. ©2007 American Heart Association.); Reprinted from [116], [www.nice.org.uk/CG64](http://www.nice.org.uk/CG64) with NHS permission

|   | Dental procedures not requiring prophylaxis   |
|---|---|
| French recommendations 2002   | <b>Noninvasive buccodental procedure (without important risk of bleeding):</b> <ul style="list-style-type: none"> <li>• Preventive procedures</li> <li>• Application of fluor</li> <li>• Coronal sealing</li> <li>• Conservative procedures (coronal restoration)</li> <li>• Non-bleeding prosthetic procedures (impression)</li> <li>• Postoperative removal of sutures</li> <li>• Inserting removable orthodontic prostheses</li> <li>• Inserting or adjusting orthodontic devices</li> <li>• Dental X-rays</li> <li>• Non-intraligamentous local anesthesia</li> </ul> |
| British Society for Antimicrobial Chemotherapy recommendations 2006   | <b>Not given</b>  |
| American Heart Association recommendations 2007, 2008   | <ul style="list-style-type: none"> <li>• Routine anesthetic injections through noninfected tissue</li> <li>• Taking dental radiographs</li> <li>• Placement of removable prosthodontic or orthodontic appliances</li> <li>• Adjustment of orthodontic appliances</li> <li>• Placement of orthodontic brackets</li> <li>• Shedding of primary teeth</li> <li>• Bleeding from trauma to the lips or oral mucosa</li> </ul>  |
| Australasian Society of Cardiac and Thoracic Surgeons and the Cardiac Society of Australia and New Zealand recommendations 2008                   | <ul style="list-style-type: none"> <li>• Oral examination</li> <li>• Infiltration and block local anesthetic injection</li> <li>• Restorative dentistry</li> <li>• Supragingival rubber dam clamping and placement of rubber dam</li> <li>• Intracanal endodontic procedures</li> <li>• Removal of sutures</li> <li>• Impressions and construction of dentures</li> <li>• Orthodontic bracket placement and adjustment of fixed appliances</li> <li>• Application of gels</li> <li>• Intraoral radiographs</li> <li>• Supragingival plaque removal</li> </ul>             |
| National Institute for Health and Clinical Excellence recommendations 2008  | All dental procedures   |
| Task Force on the Prevention, Diagnosis, and Treatment of Infective Endocarditis of the European Society of Cardiology (ESC) recommendations 2009 | <ul style="list-style-type: none"> <li>• Local anesthetic injections in noninfected tissue</li> <li>• Removal of the sutures</li> <li>• Dental X-rays</li> <li>• Placement or adjustment of removable prosthodontic or orthodontic appliances or brace</li> <li>• Shedding of deciduous teeth</li> <li>• Trauma to the lips and oral mucosa</li> </ul>  |

**Table 2.10** Factors that may help in choosing whether antibiotic prophylaxis will be prescribed when prophylaxis is optional (Reproduced from [36]. With permission from BMJ Publishing Group Ltd)

| Arguments for prescription   |  |
|--|--|
| Age >65 years  |  |
| Associated conditions  |  |
| Cardiac, renal, respiratory, and hepatic insufficiency   |  |
| Diabetes mellitus  |  |
| Acquired, constitutional or therapeutic (corticosteroids, immunosuppressive agents) immunodepression |  |
| Oral or dental condition   |  |
| Inadequate oral or especially dental hygiene   |  |
| Procedure  |  |
| Important bleeding (intensity, duration)   |  |
| Technically difficult procedure (prolonged procedure)  |  |
| Patient's opinion after receiving information  |  |
| Arguments against prescription   |  |
| Allergy to several antibiotics   |  |
| Patient's opinion after receiving information  |  |

- Surgical procedures:
  - Radicular amputation
  - Transplantation/reimplantation
  - Periapical surgery
  - Periodontal surgery
  - Implant surgery
  - Inserting filling material

- Dentofacial orthopedics

- Preorthodontal surgery of impacted teeth

Pulp diseases, periodontal diseases, and trauma require extraction [55].

For other cardiac conditions, antibiotic prophylaxis is not recommended. Before valve surgery, antibiotic prophylaxis is indicated as for high-risk group patients. A complete radiological dental assessment must be made; only pulped teeth or teeth that have had a perfect endodontic treatment (more than a year before) without periodontal enlargement and with a healthy periodontium are kept. Pulpectomized teeth with incomplete endodontic treatment and teeth presenting with periodontal lesions and persisting roots and apex are extracted at least 15 days before cardiac surgery. In case of emergency surgery, dental care is given as soon as possible according to the context [36].

Modalities of antibiotic prophylaxis recommended by the French working group are given in Table 2.11.

In the surgical dentistry and maxillo-facial surgery with opening oropharyngeal (mainly neoplastic surgery) the risk of infection is high (about 30% of patients). The duration of the antibiotic prophylaxis should not exceed 48 h, as demonstrated by studies methodologically correct. Positioned beyond this time, it is an antibiotic cure [56].

**Table 2.11** Antibiotic prophylaxis for IE in dental care and upper respiratory tract procedures: administration of antibiotics respecting contraindications and usual conditions of use and surveillance (French Society of Cardiology recommendations (Reproduced from [36]. With permission from BMJ Publishing Group Ltd)

| Ambulatory care <sup>a</sup>    | Antibiotic   | Dosage and route of administration             |                                  |
|---------------------------------|--|--|----------------------------------|
|                                 |  | Single dose in the hour before the procedure   |                                  |
| No allergy to β lactams         | Amoxicillin  | 3 g orally <sup>c</sup>                        |                                  |
| Allergy to β lactams            | Pristinamycin <sup>b</sup> or Clindamycin <sup>b</sup> | 1 g orally<br>600 mg orally                    |                                  |
| General anesthesia <sup>d</sup> |  | Before (in the hour before the procedure)      | After (6 h later)                |
| No allergy to β lactams         | Amoxicillin  | 2 g iv (infusion 30 min)                       | 1 g orally                       |
| Allergy to β lactams            | Vancomycin or Teicoplanin                              | 1 g iv (infusion ≥60 min)<br>400 mg iv (bolus) | No second dose<br>No second dose |

<sup>a</sup>Oral pediatric dosages: amoxicillin 75 mg/kg; clindamycin 15 mg/kg; pristinamycin 25 mg/kg

<sup>b</sup>2 g orally if the patient's weight is <60 kg

<sup>c</sup>The respective percentage of streptococci strains with a decreased susceptibility to these two antibiotics must be taken into account for the choice

<sup>d</sup>Pediatric dosages: amoxicillin 50 mg/kg intravenously (iv) before, 25 mg/kg orally 6 h later; vancomycin 20 mg/kg (maximum 1 g); teicoplanin: no official approval for antibiotic prophylaxis in children

### **2.2.5.2 2004: The British Cardiac Society Medical Practice Committee and the Royal College of Physicians (RCP) Clinical Effectiveness and Evaluation Unit Recommendations**

These guidelines were developed in accordance with the principles laid down by the AGREE (Appraisal of Guidelines for Research and Evaluation Instrument) Collaboration ([www.agreecollaboration.org/](http://www.agreecollaboration.org/)). The guidelines are presented in more detail, with a full reference list, on the Royal College of Physicians (RCP) website: [www.rcplondon.ac.uk](http://www.rcplondon.ac.uk). An extensive referenced document, which includes also the list of dental and “surgical” procedures requiring antibiotic prophylaxis is also available on the British Cardiac Society’s web site: [www.bcs.com](http://www.bcs.com) ([http://www.bcs.com/documents/restricted/registered/affiliates/endocarditis\\_guidance.pdf?Submit=Download](http://www.bcs.com/documents/restricted/registered/affiliates/endocarditis_guidance.pdf?Submit=Download)) Accessed on 17.05.2010)

In order to prevent IE, prophylactic antibiotic treatment is recommended prior to dental or other “surgical” procedures for those at moderate or high risk of developing IE, should bacteremia be induced.

1. Patients should be informed of their risk of IE and the need for antibiotic prophylaxis, and be told to inform any doctor or dentist who is responsible for providing care. They should be given a card to carry indicating the type of cardiac lesion, the risk, and how to avoid IE.
2. Patients at moderate risk or high risk of IE should be given antibiotic prophylaxis with appropriate antibiotics based upon the type of dental or surgical procedure being performed.

### **2.2.5.3 2006: British Society for Antimicrobial Chemotherapy Recommendations**

Despite the lack of evidence of the benefit for prophylactic antibiotics to prevent endocarditis associated with dental procedures, the Working Party of the British Society for Antimicrobial Chemotherapy considered that many clinicians would be reluctant to accept the radical, but logical, step of withholding antibiotic prophylaxis for dental procedures. It was therefore agreed to compromise and recommended prophylaxis only for

those patients in whom the risk of developing endocarditis is high and, if infected, would carry a particularly high mortality. Thus the indication for antibiotic prophylaxis for all dental treatment procedures involving dento-gingival manipulation or endodontics was restricted to patients who have a history of previous endocarditis, or who have had cardiac valve replacement surgery, or those with a surgically constructed systemic or pulmonary shunt or conduit [59] (Tables 3.7–3.9).

For high-risk patients ≥ 10 years of age the Working Party recommended a single 3 g oral dose of amoxicillin (<5 years of age: 750 mg; ≥ 5 to <10 years of age: 1.5 g) to be given 1 h prior to the procedure, whether the procedure is performed using a general or a local anesthetic [59] (Table 2.12).

For i.v. administration, the British Society for Antimicrobial Chemotherapy (2006) recommended a single dose of 1 g amoxicillin for patients ≥ 10 years of age (<5 years of age: 250 mg; ≥ 5 to <10 years of age: 500 mg), given just before the procedure or at induction of anesthesia. If the patient (≥ 10 years of age) has a documented penicillin allergy, a single dose of oral 600 mg clindamycin (<5 years of age: 150 mg; ≥ 5 to <10 years of age: 300 mg) should be given 1 h before the procedure. For i.v. administration, a single dose of 300 mg clindamycin (given over at least 10 min) (<5 years of age: 75 mg; ≥ 5 to <10 years of age: 150 mg) was recommended. For those patients who are allergic to penicillin and cannot swallow capsules, oral azithromycin suspension (≥ 10 years: 500 mg; <5 years of age: 200 mg; ≥ 5 to <10 years of age: 300 mg) given 1 h before the procedure can be used as an alternative [59].

In addition, where practicable, a preoperative mouthwash of chlorhexidine gluconate (0.2%) should be administered and held in the mouth for 1 min [59].

For patients requiring sequential dental procedures, these should ideally be performed at intervals of at least 14 days to allow healing of oral mucosal surfaces. If further dental procedures cannot be delayed, British Society for Antimicrobial Chemotherapy (2006) suggested alternating amoxicillin and clindamycin. In this scenario if the patient has a penicillin allergy, it was suggested that expert advice to be sought [59].

The Working Party acknowledged that the change in guidance may result in patient or carer concern. The

**Table 2.12** Recommendations for infective endocarditis prophylaxis regimen by the British Society for Antimicrobial Chemotherapy (Reprinted from [59]. With permission from Oxford University Press)

| Population   | Age                                |                                    |                                   | Timing of dose  |
|--|------------------------------------|------------------------------------|-----------------------------------|---|
|  | >10 years                          | 5–10 years                         | <5 years before procedure         |   |
| General  | Amoxicillin 3 g po                 | Amoxicillin 1.5 g po               | Amoxicillin 750 mg                | 1 h   |
| Allergic to penicillin                                   | Clindamycin 600 mg po              | Clindamycin 300 mg po              | Clindamycin 150 mg po             | 1 h   |
| Allergic to penicillin and unable to swallow capsules    | Azithromycin 500 mg po             | Azithromycin 300 mg po             | Azithromycin 200 mg po            | 1 h   |
| Intravenous regimen expedient                            | Amoxicillin 1 g iv                 | Amoxicillin 500 mg iv              | Amoxicillin 250 mg iv             | Just before the procedure or at induction of anesthesia |
| Intravenous regimen expedient and allergic to penicillin | Clindamycin 300 mg iv <sup>a</sup> | Clindamycin 150 mg iv <sup>a</sup> | Clindamycin 75 mg iv <sup>a</sup> | Just before the procedure or at induction of anesthesia |

Where a course of treatment involves several visits, the antibiotic regimen should alternate between amoxicillin and clindamycin  
Preoperative mouth rinse with chlorhexidine gluconate 0.2% (10 mL for 1 min)

*Po* oral, *iv* Intravenous

<sup>a</sup>Given over at least 10 min

added Appendix contains a patient information sheet, which may be helpful for dental professionals when they are explaining these changes:

“Prevention of Infective Endocarditis Guidelines Information for Patients and Parents February 2006. A BSAC group of experts has spent a lot of time carefully looking at whether dental treatment procedures are a possible cause of infective endocarditis (IE) [sometimes called bacterial endocarditis (BE)], which is infection of the heart valve. After a very detailed analysis of all the available evidence they have concluded that there is no evidence that dental treatment procedures increase the risk of these infections. Therefore it is recommended that the current practice of giving patients antibiotics before dental treatment be stopped for all patients with cardiac abnormalities, except for those who have a history of healed IE, prosthetic heart valves and surgically constructed conduits. The main reasons for this are the lack of any supporting evidence that dental treatment leads to IE and the increasing worry that administration of antibiotics may lead to other serious complications such as anaphylaxis (severe allergy) or antibiotic resistance. The advice from the BSAC is that patients should concentrate on achieving and keeping a high standard of oral and dental health, as this does reduce the risk of endocarditis. Help for this will be

provided by your Dental Professional. British Society for Antimicrobial Chemotherapy, 2 February 2006.”

#### 2.2.5.4 2007 American Heart Association (AHA) Rheumatic Fever, Endocarditis, and Kawasaki Disease Committee, Council on Cardiovascular Disease in the Young, and the Council on Clinical Cardiology, Council on Cardiovascular Surgery and Anesthesia, and the Quality of Care and Outcomes Research Interdisciplinary Working Group Recommendations

The AHA has made recommendations for the prevention of IE for more than 50 years. The 2007 recommendations are the 10th version issued by AHA.

The AHA 2007 guidelines recommended antimicrobial prophylaxis to prevent IE in patients with underlying cardiac conditions who underwent bacteremia-producing procedures on the basis of the following factors: (1) bacteremia causes endocarditis; (2) viridans group streptococci are part of the normal oral flora, and enterococci are part of the normal gastrointestinal or genitourinary tract flora; (3) these microorganisms were usually susceptible to antibiotics recommended

for prophylaxis; (4) antibiotic prophylaxis prevents viridans group streptococcal or enterococcal experimental endocarditis in animals; (5) a large number of poorly documented case reports implicated a dental procedure as a cause of IE; (6) in some cases, there was a temporal relationship between a dental procedure and the onset of symptoms of IE; (7) an awareness of bacteremia caused by viridans group streptococci associated with a dental procedure exists; (8) the risk of significant adverse reactions to an antibiotic is low in an individual patient; and (9) morbidity and mortality from IE are high. Most of these factors remain valid, but collectively, they do not compensate for the lack of published data that demonstrate a benefit from prophylaxis, as a placebo-controlled, multicenter, randomized, double-blinded study to evaluate the efficacy of IE prophylaxis in patients who undergo a dental procedure has not been done. Such a study would require a large number of patients per treatment group and standardization of the specific invasive procedures and the patient populations [168].

Previous AHA guidelines categorized underlying cardiac conditions associated with the risk of IE as those with high risk, moderate risk, and negligible risk, and recommended prophylaxis for patients in the high- and moderate-risk categories. For the 2007 guidelines on prevention of IE, the Committee considered three distinct issues: (1) What underlying cardiac conditions over a lifetime have the highest predisposition to the acquisition of endocarditis? (2) What underlying cardiac conditions are associated with the highest risk of adverse outcome from endocarditis? (3) Should recommendations for IE prophylaxis be based on either or both of these two conditions? Only for patients with risk of acquisition of IE and highest risk of adverse outcome from IE, is IE prophylaxis for dental procedures reasonable? [168] (Table 2.7).

Dental procedures for which antibiotic prophylaxis is indicated are all dental procedures that involve the gingival tissues or periapical region of a tooth and for those procedures that perforate the oral mucosa (Table 2.8). Although IE prophylaxis is reasonable for these patients, its effectiveness is unknown. This includes procedures such as biopsies, suture removal, and placement of orthodontic bands, but it does not include routine anesthetic injections through noninfected tissue, the taking of dental radiographs, placement of removable prosthodontic or orthodontic appliances, placement of orthodontic brackets, or

adjustment of orthodontic appliances. Finally, there are other events that are not dental procedures and for which prophylaxis is not recommended, such as shedding of deciduous teeth and trauma to the lips and oral mucosa [168].

An antibiotic for prophylaxis should be administered in a single dose before the procedure. If the dosage of antibiotic is *inadvertently* not administered before the procedure, the dosage may be administered up to 2 h after the procedure (Table 2.13) [168].

Patients receiving parenteral antibiotic therapy for IE may require dental procedures during antimicrobial therapy, particularly if subsequent cardiac valve replacement surgery is anticipated. In these cases, the parenteral antibiotic therapy for IE should be continued and the timing of the dosage adjusted to be administered 30–60 min before the dental procedure. This parenteral antimicrobial therapy is administered in such high doses that the high concentration would overcome any possible low-level resistance developed among mouth flora (unlike the concentration that would occur after oral administration) [168].

#### **2.2.5.5 2008: American College of Cardiology/American Heart Association (ACC/AHA) Task Force on Practice Guidelines Updates Valvular Heart Disease: Focused Update on Infective Endocarditis**

In August 2008, the American College of Cardiology (ACC) and the AHA published a “focused update” on IE prophylaxis [118]. Previously, the writing of recommendations could last 3 years. Since 2006, the ACC and the AHA have settled a faster update procedure. New data are reviewed twice a year. However, the text does not cite the recommendations by NICE that were published in April 2008 [40].

#### **2.2.5.6 2008: Australasian Society of Cardiac and Thoracic Surgeons and the Cardiac Society of Australia and New Zealand Recommendations**

The Australian guidelines for the use of antibiotics to prevent infective endocarditis have been revised recently and published by Therapeutic Guidelines,

**Table 2.13** American Heart Association regimens for infective endocarditis prophylaxis (Reprinted from [168] Adapted 2010 with permission of the American Dental Association. ©2007 American Dental Association. Excerpted by JADA by permission of Circulation. Circulation 2007. ©2007 American Heart Association.)

| Patient group   | Route                         | Antibiotic   | Regimen: Single Dose 30–60 min before procedure |                                  |
|---|-------------------------------|--|---|----------------------------------|
|   |                               |  | Adults  | Children                         |
| Standard general prophylaxis for patients at risk                         | Oral                          | Amoxicillin  | 2 g   | 50 mg/kg                         |
| Unable to take oral medication  | Intramuscular and intravenous | Ampicillin<br>OR<br>Cefazolin or ceftriaxone   | 2 g<br>1 g                                      | 50 mg/kg<br>50 mg/kg             |
| Allergic to penicillins or ampicillin                                     | Oral                          | Cephalexin <sup>a,b</sup><br>OR<br>Clindamycin<br>OR<br>Azithromycin or clarithromycin | 2 g<br>600 mg<br>500 mg                         | 50 mg/kg<br>20 mg/kg<br>15 mg/kg |
| Allergic to penicillins or ampicillin and unable to take oral medications | Intramuscular and intravenous | Cefazolin or ceftriaxone<br>OR<br>Clindamycin  | 1 g<br>600 mg                                   | 50 mg/kg<br>20 mg/kg             |

<sup>a</sup>Or other first- or second-generation oral cephalosporin in equivalent adult or pediatric dosage

<sup>b</sup>Cephalosporins should not be used in a person with a history of anaphylaxis, angioedema, or urticaria with penicillins or ampicillin

Prevention of Endocarditis. (Infective Endocarditis Prophylaxis Expert Group [74]). The Australian guidelines follow the lead of the US guidelines, reducing the categories of patients for whom prophylaxis is recommended [168]. Consideration was given to deleting the moderate risk group of dental procedures (may occur in 30% or more cases) [34]. Antibiotic prophylaxis is now recommended only for those patients with cardiac conditions associated with the highest risk of adverse outcomes from endocarditis, rather than those patients with an increased lifetime risk of infective endocarditis [76].

Antibiotic prophylaxis is recommended only in those patients with the highest risk of *adverse outcomes* from endocarditis. Standard prophylaxis is *amoxicillin 2 g orally 1 h before the procedure*. It should be noted that antibiotic prophylaxis is no longer recommended in patients with other forms of valvular or structural heart disease, including mitral valve prolapse [74, 76].

An important difference between the Australian and American guidelines is the description of dental procedures requiring antibiotic prophylaxis. In the American guidelines, the dental procedures for which IE prophylaxis is recommended are “Dental procedures that

involve manipulation of gingival tissue or the periapical region of teeth or perforation of the oral mucosa.” [168]. “Manipulation of gingival tissue” is a confusing description and therefore the new Australian guidelines have sought to identify which dental procedures are likely to have a high incidence of bacteremia (may occur in 70% or more of cases). However, in a lengthy appointment in which multiple treatments are being performed, particularly in a patient with periodontal disease, consideration must be given to providing antibiotic prophylaxis [34] (Table 2.8). If possible, it is preferable to structure appointments for patients requiring antibiotic prophylaxis so that multiple treatments are performed at one sitting, thus avoiding the need for repeated visits under antibiotic prophylaxis [34].

Dental procedures for which antibiotic prophylaxis is not required are shown in Table 2.9 [74, 76].

If, after evaluation of both the cardiac condition and the dental procedure, antibiotic prophylaxis is considered necessary, a single dose of antibiotic should be given before the procedure. Follow-up doses are not recommended. Standard prophylaxis is amoxicillin 2 g orally 1 h before the procedure. For alternatives (penicillin-sensitive patients, etc.), refer to Table 2.14 [74, 76].

**Table 2.14** Recommendations for infective endocarditis prophylaxis regimen by the The Task Force on the Prevention, Diagnosis, and Treatment of Infective Endocarditis of the European Society of Cardiology (ESC) (2009) (Reprinted with permission from Oxford University Press)

| Situation                              | Antibiotic                              | Single dose 30–60 min before procedure |                   |
|--|---|--|-------------------|
|  |   | Adults                                 | Children          |
| No allergy to penicillin or ampicillin | Amoxicillin or ampicillina <sup>a</sup> | 2 g po or iv                           | 50 mg/kg po or iv |
| Allergy to penicillin or ampicillin    | Clindamycin                             | 600 mg po or iv                        | 20 mg/kg po or iv |

Cephalosporins should not be used in patients with anaphylaxis, angioedema, or urticaria after intake of penicillin and ampicillin

<sup>a</sup>Alternatively cephalexin 2 g i.v. or 50/kg i.v. for children, cefazolin or ceftriaxone 1 g i.v. for adults or 50/kg i.v. for children

### 2.2.5.7 2008: National Institute for Health and Clinical Excellence (NICE) Recommendations

Recently, NICE, an independent organization in charge of issuing recommendations on health, published recommendations on the prevention of IE. In summary, this guideline recommends that antibiotic prophylaxis to prevent infective endocarditis should not be given to adults and children with structural cardiac defects at risk of infective endocarditis undergoing dental and non-dental interventional procedures. The basis for this recommendation is [148].

- There is no consistent association between having an interventional procedure, dental or non-dental, and the development of infective endocarditis.
- Regular toothbrushing almost certainly presents a greater risk of infective endocarditis than a single dental procedure because of repetitive exposure to bacteremia with oral flora.
- The clinical effectiveness of antibiotic prophylaxis is not proved.
- Antibiotic prophylaxis against infective endocarditis for dental procedures is not cost-effective and may lead to a greater number of deaths through fatal anaphylaxis than a strategy of no-antibiotic prophylaxis.

In addition, given the difficulties of defining relative risk, a simple classification of conditions into groups at risk and those considered not to be at any risk greater than the general population was undertaken.

The detailed consideration of the evidence for this guideline is available in the full version (<http://www.nice.org.uk/CG064>, accessed 28 April 2010).

A systematic review of the literature was undertaken and a health economic model for dental antibiotic prophylaxis was developed [148].

Reference [116] (Available from [www.nice.org.uk/CG064](http://www.nice.org.uk/CG064) Reproduced with permission) recommended that health care professionals should offer people at risk of infective endocarditis clear and consistent information about prevention, including:

- The benefits and risks of antibiotic prophylaxis, and an explanation of why antibiotic prophylaxis is no longer routinely recommended
- The importance of maintaining good oral health
- Symptoms that may indicate infective endocarditis and when to seek expert advice

Antibiotic prophylaxis against infective endocarditis was not recommended for people undergoing dental procedures. Moreover, they suggested that Chlorhexidine mouthwash should not be offered as prophylaxis against infective endocarditis to people at risk of infective endocarditis undergoing dental procedures.

However, NICE recommended that any episodes of infection in people at risk of infective endocarditis should be investigated and treated promptly to reduce the risk of endocarditis developing.

### 2.2.5.8 2009: Task Force on the Prevention, Diagnosis, and Treatment of Infective Endocarditis of the European Society of Cardiology (ESC) Recommendations

Reviewing the recent guideline committees of national cardiovascular societies that have reevaluated the existing scientific evidence in this field [36, 59, 114, 168], it was revealed that although the individual recommendations of these committees differ in some aspects, they did uniformly and independently draw four conclusions:

- The existing evidence does not support the extensive use of antibiotic prophylaxis recommended in previous guidelines.
- Prophylaxis should be limited to the highest risk patients (patients with the highest incidence of IE and/or highest risk of adverse outcome from IE).

- The indications for antibiotic prophylaxis for IE should be reduced in comparison with previous recommendations.
- Good oral hygiene and regular dental review are of particular importance for the prevention of IE.

Recently, the Task Force of the European Society of Cardiology [152] decided to revise its previous guidelines from 2004 ([71]; [www.esacardio.org](http://www.esacardio.org)). The main reasons justifying the revision of previous recommendations are the following:

1. Incidence of bacteremia after dental procedures and during daily routine activities. As a large proportion of IE-causing bacteremia may derive from daily routine activities: toothbrushing, flossing, or chewing and because in patients with poor dental health, bacteremia cannot only be observed independently of dental procedures, but also rates of post-procedural bacteremia are higher in this group, it emphasizes the importance of good oral hygiene and regular dental review to prevent IE.

#### 2. Risks and benefits of prophylaxis

The following considerations are critical with respect to the assumption that antibiotic prophylaxis can efficiently prevent IE in patients who are at increased lifetime risk of the disease:

- (a) Increased lifetime risk of IE is not an ideal measure of the extent to which a patient may benefit from antibiotic prophylaxis for distinct procedures.
- (b) In the majority of patients, no potential index procedure preceding the first clinical appearance of IE can be identified.
- (c) Antibiotic administration carries a small risk of anaphylaxis.
- (d) Widespread and often inappropriate use of antibiotics may result in the emergence of resistant microorganisms.

3. Lack of scientific evidence for the efficacy of infective endocarditis prophylaxis as studies reporting on the efficacy of antibiotic prophylaxis to prevent or alter bacteremia in humans after dental procedures are contradictory, and so far there are no data demonstrating that reduced duration or frequency of bacteremia after any medical procedure leads to a reduced procedure-related risk of IE.

Although recent guidelines proposed limitation of prophylaxis to patients at increased risk of adverse outcome of IE [168] or even complete cessation of

antibiotic prophylaxis in any patient groups [134], the ERC Task Force decided

- To maintain the principle of antibiotic prophylaxis when performing procedures at risk of IE in patients with predisposing cardiac conditions, but
- To limit its indication to patients with the highest risk of IE (Table 2.7) undergoing the highest risk procedures (Table 2.8) (manipulation of the gingival or periapical region of teeth or perforation of the oral mucosa, including scaling and root-canal procedures).

The ESC Task Force does not recommend prophylaxis in cardiac transplant recipients who develop cardiac valvulopathy. Prophylaxis is not recommended for any other form of native valve disease (including the most commonly identified conditions, bicuspid aortic valve, mitral valve prolapse, and calcific aortic stenosis).

The main targets for antibiotic prophylaxis in these patients are oral streptococci. The impact of increasing resistance of these pathogens for the efficacy of antibiotic prophylaxis is unclear. Fluoroquinolones and glycopeptides are not recommended due to their unclear efficacy and the potential induction of resistance.

In summary, the Task Force proposes limitation of antibiotic prophylaxis to patients with the highest risk of IE undergoing the highest risk dental procedures. Good oral hygiene and regular dental review have a very important role in reducing the risk of IE. Aseptic measures are mandatory during venous catheters manipulation and during any invasive procedures in order to reduce the rate of health care-associated IE. It was also recommended to discuss the potential benefit and harm of antibiotic prophylaxis with the patients before a final decision is made. Following informed review and discussion, many may wish to continue with routine prophylaxis, and these views should be respected [92, 106].

## 2.2.6 Discussion

Antibiotic prophylaxis may be defined as the use of an antimicrobial agent before any infection has occurred for the purpose of preventing a subsequent infection [22]. Criteria for antibiotic prophylaxis against infection include the following: the health benefits must outweigh the antibiotic risks, the choice of antibiotic should be made on the single microorganism most

likely to cause an infection, and the cost–benefit ratio must be acceptable [121].

There are currently *insufficient primary data to know whether antibiotic prophylaxis before invasive dental procedures in people at high risk of endocarditis does actually prevent endocarditis*, deaths, or other serious illness [43, 119]. A recent Cochrane review [119] was performed with the aim to determine whether prophylactic antibiotic administration compared to no such administration or placebo before invasive dental procedures in people at increased risk of bacterial endocarditis influences mortality, serious illness, or endocarditis incidence. The search strategy run on MEDLINE (1950 to June 2008) and adapted for use on the Cochrane Oral Health, Heart, and Infectious Diseases Groups' Trials Registers, as well as the following databases: CENTRAL (The Cochrane Library 2008, Issue 2); EMBASE (1980 to June 2008); and the metaRegister of Controlled Trials (to June 2008). No randomized controlled trials (RCTs), controlled clinical trials (CCTs), or cohort studies were included. The only included case-control study [158], which included all of the people in The Netherlands who developed endocarditis following an invasive dental procedure while at known cardiac risk over a 2-year period (24 individuals who underwent a procedure definitely requiring prophylaxis, and a further 20 which may possibly have required prophylaxis), provided no conclusive evidence about whether antibiotic prophylaxis is effective or ineffective against bacterial endocarditis in such high-risk individuals about to undergo an invasive dental procedure [119].

Existing guidelines identified the gaps and inconclusive nature of the evidence available relating to antibiotic prophylaxis, although there is more evidence available for dental than for non-dental procedures. They also identified a lack of prospective, randomized RCTs on the efficacy of antibiotic prophylaxis to prevent IE. The AHA guideline [168] noted that some studies reported that antibiotics administered prior to a dental procedure reduced the frequency, nature, and/or duration of bacteremia whereas others did not. The BSAC guideline [59] commented on the need for a prospective double-blind study to evaluate the risk/benefit of prophylactic antibiotics, but also noted that this is unlikely to be undertaken due to the numbers of patients that would be required and while guidelines continue to

recommend prophylaxis. The ESC guideline [71] discussed that antibiotic prophylaxis may not be effective in preventing bacterial endocarditis if the amount of bacteremia in terms of colony forming units (CFU) is very large. These guidelines assessed and discussed the available evidence and reached conclusions that ranged in emphasis with the AHA taking an approach that would involve fewer patients than previously getting antibiotic prophylaxis, while the BCS/RCP [132] continued to recommend antibiotic prophylaxis for many dental and non-dental procedures [116].

On review of the *international and national guidelines* it is clear that there are also several points of agreement. All agree that there is a high-risk cardiac group where the risk of endocarditis exceeds the risk of antibiotic prophylaxis from dental at-risk procedures, thus warranting antibiotic prophylaxis. There are, however, some variations as to which conditions constitute high-risk cardiac states. All agree that there is a large group of patients with a cardiac history who are at no greater risk of endocarditis than the general population. In these patients the risk of anaphylaxis and similar adverse events from the antibiotic prophylaxis is demonstrably greater than the risk of endocarditis. Thus, in these circumstances, antibiotic prophylaxis is not warranted for any type of dental treatment [145]. To date, only the 2002 French recommendations take into consideration the patient's health background in modulating the indication of prophylaxis [43].

In regard to dental interventions, there is consensus that dental manipulations of either the hard or soft tissues will result in a bacteremia. However, there are variations as to where exactly to draw the line. There is also general consensus as to those procedures which have only a physiologic level of bacteremia and thus do not require prophylaxis. Again there are some differences [145].

There are some points of consensus on the choice of antibiotic dose and route for prophylaxis. If it is indicated then an appropriate antibiotic should be chosen to cover the most likely causative organisms, that is, the oral flora [145]. Antibiotics should act on the primary bacteremia responsible for infective endocarditis: oral streptococci for dental procedures, and enterococcus and group D streptococci for digestive or urinary-tract procedures. The main antibiotics to have proved effective in experimental models are

amoxicillin, vancomycin, teicoplanin, clindamycin, synergistins, azithromycin, and clarithromycin. When antibiotic prophylaxis is recommended before dental and upper respiratory procedures, amoxicillin is used in most countries, at doses varying from 2 to 3 g (1 h before procedure). For patients allergic to  $\beta$ -lactams, clindamycin or a glycopeptide are substituted; azithromycin or clarithromycin are recommended in some guidelines. For dental procedures, antibiotic prophylaxis is generally administered as a single dose in the hour before the procedure [43]. There is little value from a follow-up dose on the same day. Antibiotics in the preceding days and days after the procedure are not clinically indicated as they offer no additional advantage [145].

Other methods of antimicrobial prophylaxis have also been proposed for dental procedures, notably the use of *topical oral antimicrobials*, although there has also been concern that their routine use may provoke the selection of resistant microorganisms [22]. However, contradictory evidence and conclusions were identified also regarding topical antiseptics. The AHA guideline considered that the body of evidence showed no clear benefit [168]; the BCS/RCP guideline [135] advised the use of chlorhexidine as an oral rinse, although it did note that recent work has questioned its effectiveness [116].

Differences in such recommendations have caused much debate and continue to stir controversy in the international medical and dental communities [28].

It is thus important for dentists to be familiar with the current guidelines and the rationale behind them. This will allow them to alleviate their own concerns, reeducate their patients about the changes, and communicate effectively with their medical colleagues to optimize the continued safe delivery of patient care [84].

There is no doubt that the changes in IE guidelines will have also an *impact on patients*. Patients who, for many years, have always had antibiotic prophylaxis provided for dental treatment will need to be advised and counseled as to why this is no longer necessary. Some of these patients will be relieved but some will be confused as to why the use of antibiotics, which they were once informed was essential, is no longer advised. Traditional practice in any aspect of culture, including medicine and dentistry, always takes time to be changed. There will no doubt be some medical and dental practitioners who will be resistant to change and

will still want to give antibiotic prophylaxis to their patients. This is particularly so if the practitioner underestimates the possibility of an adverse reaction to the antibiotic and the broader community issue of bacterial resistance [34].

Despite the varying guidelines produced over the years and the recent significant change recommended by NICE, it is important for medical and dental practitioners to remember that patients remain at risk from developing endocarditis. Many patients will develop endocarditis with organisms that have originated from the oral cavity. While there is no evidence that dental treatment is directly related to the development of the disease or not, nor that prophylactic antibiotics can prevent the development of the disease or not, it would appear logical to recommend that the *highest level of oral health* should be achieved and maintained in at-risk patients and regular preventive dental checkups done [43, 119].

## 2.3 Antibiotic Prophylaxis before Invasive Dental Procedure in Patients with Joint Replacements

Approximately 450,000 total joint arthroplasties are performed annually in the USA. Deep infections of these total joint replacements usually result in failure of the initial operation and the need for extensive revision. Owing to the use of perioperative antibiotic prophylaxis and other technical advances, deep infection occurring in the immediate postoperative period resulting from intraoperative contamination has been reduced markedly in the last 20 years [9].

Infections in joint replacements are divided into early or late occurring. Early infections, that is within the first 3 months following implantation, primarily relate to infection introduced at the time of the operation, either sourced from the patient or the surgical staff. The incidence of this is low and of the order of 0.39% [107, 125, 142].

Later infections, more than 3 months after primary implantation, are usually secondary to bacteremia. The incidence is low and of the order of 0.97%. The prevalent bacteria are *Staphylococcus aureus* (35%) and *Staphylococcus epidermidis* (15%). These are of skin origin. Some or most of these may even have been

introduced at the time of surgery but have a delayed presentation. Group A streptococci, which are mainly of oropharyngeal origin, occurred in about 8% of cases. *Escherichia coli*, which is the classic alimentary tract bacteria, were involved in about 4% of cases. Thus bacteremic-related joint infections may occur but generally at a low incidence. Skin organisms are the predominant group. The risk of oral-related infections is very low with figures in the range of 0.04–0.07% [90, 102, 142].

Most orthopedic surgeons are aware that the former catastrophes in joint replacement surgery were caused by the common practice of using implants without documented good long-term results. Unfortunately, this practice is still common in most countries [69].

For example, after 15 years of use, there is no good documentation of long-term results from clinical studies of patients operated on with prostheses based on the metal-on-metal principle, although 60,000 of these prostheses have been used in central Europe. There is still uncertainty about which metal (chrome cobalt, stainless steel, or titanium) is better in joint prostheses, and about the surface and geometry of cemented hip prostheses (polished, matt, precoated, taper, collar). Of the uncemented implants that are presently used, very few have reportedly good long-term results. The problems with polyethylene wear of the uncemented cups have still not been solved. Many orthopedic surgeons consider the new highly cross-linked polyethylenes to be the solution of the wear problems. Testing of these products in laboratory situations is promising, but they have now been marketed before clinical results became available. To complicate the situation further, many of the manufacturers of prostheses change their products before the long-term results are known. New cements, without documentation on good clinical results, were introduced on the market in 1999, even though it was well known that changes in the cement formulae might give unexpected problems [69].

There is a lack of robust evidence linking dental procedures to an increased risk of infection of prosthetic joints [120]. The several observational studies, case reports and series, or surveys in the literature of infected joints thought to have been caused by hematogenous spread from an oral focus, as reviewed by several authors [27, 30, 91, 149, 157] are summarized in Table 2.15.

### 2.3.1 Existing Guidelines

The use of antibiotics to prevent infections of prosthetic joints following dental treatment remains controversial. Numerous different guidelines on prophylactic regimes have been proposed, including those from the The Australian Orthopaedic Association, British Orthopaedic Association, Swiss Society for Infectious Diseases, and New Zealand Orthopaedic Association, that have all made very similar suggestions for prescribing prophylactic antibiotics only for patients at an increased risk of infection. For their respective risk-factor lists, the corresponding guideline of each association should be referred to. While dental associations attempt to identify high-risk patients for the prescription of prophylactic antibiotics, the American Academy of Orthopaedic Surgeons published a more inclusive guideline in 2009 [82, 61, 157].

#### 2.3.1.1 2003: American Dental Association and American Academy of Orthopaedic Surgeons

In 1997, an expert panel of dentists, orthopedic surgeons, and infectious disease specialists convened by the American Dental Association and the American Academy of Orthopaedic Surgeons, or AAOS, performed a thorough review of all available data to determine the need for antibiotic prophylaxis to prevent hematogenous prosthetic joint infections in dental patients who have undergone total joint arthroplasties. The 2003 advisory statement was the first periodic update of the 1997 statement. In addition, the organizations have created a new patient handout (included at the end of the statement) that dentists may share with their patients.

The statement emphasizes that patients should be in optimal oral health prior to having total joint replacement and should maintain good oral hygiene and oral health following surgery. Any patient with a total joint prosthesis with acute orofacial infection should be vigorously treated as any other patient with elimination of the source of the infection (incision and drainage, endodontics, extraction) and appropriate therapeutic antibiotics when indicated [2].

**Table 2.15** Summary of studies of infected joints replacements due to hematogenous spread from an oral focus

| Reference                | Type of study | Study population  | Main outcomes   |
|--------------------------|---------------|---|---|
| Jacobsen & Murray [75]   | Retrospective | 1,855 hip prosthesis placed between 1970 and 1975   | Thirty-three cases of infected hips out of a total of 1,855 hip prosthesis placements. The infections were classified as early (less than 6 months after placement) or late (greater than 6 months after placement). In the patients studied, the risk of infection associated with dental procedures was extremely low (0.05%). <i>Staphylococcus aureus</i> was the organism most often isolated from the infected hips, and its incidence was twice as high in the late as in the early infections   |
| Ainscow & Denham [3]     | Prospective   | 1,000 patients who received 1,112 total joint replacements between 1966 and 1980 were followed up prospectively for an average of 6 years. These patients were not advised to take antibiotics prophylactically to cover subsequent dental or surgical procedures and, so far, only three cases of hematogenous infection at the site of the joint replacement have developed | 450 patients had not been at risk of transient bacteremia since receiving their implant, and that 224 patients had undergone dental or surgical procedures. None of these patients developed hematogenous infection. However, 288 patients had developed urinary tract infection, respiratory tract infection, or multiple infections at various sites; some of these patients had also undergone dental or surgical procedures but none developed hematogenous infections. Of the 40 patients who had recurrent skin ulceration and infection, three developed hematogenous infection (7.5%) ( $P < 0.01$ ). These results suggest that transient bacteremia is not likely to infect a replaced joint in otherwise healthy patients  |
| Lindqvist & Sjästis [89] | Case report   | Case reports of three patients with late hip replacement infection  | The microorganism related to the hip replacement infection was microaerophilic <i>Streptococcus viridans</i> , an oral organism, in all patients. Dental procedures had preceded the onset of the hip infection in all cases, and severe periodontal disease was observed on subsequent admissions  |
| Strazzeri & Anzel [149]  | Case report   | Case report of a 61-year-old woman developed an infected total hip arthroplasty after dental work   | The patient denies ever being instructed to take prophylactic antibiotics by her orthopedic surgeon, by her internist, or by her dentist  |
| Grogan et al. [62]       | Retrospective | Evaluation of 821 total knee arthroplasties performed in 604 patients, all of whom received perioperative antibiotics, performed between 1971–1982 at the University of California at Los Angeles Medical Center, USA   | Deep sepsis, proved by a positive culture of a specimen obtained by postoperative arthrocentesis, developed 14 times in 13 knees of 12 patients, an incidence of 1.71%. In one of these patients, who had systemic lupus erythematosus and bilateral knee replacement, the right knee became infected with two distinct organisms on two different occasions (separated by 10 months). The first infection, was probably hematogenous while the second, developing after a dental procedure, definitely was. Over-all, five infections were hematogenous with an identified source and one other was suspected of having a hematogenous origin. The time from operation to the diagnosis of sepsis averaged 8.3 months overall, but five of the 14 infections were recognized less than 2 months after arthroplasty. For the six infections that were assumed to be hematogenous, the time from operation to the diagnosis of sepsis averaged 16.4 months |

(continued)

Table 2.115 (continued)

| Reference                    | Type of study   | Study population  | Main outcomes  |
|------------------------------|---|---|--|
| Ching et al.<br>[27]         | Review of the English language literature from 1973 to 1987 | 110 case reports of hematogenous infection of large prosthetic joints.  | 48 cases (43–6%) were due to <i>Staphylococcus aureus</i> , 16 to β-hemolytic streptococci (groups A, B and G), 19 to enterobacteria (including one <i>Salmonella typhimurium</i> ), seven to <i>Streptococcus pneumoniae</i> , seven to <i>Staph.epidermidis</i> , four to viridans streptococci, four to <i>Stfaecalis</i> , four to <i>Pseudomonas aeruginosa</i> , two to <i>Bacteroides fragilis</i> , and one each to <i>Clostridium perfringens</i> , a diphteroid, <i>Hemophilus influenzae</i> and <i>Mycobacterium tuberculosis</i> . Five were mixed infections. Twelve cases were attributed to the dental abscesses or periodontal disease, some in association with dental procedures. (However, only four infections with viridans streptococci are recorded)   |
| Sullivan et al.<br>[150]     | Case report   | Case report of a 44-year-old woman who had had bilateral total hip replacement in May 1983 for bilateral congenital dysplasia of the hip and developed an hip infection 5 years later after dental treatment  | The sequence of events in the patient suggests that an infection can occur in the area of a total joint replacement after dental manipulation when no oral abscess is present. A temporary crown was placed without difficulty, the procedure being preceded and followed by oral administration of antibiotics. Twelve days later, the tooth became painful. The dentist noticed some food impacted under the temporary crown and treated this promptly by removing the food, but the patient already had gingival irritation. Manipulation during this urgent dental procedure, as during routine dental procedures, could result in bacteremia. The patient took erythromycin after treatment. Two days after the second dental treatment, without any other recognizable cause, she began to have pain in the left hip. After thorough investigation, this proved to be due to an infection with <i>Peptostreptococcus</i> species, an oral organism. The authors thank Daniel McIntroy, D |
| Jacobsen et al.<br>[75]      | Retrospective   | Hospital and dental charts of 2,693 patients in whom total prosthetic joints had been placed at the Veterans Administration Hospitals of Ann Arbor and Allen Park, Michigan, as well as at The University of Michigan Hospital, USA   | Of the thirty (1.1%) late prosthetic joint infections (greater than 6 months after placement), only one (0.04%) could be temporally associated with dental treatment. Nine of the 30 late infections occurred in insulin-dependent diabetic patients and patients on long-term immunosuppressive therapy. An analysis of the organisms isolated from the late infections shows that 54% where <i>Staphylococcus epidermidis</i> and <i>Staphylococcus aureus</i> . The authors concluded that these data do not support the practice of prescribing prophylactic antibiotic coverage of prosthetic hip and knee joints prior to all dental therapy. Rather, use of antibiotics during dental treatment appears warranted only if a chronic bacteremia is anticipated or where a predisposing systemic condition may exist  |
| Bartzokas<br>et al. [16]     | Case series   | Four cases of adults aged 58–83 year with deep infection – three in total knee replacements and one in a total hip replacement – caused by <i>Streptococcus sanguis</i> (an (alpha)-hemolytic streptococcus of the viridans group), a common constituent of the oral microflora | For each patient the strain of <i>S. sanguis</i> isolated from the mouth was indistinguishable from that isolated from the prosthesis. All patients had severe periodontal disease and caries  |
| Skiest & Coykendall<br>[146] | Case report   | Case report of a 39-year-old man with systemic lupus erythematosus (SLE) and osteonecrosis of both hips who had a right total hip replacement   | The patient had a prosthetic hip infection with <i>Streptococcus oralis</i> despite receiving prophylaxis with erythromycin for a comprehensive dental procedure, at which time he received a complete oral examination with periodontal probing, scaling, and flossing. At the dental examination several carious lesions were found, and pocket depths of 5 and 6 mm were seen between some upper posterior teeth with normal sulcal depths elsewhere  |

|                      |  |   |  |
|----------------------|--|---|--|
| Waldman et al. [166] | Retrospective  | Records of 3,490 patients treated with total knee arthroplasty by the authors between 1982 and 1993   | 62 total knee arthroplastics with late infections (greater than 6 months after their procedure) were identified, and of these, seven infections were associated strongly with a dental procedure temporally and bacteriologically. These seven cases represented 11% of the identified infections or 0.2% of the total knee arthroplasty procedures performed during this period. In addition, among 12 patients referred for infected total knee arthroplastics from outside institutions, two infections were associated with a dental procedure. Five of the nine (56%) patients had systemic risk factors that predisposed them to infection, including diabetes and rheumatoid arthritis. All dental procedures were extensive in nature (average, 11.5 min; range, 75–205 min). Eight of the patients received no antibiotic prophylaxis. One patient had only one preoperative dose   |
| LaPorte et al. [85]  | Retrospective  | The records of 2,973 patients after total hip arthroplasty (THA) between 1982 and 1994 were reviewed  | 52 (1.7%) had a deep late infection, defined as being diagnosed more than 6 months after operation. Infection occurring earlier than this was not included since it may have been related to the primary operation. Factors identified which may have predisposed patients to infection included the use of corticosteroids, rheumatoid arthritis, diabetes mellitus, and open wounds. Three patients had a dental procedure within the 2 weeks before onset of evidence of infection in the hip, with the organism cultured being identified as oral in origin. No other source for the infection was found. The times since their primary arthroplasties were 15, 24, and 39 months, respectively. These cases contributed 6% of the 52 late infections identified and 0.1% of the 2,973 arthroplasties which had been carried out. The associated dental episodes were multiple-tooth extractions, root-canal operations, and a periodontal procedure, lasting 45, 60, and 90 min, respectively. The patients did not receive prophylactic antibiotics. The onset of symptoms in the hip followed the dental procedure by two, five, and 11 days, respectively, in the three patients. All complained of pain in the affected hip and one had both pain and swelling. None had a temperature greater than 38°C. The organisms cultured were <i>Streptococcus viridans</i> in two cases and <i>Pepostreptococcus</i> in one. They were sensitive to most tested antibiotics, including penicillin. All the patients were treated with a staged reimplantation. Appropriate antibiotics were administered intravenously for 6 weeks |
| Berbari et al. [19]  | Prospective single-center, case-control study for the period 2001–2006 | 339 case patients hospitalized with total hip or knee infection. 339 control subjects were patients who underwent a total hip or knee arthroplasty but without a prosthetic joint infection who were hospitalized during the same period on the same orthopedic floor | There was no increased risk of prosthetic hip or knee infection for patients undergoing a high-risk or low-risk dental procedure who were not administered antibiotic prophylaxis (adjusted OR = 0.8; 95% CI: 0.4–1.6), compared with the risk for patients not undergoing a dental procedure (adjusted OR = 0.6; 95% CI: 0.4–1.1, respectively). Antibiotic prophylaxis in high-risk or low-risk dental procedures did not decrease the risk of subsequent total hip or knee infection (adjusted OR = 0.9, 95% CI: 0.5–1.6 and 1.2, 95% CI: 0.7–2.2, respectively). The authors concluded that dental procedures were not risk factors for subsequent total hip or knee infection. The use of antibiotic prophylaxis prior to dental procedures did not decrease the risk of subsequent total hip or knee infection   |

Practitioners should maintain a high index of suspicion for any unusual signs and symptoms (such as fever, swelling, pain, joint that is warm to touch) in patients with total joint prostheses [2].

Related to antibiotic prophylaxis the panel suggests that

1. Antibiotic prophylaxis is not indicated for dental patients with pins, plates, and screws, nor is it routinely indicated for most dental patients with total joint replacements.
2. There is limited evidence that some immunocompromised patients with total joint replacements (inflammatory arthropathies such as rheumatoid arthritis, systemic lupus erythematosus, and drug- or radiation-induced immunosuppression) may be at higher risk of experiencing hematogenous infections. Antibiotic prophylaxis may be considered for such patients undergoing dental procedures with a higher bacteremic risk, as defined by Dajani et al. [32].
  - Dental extractions
  - Periodontal procedures, including surgery, subgingival placement of antibiotic fibers/strips, scaling and root planning, probing, recall maintenance
  - Dental implant placement and replantation of avulsed teeth
  - Endodontic (root canal) instrumentation or surgery only beyond the apex
  - Initial placement of orthodontic bands but not brackets
  - Intraligamentary and intraosseous local anesthetic injections
  - Prophylactic cleaning of teeth or implants where bleeding is anticipated should be considered using an empirical regimen
3. Antibiotic prophylaxis may be also considered when the higher-risk dental procedures are performed on dental patients within 2 years post-implant surgery, on those who have had previous prosthetic joint infections and on those with some other conditions (malnourishment, hemophilia, HIV infection, insulin-dependent (type 1) diabetes, malignancy).

The ADA/AAOS advisory statement, which lists high-risk dental procedures and suggested antibiotic prophylaxis regimens, is found at:<sup>\*</sup> <http://jada.ada.org/cgi/content/full/134/7/895>

### **2.3.1.2 2003: New Zealand Dental Association**

The New Zealand Dental Association code of practice has the following suggestions regarding antibiotic prophylaxis for dental treatment of patients with prosthetic joint replacements:

- Patients with oral pathology, such as abscesses and/or periodontal disease, are theoretically at increased risk of prosthetic joint infection. It is therefore prudent that all patients scheduled for prosthetic joint replacement should have a dental examination, and treatment as required, to reduce and remove sources of oral bacteremia.
- Patients with a prosthetic joint replacement should have a regular dental examination, and treatment as required, to remove sources of oral bacteremia.
- Routine use of antibiotic prophylaxis for all patients with a prosthetic joint replacement is not justified.
- Antibiotic prophylaxis could be considered for dental procedures producing a significant bacteremia

The New Zealand Dental Association code of practice regarding antibiotic prophylaxis for dental treatment of patients with prosthetic joint replacements can be found at: <http://www.nzoa.org.nz/download.php?id=31272,5,1>

### **2.3.1.3 2003: British Orthopedic Association and British Dental Association**

The guideline for antibiotic prophylaxis proposed by British Orthopaedic Association and British Dental Association guide [144] to practice on joint replacement, dental treatment, and antibiotic reveals:

1. Intuitively, good oral hygiene and regular dental advice are imperative for patients with large joint replacements or those anticipating such operations. Dental advice should be sought where there is doubt about oral sepsis.
2. Routine antibiotic prophylaxis should not be offered to all patients undergoing dental treatment.
3. Antibiotic prophylaxis is advised in patients with systemic immunosuppressive disease, for example, diabetes (type I and II), rheumatoid arthritis, haemophilia, or malignancy (either from the immunosuppressive effects of the malignancy or those of treatment).

4. Prophylaxis is clearly indicated where there is overt oral sepsis, for example, any kind of preexisting oral infection which could lead to metastatic spread.
5. Prophylaxis should be considered where dental treatment is invasive, complex, and of long duration.

The British Orthopedic Association and British Dental Association (2003) suggested the following antibiotic prophylaxis [144]:

Under local or no anesthetic:

- Patients *not allergic* to penicillin: Amoxicillin 3 g orally, 1 h prior to dental procedure
- Patients *allergic* to penicillin: Clindamycin 600 mg orally, 1 h prior to dental procedure

Under general anesthetic

- Patients *not allergic* to penicillin: Amoxicillin 1 g i.v. at induction followed by amoxicillin 500 mg orally 6 h later
- Patients *allergic* to penicillin: Clindamycin 300 mg i.v. at induction over 1 h

The need for antibiotic prophylaxis prior to dental treatment for patients with prosthetic joints seems to be driven exclusively by the orthopedists. The supportive evidence for such an indication appears equivocal at best and does not seem to be based upon a clear understanding of oral bacteremia arising either spontaneously or from dental treatment [144].

#### 2.3.1.4 2005: The Australian Orthopaedic Association

The guidelines for antibiotic prophylaxis, across the Royal Adelaide Hospital campus, were formulated in 1999 and revised in 2003. This was under the leadership of the Antibiotic Working Party, a subcommittee of the Drug Committee and it involved extensive consultation. This guideline has to a degree regulated what had been a highly individualistic approach to common problems. For patients with joint replacements, they agree that antibiotic prophylaxis was only indicated for high-risk dental procedures in immunocompromised patients with joint problems [142]:

1. Prior to placement of the first artificial joint
2. Dental problem in the first 3 months following artificial joint placement

3. Dental treatment after 3 months in a patient with a normally functioning artificial joint
4. Dental treatment for patients with significant risk factors for artificial joint infection
  - Immunocompromised patients include
  - Failing, particularly chronically inflamed, artificial joints
  - *Previous history of infected artificial joints*
    - Routine nonsurgical dental treatment – no prophylaxis indicated
    - Antibiotic prophylaxis recommended for
      - All extractions
      - Deep periodontal scaling
    - Regular dental reviews mandatory

5. Established infection by oral organisms on an artificial joint
  - Urgent referral to *dentist* to determine and eliminate any oral cause
  - Aggressive treatment by removal of the cause, extraction or endodontic under antibiotic prophylaxis

Logically, the first step should be that all patients undergoing joint replacement should be dentally fit. Dental treatment in the preimplantation phase should be aggressive to eliminate current foci of infection. If the condition cannot be rapidly resolved by restorative, endodontic, or periodontal treatment the involved teeth should be extracted. Antibiotic prophylaxis would not usually be required for such preimplantation treatment [142].

Standard antibiotic prophylactic regimens are presented in Table 2.16 [142].

If the patients demand that they have antibiotic treatment for all dental treatment because their orthopedic surgeon has advised them that they should, one does need to carefully evaluate the precise nature of the dental problem and the treatment needs. One then needs to communicate with the patient's orthopedic surgeon so that the actual risk–benefit situation can be determined. If there is no indication for antibiotic prophylaxis in accordance with these guidelines but the orthopedic surgeon is insistent that antibiotics are given, then an informed consent decision needs to be made. If antibiotics are requested, it needs to be recorded that the orthopedic surgeon requested prophylaxis be given. Hence, the orthopedic surgeon would bear the responsibility for any adverse outcome related to the administration of the antibiotic therapy [142].

**Table 2.16** Recommended antibiotic regimens (Australian Orthopaedic Association; Reprinted from [142]. With permission from John Wiley and Sons)

| Condition  | Drug regimes  |
|--|---|
| 1. Dental clinic LA extractions or deep curettage  | Amoxycillin 2–3 g orally 1 h prior to procedure   |
| 2. Theatre procedures  | Amoxycillin 1 g I/V at induction<br>Followed by 500 mg amoxicillin i.v. or orally 6 h later.  |
| 3. Penicillin hypersensitivity, long-term penicillin, recent penicillin/other $\beta$ -lactam    | Clindamycin 600 mg 1 h prior to procedure or Vancomycin 1g i.v. 1 h to finish 2 h or Lincomycin 600 mg just prior to the procedure  |
| 4. High-risk case (i.e., Gross oral sepsis/ severely immunocompromised/previous joint infection) | Gentamicin 2 mg/kg i.v. just before procedure ( <i>can be administered 3 mg/kg provided there is no concomitant renal disease</i> )<br>PLUS Amoxycillin 1 g i.v. just before procedure followed by 500 mg I/V or orally 6 h later<br>If hypersensitive to penicillin replace amoxicillin with Vancomycin 1 g I/V over 1 h to finish just before procedure |

- All patients with prosthetic joint replacement
- Immunocompromised/immunosuppressed patients
- Inflammatory arthropathies (e.g., rheumatoid arthritis, systemic lupus erythematosus)
- Drug-induced immunosuppression
- Radiation-induced immunosuppression
- Patients with co-morbidities (e.g., diabetes, obesity, HIV, smoking)
- Previous prosthetic joint infections
- Malnourishment
- Hemophilia
- HIV infection
- Insulin-dependent (Type 1) diabetes
- Malignancy
- Megaprostheses

In order to prevent bacteremia, an appropriate dose of a prophylactic antibiotic should be given prior to the procedure so that an effective tissue concentration is present at the time of instrumentation or incision in order to protect the patient's prosthetic joint from a bacteremia-induced periprosthetic sepsis. Current prophylactic antibiotic recommendations for dental procedures are cephalexin, cephadrine, or amoxicillin, 2 g orally, 1 h prior procedure.

AAOS (2009) considered that the statement provides recommendations to supplement practitioners in their clinical judgment regarding antibiotic prophylaxis for patients with a joint prosthesis. It is not intended as the standard of care nor as a substitute for clinical judgment as it is impossible to make recommendations for all conceivable clinical situations in which bacteremias may occur. The treating clinician is ultimately responsible for making treatment recommendations for his/her patients based on the clinician's professional judgment. Any perceived potential benefit of antibiotic prophylaxis must be weighed against the known risks of antibiotic toxicity, allergy, and development, selection, and transmission of microbial resistance. Practitioners must exercise their own clinical judgment in determining whether or not antibiotic prophylaxis is appropriate.

However, several critical articles were published recently with regard to the AAOS (2009) recommendations [105, 112, 115].

The 2009 AAOS advisory statement can be found at <http://www.aaos.org/about/papers/advistmt/1033.asp>

### 2.3.1.5 2009: American Academy of Orthopaedic Surgeons

In 2009, the American Academy of Orthopaedic Surgeons released the Information Statement: "Antibiotic Prophylaxis for Bacteremia in Patients with Joint Replacements," developed as an educational tool.

Given the potential adverse outcomes and cost of treating an infected joint replacement, the American Academy of Orthopaedic Surgeons (2009) recommends that clinicians consider antibiotic prophylaxis for all total joint replacement patients prior to any invasive procedure that may cause bacteremia. This is particularly important for those patients with one or more of the following risk factors:

## 2.4 The Prevention of Infection Following Dental Surgical Procedures

There are numerous local surgical and dental surgical procedures and medical conditions that are routinely covered by systemic antibiotics in an attempt to prevent postoperative infections. These can be considered as follows [143].

- Local wound infection that may not jeopardize the procedure (e.g., removal of an impacted lower third molar)
- Local infection that may jeopardize the procedure (e.g., installation of endosseous implants)
- Distant metastatic infection (e.g., infection of an indwelling vascular stent)
- Fulminant sepsis (e.g., the severely immunocompromised patient)

However, present evidence on the need for antibiotic prophylaxis in relation to periodontal surgery and implant placement is equivocal. Postoperative regimens afford little advantage for implant survival [143].

Patients with a lowered local or general resistance to infection may be placed at special risk by invasive dental procedures. It is probable that the risks to these patients are higher than those in the categories considered earlier and that their relative rarity does not raise the same public-health issues [143].

However, two recent systematic reviews, [91] and [153] found no evidence to support the practice of antibiotic prophylaxis before undergoing dental procedures or to demonstrate that it prevents distant site infections for any patients with:

- Cardiac-native heart valve disease, prosthetic heart valves, and pacemakers
- Hip, knee, and shoulder prosthetic joints
- Renal dialysis shunts
- Cerebrospinal fluid shunts
- Vascular grafts
- Immunosuppression secondary to cancer and cancer chemotherapy
- Systemic lupus erythematosus
- Insulin-dependent (type 1) diabetes mellitus

Termine et al. [153] suggested that antibiotic prophylaxis is advisable only in a small percentage of patients

where a risk of severe infective complications (i.e., infective endocarditis and prosthetic joint infection, septicemia in severely immunocompromised patients, bisphosphonate-related osteonecrosis of the jaw) exists.

### 2.4.1 The Asplenic Patient

Surgical removal of the spleen is performed for several reasons, including trauma, immunological diseases, hypersplenism, and malignancy. Patients with functional or anatomic asplenia are at a significantly increased risk of overwhelming infection (postsplenectomy sepsis [PSS]), particularly involving the encapsulated bacteria *Streptococcus pneumoniae* and *Haemophilus influenzae*. The risk is highest in infants and young children, but adults also have an increased risk of infection. Preventive strategies are very important and fall into three major categories: immunoprophylaxis, antibiotic prophylaxis, and education [108].

There is a consensus among the various guidelines regarding immunization prior to elective splenectomy and posttrauma. The British Committee for Standards in Hematology guideline update, based on Medline, BID Embase, and Cochrane Library concluded that triple vaccination with Pneumococcal, *Hemophilus Influenzae*, and meningococcal vaccines is essential. It is accepted, however, that compliance may be a problem with lifelong oral antibiotic prophylaxis (<http://www.bcsghguidelines.com/pdf/SPLEEN21.pdf> accessed 4.05.2010) [37, 79, 87].

The Working Party of the British Committee for Standards in Clinical Hematology Task Force [35] recommends lifelong prophylaxis in all cases, especially in the first 2 years after splenectomy, for all children aged up to 16 years. A more flexible approach has been suggested for those patients who find it difficult to comply with a lifelong regimen [79, 167].

### 2.4.2 Transplantation Patients

Organ transplantation has become a widely accepted treatment modality for end-stage diseases. The progressive success of this therapeutic modality is a result

of advances in surgical techniques, organ preservation, improved anti-rejection therapy, and more effective antibiotics to prevent and to treat infectious complications. Because infection and rejection treatment are closely interrelated, the advent of newer and more potent immunosuppressive agents aimed at decreasing allograft rejection have led to an increase in the risk and morbidity of infectious complications in transplant recipients [63, 163].

Bacterial infection and sepsis are major complications following organ transplantation. Infection from a dental source is a potential threat for both organ transplant candidates and recipients since dental disease is a ubiquitous condition and is also likely to be more severe and untreated in the transplant population. The cumulative effects of poor dental health, untreated dental disease, and increased susceptibility to infection suggest that dental infections could pose a significant risk for the physically debilitated transplant candidate, as well as immunosuppressed transplant recipients [64].

The recipient of a transplant is likely to have fewer and less complex medical-dental management predicaments, but the dental practitioner may still be confronted by a number of other issues. Among these are metabolic derangements, including electrolyte disturbances and diabetes, which may be caused by the immunosuppressive drugs. The patient's immunosuppressive therapy

protocol may include the use of corticosteroids, which the dentist will also need to address [63].

Recommendations for pre- and post-transplantation dental care have been published and are summarized in Table 2.17.

### 2.4.3 Hematological Patients

Infection is a frequently occurring and serious complication in patients with hematological malignancies, due to periods of cytopenia following chemotherapy [161].

The BCSH Clinical Haematology Task Force last produced guidelines on levels of care relating to the provision of facilities for patients with hematological malignancies and severe bone marrow failure in 1995. Since then, the range of diagnostic methods for hematological malignancies has broadened, whilst the complexity and toxicity of many of the treatments for these diseases has increased considerably. Furthermore, there have been numerous important national developments in the provision of care for patients with cancer in general and for those with hematological malignancies in particular. These developments, as cited by BCSH [23], include: A Policy Framework for Commissioning Cancer Services (Calman Hine

**Table 2.17** Pre- and Post-transplantation dental care guidelines (Reprinted from [63]. With permission from Elsevier)

| Pretransplantation dental care guidelines   | Posttransplantation dental care guidelines   |   |  |
|---|--|---|--|
|   | Immediate posttransplantation period   | Stable posttransplantation period   | Posttransplantation rejection period   |
| <ul style="list-style-type: none"> <li>• Consult with patients' physician</li> <li>• Perform dental prophylaxis</li> <li>• Treat all active dental disease</li> <li>• Postpone elective treatment</li> <li>• Remove all potential sources of acute or chronic infection, including partially erupted third molars</li> <li>• Remove all nonrestorable teeth</li> <li>• Perform necessary denture adjustments</li> <li>• Reinforce oral hygiene and home care instructions</li> <li>• Initiate daily antibacterial mouth rinses</li> </ul> | <ul style="list-style-type: none"> <li>• Consultation with the physician/transplant coordinator</li> <li>• Emergency care of dental infections only</li> </ul> | <ul style="list-style-type: none"> <li>• Consultation with the physician/transplant coordinator</li> <li>• Frequent recall and prophylaxis</li> <li>• Daily antibacterial mouth rinses</li> <li>• All indicated dental care</li> <li>• No NSAIDS</li> </ul> | <ul style="list-style-type: none"> <li>• Consultation with the physician/transplant coordinator</li> <li>• Emergency care of dental infections only</li> </ul> |

Report – 1995), The NHS Plan (2000), The NHS Cancer Plan (2000), Cancer in Scotland: Action for change. The Scottish Executive Health Department (2001), National Institute for Clinical Excellence. Guidance on Cancer Services Improving Outcomes in Haematological Cancer – The Manual (2003), National Institute for Clinical Excellence. Guidance on Cancer Services Improving Supportive and Palliative Care for Adults with Cancer – The Manual (2004), Manual of Cancer Services (2004), The National Cancer Peer Review Programme, Better Cancer Care, An Action Plan. The Scottish Government (2008), HSC 2003/010 Updated National guidance on the safe administration of Intrathecal Chemotherapy (2008), For Better, for Worse? A report by the National Confidential Enquiry into Patient Outcome and Death (2008), Chemotherapy Services in England – Ensuring quality and safety. A report from the National Chemotherapy Advisory Group (August 2009).

The measures defined in these initiatives provide for a detailed specification of the standards that need to be met with respect to the care of patients with hematological cancers and for their external peer review assessment. In view of the developments in this area of clinical work, the BCSH felt it appropriate to review its document of 1995 so as to provide an updated guideline in 2010 for use both by providers of this clinical care and by those who commission it (The document can be found at [http://www.bcsghguidelines.com/pdf/draft\\_levelsofcare\\_042010.pdf](http://www.bcsghguidelines.com/pdf/draft_levelsofcare_042010.pdf)).

The primary focus in management of leukemic patients and those in whom myelosuppression is anticipated, is treatment of preexisting infection and prevention and treatment of acute complications during medical therapy. The long-term effects of chemotherapy are of less concern than those associated with high-dose radiation therapy, because chemotherapy effects are reversible. In neutropenic patients, the risk of infection during medical therapy may require aggressive antimicrobial therapy and is potentially life-threatening. Studies have shown that oral and periodontal assessment and management reduce the risk of infection and fever associated with oral conditions. Patients with chronic periodontal disease receiving high-dose chemotherapy may develop acute exacerbations from preexisting sites of disease during periods of neutropenia. Assessment of periodontal flora during chemotherapy showed a shift to increased Gram-negative bacilli in less than 50% of patients. Of

these, *Pseudomonas* species predominated, although *Klebsiella pneumoniae* was also present. Other potential pathogens identified were *Staphylococcus epidermidis*, *C. albicans*, *S. aureus*, and *P. aeruginosa* in primary infection or mixed culture, that appear to represent exacerbation of chronic infection due to indigenous flora when compared with noncancer patients. Thus, preexisting periodontal disease may serve as a source of infection in neutropenic patients. An oral source of septicemia was suspected in 25% of acute leukemic patients who received dental care with scaling prior to chemotherapy compared to 77% among patients who did not receive dental care prior to chemotherapy. The primary sources were pericoronitis or preexisting periodontal infections. No difference in the incidence of fever or bacteremia was seen in a study of leukemic patients receiving or not provided with a probing assessment and scaling of the teeth. Oral preventive care has been shown to not result in increased risk of bacteremia or in fever and is associated with less severe oral mucositis. Periodontal evaluation and treatment may reduce the severity of mucositis during treatment and reduce the potential for septicemia from periodontal sources, and should be a part of the management of patients at risk of neutropenia due to medical therapy ([149], reprinted with permission from Elsevier).

#### 2.4.4 HIV Infection

HIV disease is now considered a chronic illness requiring continued management and monitoring. However, for those with poor access to antiretroviral medications, the disease continues to be associated with higher morbidity and mortality. With the expansion of the HIV pandemic into vulnerable subpopulations, HIV care requires coordinated and integrated care for a complex mix of psychosocial and clinical services that must include oral health care [169].

The incidence of opportunistic infections (OIs) in HIV-infected persons declined substantially with the widespread use of combination antiretroviral therapy. This suggested the approach of discontinuing antibiotic prophylaxis in patients who experienced effective immune reconstitution (CD4 cell count increased to levels indicative of a low risk of opportunistic infection), thereby reducing toxicity and drug interactions, lowering

costs of care, and potentially improving adherence to antiretroviral regimens. Accordingly, the US Public Health Service/Infectious Disease Society of America (USPHS/IDSA) guidelines were updated in 2002, and specify the same or slightly more conservative CD4 count thresholds for discontinuing prophylaxis as those for initiating prophylaxis [60].

Guidelines for Prevention and Treatment of Opportunistic Infections in HIV-Infected Adults and Adolescents, Recommendations from CDC, the National Institutes of Health, and the HIV Medicine Association of the Infectious Diseases Society of America, 2009 can be found on <http://www.cdc.gov/mmwr/preview/mmwrhtml/rr5804a1.htm> [77].

A recent update of the HIV Medicine Association of the Infectious Diseases Society of America, replaces those published in 2004. The guidelines are intended for use by health care providers who care for HIV-infected patients or patients who may be at risk for acquiring HIV infection [1]. The 2009 version can be found at <http://www.idsociety.org/Content.aspx?id=1922>.

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## The Systemic Use of Antibiotics in Periodontal Therapy

Periodontal treatment aims at restoring a microbiota compatible with periodontal health. Effective therapy implies a reduction of pathogenic levels of indigenous oral microorganisms and elimination of exogenous pathogens and of organisms outside their ecological niche such as enteric rods and pseudomonas. Since both microbial and host susceptibility factors determine the periodontal health status, the composition and number of subgingival organisms associated with periodontal health may vary from individual to individual. Mechanical periodontal treatment can reduce total supra- and subgingival bacterial mass, but major pathogens may escape the effect of treatment due to their ability to invade periodontal tissues or to reside in furcations or other tooth structures outside the reach of periodontal instrumentarium, or due to poor host defence mechanisms. Periodontal antibiotic therapy aims to reinforce mechanical periodontal treatment and to support the host defence system in overcoming the infection by killing subgingival pathogens that remain after conventional mechanical periodontal therapy [207].

### 3.1 Advantages of Systemic Antibiotic Therapy in Periodontics

Systemic antibiotics present several advantages compared with mechanical debridement and topical application of antiseptics: via serum, systemic antibiotics can reach microorganisms at the base of deep periodontal pockets and furcation areas and may also affect organisms residing within gingival epithelial and connective tissues. Systemic antibiotics may be also capable of eradicating periodontal pathogens colonizing

oral mucosa and other extra-dental sites, and thus, reducing the risk of subgingival recolonization of pathogens and for future disease activity [207]. Motile rods were found significantly more often on the dorsum of the tongue and in the tonsillar area in subjects with periodontal breakdown compared with subjects without. Also, spirochetes were only found on the dorsum of the tongue in cases with periodontal breakdown. Black-pigmented *Bacteroides* spp., in untreated adult periodontitis, were identified at various sites in the oral cavity, including saliva [208]. Different prevalence values of *Aggregatibacter actinomycetemcomitans* were found in different patient populations. The organisms were isolated in pooled subgingival plaque of 68% early onset periodontitis and 24% adult periodontitis patients. While *A. actinomycetemcomitans* was recovered from cheek mucosa and saliva in early onset periodontitis, almost as frequently as from pooled subgingival plaque, the proportion of culture-positive extracrevicular samples in adult periodontitis patients was considerably lower. A significant positive association between the incidence of *A. actinomycetemcomitans* in infected individuals was identified for deep and normal periodontal sites, periodontal pockets and cheek, tongue and saliva, and cheek and saliva [128]. The long-term microbiological and clinical effects of mechanical debridement followed by metronidazole plus amoxicillin therapy in *A. actinomycetemcomitans*-associated periodontitis showed that this combined therapy was very effective in suppressing *A. actinomycetemcomitans* below cultivable levels, not only at the periodontal area but at all sites investigated, i.e., tongue, tonsillar area, cheeks, and saliva. This elimination was paralleled by a continuing reduction in periodontal probing depth (PPD) and bleeding index and gain in probing attachment level (AL) during a 24-month test period [148].

### 3.2 Disadvantages of Systemic Antibiotic Therapy in Periodontics

The use of antibiotics in periodontal therapy carries potential risks for the periodontium. Elevated levels of penicillin- and tetracycline-resistant bacteria on subgingival microflora were demonstrated in periodontitis patients. High proportions of enteric rods, pseudomonas, or yeasts in the subgingival flora of some patients who had received several courses of one or more systemic antimicrobial agents were reported. Prolonged or frequent courses of systemic antibiotic therapy may enable superinfecting organisms to persist in the subgingival microbial community over extended time periods. Opportunistic pathogens may sustain the periodontal pathology and may give rise to systemic complications [185].

Another potential disadvantage to long-term tetracycline therapy was suggested to be the permanent discoloration of the dentition that may not be limited only to tooth development in the child, but can also affect the adult dentition. When given over long periods of time in adults, the tetracycline molecule is incorporated into the continuously forming secondary dentin. The chronic sun exposure of the incorporated tetracycline may cause the formation of a reddish-purple oxidation product, resulting in discoloration of the permanent teeth [28].

Patient noncompliance can be a significant confounding factor in controlling a patient's medical condition. Compliance can be assured by having the patients take the medication under supervision, but is impracticable with most ambulatory patients. Compliance can be verified by examining biological fluids such as blood, urine, or saliva for the presence of the agent. An electronic system, built into the cap of the prescription bottle, which records the time and date when the bottle was opened was also suggested. The persistence of elevated levels or proportions of spirochetes in the plaque samples of any patient given metronidazole would be an indication that the patient was noncompliant [100].

### 3.3 Efficacy of Systemic Antibiotic Therapy in Periodontics

Factors that may influence the efficacy of antibiotics in the periodontal areas include:

- **The substantivity of a drug**, that is the ability of the drug to be retained in the mouth by adsorption to the teeth and soft tissues, is an important criterion for the success of a drug as an inhibitor of plaque formation. Two drugs with equal antimicrobial activity but different capacities for retention in the oral cavity would not be expected to have the same activity in inhibiting the formation of dental plaque. Thus, antimicrobial activity alone will not predict the efficacy of a drug as a plaque-preventive agent. It has been shown that tetracycline and its derivatives minocycline, oxytetracycline, and chlorotetracycline strongly adsorb to tooth surfaces retaining their antimicrobial activity. The other antibiotics including tyrothricin, vancomycin, spirofloxacin, streptomycin, kanamycin, neomycin, and actinobolin bind weakly to the tooth surface [11].
- **The protection of key organisms through binding and/or consumption of the drug by nontarget microorganisms.** This phenomenon may be significant in periodontal infections with a complex microbiota. *Enterococcus faecalis* can protect *Bacteroides fragilis* by inactivating metronidazole, hereby enabling *B. fragilis* to survive in a mixed microbiota of medical importance [207].
- **The efficacy of an antibiotic may be decreased by the localization of bacteria within gingival tissues from various forms of human periodontal disease.** The presence of *A. actinomycetemcomitans* in the gingival biopsies taken from sites adjacent to periodontal lesions in juvenile periodontitis patients was investigated by selective culture. Moreover, detection of viable organisms by culture techniques is always associated with *A. actinomycetemcomitans* in the pocket samples and also correlated with the presence of antigens in the tissues as estimated by immunofluorescence methods using antisera specific for *A. actinomycetemcomitans* [31]. Manor et al. [114] have demonstrated in cases of advanced periodontitis, bacterial penetration into the soft tissues of the apical zone of the pocket. Bacterial penetration occurred both in epithelial and connective tissue and appeared to follow enlarged epithelial intercellular spaces and bacteria were often located among debris of disintegrated epithelial cells.
- **The total bacterial load in the periodontal pocket** in relation to the maximal achievable antibiotic concentration may be too large to allow the elimination phenomenon known as inoculum effect. The

inoculum effect may be most critical for bacteriostatic agents such as the tetracyclines. However, it may also be applicable to the bactericidal agent metronidazole [207].

- In the case of **bacteriostatic tetracyclines**, they can suppress susceptible periodontal pathogens but are not able to completely eradicate some key subgingival organisms. The effectiveness of bacteriostatic antibiotics strongly depends on the host defence system, which may be impaired in the periodontal pocket [207].
- Penicillin-resistant strains were isolated from periodontally diseased sites which were positive for  $\beta$ -lactamase activity. However, it was found that greater than 50% of the activity may be inhibited by clavulanic acid, and inhibition appeared to be both irreversible and progressive [213].
- **Periodontal pathogens:** motile rods, spirochetes, black-pigmented *Bacteroides* spp., *A. actinomycetemcomitans*, were identified at various sites in the oral cavity, including saliva [129, 149, 208]. These organisms may seed the subgingival area after periodontal therapy and may also be transmitted to other individuals. Organisms from extradental sites are usually not affected by scaling and root planning (SRP) and other periodontal site-specific mechanical tooth-cleaning approaches but may be eradicated by appropriate systemic antibiotic therapy [207].

### 3.4 Microbiological Analysis

In clinical research and dental office settings, antibiotics should not be selected merely on the basis of clinical status, radiographic analysis, or limited microbiological testing. A risk of treating chemotherapeutically with insufficient information is failure to control the pathogens and/or overgrowth of new pathogens. The selection of antibiotics should be based on proper analysis of the composition of the subgingival microflora and the in vitro antimicrobial susceptibility of important organisms, as well as considerations to the medical status of the patient, potential side effects of the drug, and possible drug interactions [185].

Microbiology testing should be performed after completion of conventional mechanical therapy to assess the need for additional antibiotic treatment.

Microbial testing should be comprehensive enough to identify the most optimal drug regimen in case antibiotic therapy is warranted. Microbiology testing might be repeated 1–3 months after the antimicrobial therapy to verify the elimination or marked suppression of the pathogen(s) and to screen for possible superinfecting organisms. Ideally, the composition of the subgingival microbiota post treatment should be similar to that in periodontal health [207].

Microbial sampling sites can include individual pockets with recent disease activity or several pooled subgingival sites [207]. Mombelli et al. [128] showed that in some periodontitis patients the outcome of a test depends greatly upon the number of samples taken and upon the strategy of site selection; the most efficient method of sampling seems to be the selection of the deepest pocket in each quadrant of the mouth. If several pockets are equally deep, the most mesial is used. Subgingival samples can be collected with either sterile points, left in place for 10 s each, or a curette after isolating the area with cotton rolls and removing supragingival deposits with a scaler [207]. The evaluation of the periodontal microflora of patients who are not treated in the vicinity of the laboratory where the samples are analyzed requires information on the influence of the transport conditions on the survival of the bacteria. The survival of *A. actinomycetemcomitans*, *Porphyromonas gingivalis*, and *Prevotella intermedia* in three transport media (RTF, RTFFF, and VMGA III) was tested in conditions simulating the transit from the dental office to the laboratory. For transport within 4 h, all three transport media were equally effective, both at room temperature and at 4°C. After 24 and 48 h, a significant increase of total viable counts was found after storage in VMGA III (viability-maintaining microbiostatic medium, anaerobically prepared) at room temperature, which made this medium less useful as a transport medium. It was suggested, for quantitative recovery, that all media should be used within 4 h of storage [204].

For the detection of putative periodontal pathogens, molecular probes have been shown to provide an alternative approach to conventional culture and immunological methods. It was reported that DNA probe analysis was able to detect *P. gingivalis* even in culture-negative samples. Polymerase chain reaction was able to identify *P. gingivalis* collagenase genes in whole plaque samples, compared with the detection methods currently used that were able to target a potential

virulence factor for identification of pathogens [16]. Other techniques include polyclonal or monoclonal antibody assays and trypsin-like enzyme-produced assay. The disadvantages of these techniques are the limited number of species that can be detected and the inability to determine the antimicrobial sensitivity testing of target microorganisms [207]. Walker et al. [218] described an antibiotic susceptibility testing method which involves determining the in vitro effect of an antibiotic on dispensed samples of subgingival plaque as an aid in selecting an appropriate agent for use in the patient with advanced destructive periodontal disease.

### 3.5 Drug Interactions

Many antibiotics may interact with other drugs and cause clinically significant effects. Interaction occurs when one drug alters the other drug's pharmacokinetics with respect to absorption, distribution, metabolism, or excretion. The list of drug-drug interactions keeps expanding and is too long to be memorized. Patients on long-term medication for cardiovascular disease, asthma, seizures, or diabetes are at particularly high risk for drug-drug interactions [188] (Table 3.1)

### 3.6 Antibiotic Regimens in Periodontal Therapy

#### 3.6.1 Single Antibiotic Regimes

##### 3.6.1.1 Tetracycline-HCl

The tetracyclines, **tetracycline-HCl, doxycycline hyolate, and minocycline-HCl**, are broad-spectrum antibiotics active against both gram-positive and gram-negative bacteria. **Adverse reactions** to the tetracyclines are relatively minimal. Similar to any other broad-spectrum antibiotic, suppression of the normal flora may occur, followed by overgrowth of resistant organisms or colonization by exogenous pathogens such as *Candida*. Gastrointestinal disturbances are not uncommon and may result in nausea and/or diarrhea. Photosensitivity, manifested by exaggerated

sunburn, may occur. True allergic reactions are relatively rare [219].

**Structurally, tetracyclines consist of four fused rings, hence the name tetracyclines.** Tetracycline derivatives, primarily doxycycline and minocycline, differ from the parent compound by minor alterations of chemical constituents attached to the basic ring structure. These minor alterations in the molecular structure make both doxycycline and minocycline more lipophilic than the parent compound, resulting in better adsorption following systemic delivery and better penetration into the bacterial cell. Thus, lower and less frequent doses of doxycycline and minocycline can be given. For this reason and due to the widespread resistance to tetracycline-HCl, *doxycycline and minocycline tend to be the tetracyclines most commonly used* [219].

Tetracycline differs from minocycline and doxycycline in many pharmacokinetic characteristics. Although all tetracyclines are incompletely absorbed, the oral availability of tetracycline is considerably lower than that of minocycline or doxycycline. In addition, minocycline and doxycycline are not affected by concomitant food ingestion. Tetracycline differs from minocycline and doxycycline in that it has a higher percentage of urinary excretion, a lower percentage of plasma binding, clearance that is more rapid, a larger volume distribution, and a shorter half-life. It has been recognized for some time that individuals differ widely in their peak plasma levels of tetracyclines, which has been attributed to variability in absorption. Generally, a peak plasma concentration of 2–2.5 mg/mL occurs 2–4 h following oral administration of repeated doses of tetracycline. Oral administration of 200 mg doxycycline creates a peak plasma concentration of 3 mg/mL at 2 h. [174]. Ciancio et al. [33] showed gingival health improvement after the administration of minocycline over an 8-day period. *The concentration of minocycline in saliva was approximately 6% of serum concentrations in contrast to the observation that the concentration in gingival crevicular fluid (GCF) was approximately 500% of the serum concentrations* [33]. It was revealed that the most important single factor controlling the ability of the tetracyclines to establish an antibacterial concentration within the periodontal environment was the plasma concentration. In other words, if the drug was not well absorbed, it did not reach antibacterial levels in the periodontal pocket [174].

**Table 3.1** Antibiotic drug–drug interactions [188] (Reprinted with permission John Wiley & Sons)

| Antibiotic                                | Interacting drug   | Effect  | Clinical significance |
|---|--|---|-----------------------|
| Clindamycin                               | Antidiarrheal agents (kaolin)  | Decreased absorption of clindamycin   | Probable              |
|   | Muscle relaxants (diazepam)  | Increased frequency and duration of respiratory paralysis                           | Probable              |
|   | Erythromycin   | Mutual antagonism   | Probable              |
| Metronidazole                             | Barbiturates and hydantoins  | Decreased effectiveness of metronidazole  | Probable              |
|   | Oral anticoagulants (warfarin)   | Increased anticoagulant effect  | Definite              |
|   | Ethanol  | Disulfiram-like (AntabuseA) reaction  | Probable              |
|   | Disulfiram (AntabuseA)   | Acute toxic psychosis   | Probable              |
| Penicillins/amoxicillin                   | Probenecid   | Increased levels of penicillins   | Probable              |
| Tetracyclines/doxycycline                 | Antacids, aluminum, bismuth iron, Mg <sup>11</sup>   | Decreased absorption of tetracyclines due to chelation                              | Probable              |
|   | Barbiturates and hydantoins  | Decreased serum half-life of doxycycline  | Probable              |
|   | Carbamazepine (TegretolA)  | Decreased serum half-life of doxycycline  | Probable              |
|   | Digoxin  | Increased serum levels of digoxin   | Probable              |
| Erythromytics/azithromycin/clarithromycin | Carbamazepine  | Increased serum levels of carbamazepine: nystagmus, nausea, vomiting, and ataxia    | Definite              |
|   | Cisapride  | Increased cisapride concentration, with the risk of life-threatening arrhythmia     | Definite              |
|   | Cyclosporine   | Increased serum levels of cyclosporine, with toxic effects                          | Probable              |
|   | Methylprednisolone   | Increased steroid concentration   | Definite              |
|   | Nonsedating antihistamines (terfenadine, astemizole)   | Increased antihistamine concentration, with the risk of life-threatening arrhythmia | Definite              |
|   | Theophylline   | Increased serum levels of theophylline, with nausea, vomiting, seizures, and apnea  | Definite              |
|   | Oral anticoagulants (warfarin)   | Increased anticoagulant effect  | Probable              |
| Fluoroquinolones (ciprofloxacin)          | Cations (Al <sup>111</sup> , Ca <sup>11</sup> , Fe <sup>11</sup> , Mg <sup>11</sup> , Zn <sup>11</sup> ) in antacids, vitamins, and dairy products | Decreased absorption of fluoroquinolones due to chelation                           | Definite              |
|   | Caffeine   | Increased caffeine concentration  | Probable              |
|   | Cimetidine   | Increased serum levels of fluoroquinolones  | Probable              |
|   | Cyclosporine   | Increased serum levels of cyclosporine  | Probable              |
|   | Nonsteroidal anti-inflammatory drugs   | Increased risk of central nervous system stimulation                                | Definite              |
|   | Probenecid   | Decreased ciprofloxacin clearance   | Probable              |
|   | Sucralfate   | Decreased absorption of fluoroquinolones  | Definite              |
|   | Theophylline   | Increased serum levels of theophylline  | Definite              |
|   | Oral anticoagulants (warfarin)   | Increased anticoagulant effect  | Probable              |

Tetracyclines are **bacteriostatic** and possess high in vitro activity against several suspected periodontal pathogens, including *A. actinomycetemcomitans*. It appears to **concentrate in the periodontal pocket**, and its antibacterial efficacy may be prolonged

through its ability to bind to both enamel and dentin surfaces. In addition, tetracyclines have been shown to **suppress collagenase activity in crevicular fluid and inflammatory cells and to inhibit osteoclast-mediated bone resorption in vitro** [138].

However, at high concentrations, such as those achieved with localized delivery of the antibiotic directly into the periodontal pocket, the tetracyclines may exert a **bactericidal effect** due to their ability to cause alterations in the cytoplasmic membrane. This may result in leakage of nucleotides and other components from the bacterial cell and result in its death [219].

**Resistance to the tetracyclines** is relatively common and is mediated by a number of genetic determinants that may be located on plasmids or on the bacterial chromosome. Resistance may occur due to the coding of an efflux pump that actively removes the drug from the bacterial cell so that sufficient drug concentration is never achieved within the cell. This is a common mechanism for the conveyance of resistance to tetracycline and, to a lesser extent, to doxycycline. Another mode of resistance is referred to as ribosome protection. With this mechanism, tetracycline antibiotics are not removed from the bacterial cell but are prevented from binding to the 30S ribosomal subunit. This mechanism generally conveys resistance equally to all tetracyclines [219]. *An overall increased frequency of tetracycline resistance has been recently reported in the medical and dental literature, especially, in countries with unrestricted antimicrobial use, and it is suggested that bacteriostatic drugs may not be suitable for treating biofilm infections [206, 226].* This may be one of the causes of treatment failure of periodontitis when using this drug, as reported by some investigators [8, 38, 126, 170, 223].

A number of clinical trials using various designs have been conducted to evaluate the **effectiveness of the adjunctive use of the tetracyclines in periodontal disease** (see Table 3.2).

Hayes et al. [75] performed a meta-analysis of 13 published studies, assessing the use of systemic tetracycline in the treatment of periodontal disease. The authors were unable to combine data from the majority of studies due to heterogeneity of the outcomes evaluated and limitations in data reported in the individual studies. Therefore, only two studies were included in the quantitative meta-analysis. On a scale of 0–1, the mean score for this group of studies was  $0.27 \pm 0.19$  for study protocol and  $0.31 \pm 0.11$  for data analysis and presentation. Mean reduction in PPD for the group treated with tetracycline plus scaling was 2.45 mm; for the group that received only scaling, 2.02 mm; for the group that received only tetracycline, 1.98 mm; and for the control group, 0.65 mm. It was concluded that data

from the published literature does not demonstrate that the use of systemic tetracycline is more beneficial than conventional treatment in the management of adult periodontal disease.

Early studies have shown controversial results, but they did not assess the pretreatment disease activity and the cultivable composition of the pocket microbiota in the study patients [91]. Several small-scale clinical trials, evaluating the **efficacy of tetracycline as an adjunct to SRP** in the treatment of **adult periodontitis**, failed to demonstrate statistically significant differences compared to SRP alone. However, mean PPD and AL were slightly improved in the tetracycline group [219].

Tetracycline therapy, when used in conjunction with surgery or root planning has been shown to be effective in controlling the progression of **juvenile periodontitis** [132, 138, 147, 179, 182, 226]. Results obtained following adjunctive use of tetracycline in the treatment of localized aggressive periodontitis were often clinically profound. Such improvements were likely due to the elimination or severe repression of *A. actinomycetemcomitans* in the infected site [219]. Christersson et al. [32] showed that systemic tetracycline-HCl alone has beneficial effects on clinical parameters in localized juvenile periodontitis patients and can suppress *A. actinomycetemcomitans*. However, *A. actinomycetemcomitans* was suppressed below detectable levels in only approximately 50% of the lesions even following an 8-week tetracycline administration [32]. Following SRP with systemic antibiotic therapy Kornman et al. [90] reported substantially reduced bacterial species, *A. actinomycetemcomitans*, *P. intermedia*, and *E. corrodens* in 236 patients with moderate to severe periodontitis. Additional studies have demonstrated that both doxycycline and minocycline, like tetracycline, may significantly suppress *A. actinomycetemcomitans* but not totally eradicate the bacterium from all sites [112, 113, 219].

Double-blind clinical studies enrolling patients characterized as having **refractory or recurrent periodontitis** found systemic tetracycline and doxycycline, in conjunction with SRP, significantly reduced PPD and resulted in increased attachment gain relative to SRP and placebo [92, 118, 120, 217].

In **periodontal abscesses** treated with drainage and supragingival scaling (SGS) in addition to systemic tetracycline demonstrated reduced PPD and bleeding on probing (BOP) as well as gain of AL after 6 months.

**Table 3.2** Clinical studies of systemic tetracycline therapy in periodontal disease

| Study                | No. patients             | Periodontal condition                | Study period | Periodontal treatment  | Outcome  |
|----------------------|--------------------------|--------------------------------------|--------------|--|--|
| Akalin et al. [4]    | 45                       | Chronic periodontitis                | 7 weeks      | 1. Systemic DOXY (200 mg for the first day, then 100 mg once daily for 14 days)±SRP (N=15)<br>2. DOXY local ±SRP (N=15)<br>3. SRP alone (N=15)   | There was no significant difference when comparing systemic DOXY+SRP versus SRP alone treatment ( $P>0.05$ ) (mean PPD change: 1.91 vs 1.72; Mean CAL change: 1.21 vs 1.10; Mean GI change: 0.66 vs 0.54; Mean SBI change: 0.84 vs 0.85; Mean PI: 0.73 vs 0.40). The systemic DOXY group showed lower PPD reduction than SRP group (1.13 vs 1.72, $P<0.05$ ), lower CAL gain (0.69 vs 1.10) and SBI change (0.69 vs 0.85). No significant difference was found between local DOXY and SRP treatments. In summary, the local DOXY alone treatment seemed more effective than systemic DOXY alone treatment on PPD reduction, but no significant difference was found between them when combined with the SRP.   |
| Al-Joburi et al. [5] | 96 at baseline, 79 final | Advanced adult chronic periodontitis | 24 weeks     | 1. SRP+spiramycin (14 UI, 2x, 14d)<br>2. SRP+TET (250, 4x, 14d)<br>3. SRP+placebo  | While the proportion of spirochetes were significantly decreased ( $P<0.05$ ) at 2- and 8-week intervals in both tetracycline and spiramycin groups (26% to 0.04% and 28% to 0.04%, respectively), compared to the placebo group (30% to 7%), only in the spiramycin group was the proportion of spirochetes significantly lower than the placebo group at the 24-week interval (3% and 11%, respectively). At week 24, the proportion of spirochetes in the tetracycline group had rebounded to 7%, which was not significantly different from the placebo group.   |
| Feres et al. [55]    | 20                       | Adult periodontitis                  | 90 days      | 1. SRP+DOXY (100, 1x, 14d) (N=10)<br>2. SRP (N=10)   | A modest but significant reduction in mean PPD from baseline to 90 days occurred in both test and control groups. A significant decrease in the percentage of sites with gingival redness occurred in the test group. There were no significant differences in proportions between test and control groups for 33 of the test species at any time point. Test subjects exhibited lower proportions of four <i>Actinomyces</i> species and an increase in three <i>Streptococcus</i> species during antibiotic administration. Periodontal pathogens including <i>B. forsythus</i> , <i>P. gingivalis</i> , <i>T. denticola</i> , and <i>A. actinomycetemcomitans</i> were not significantly altered by oral administration of DOXY using conventional therapeutic dosage.  |
| Haffajee et al. [69] | 98                       | Severe periodontal disease           | 10 months    | 1. Periodontal surgery (modified Widman flap) + AMOX/CLAV (250, 3x, 30d)<br>2. Periodontal surgery (modified Widman flap) + TET (250, 4x, 30d)<br>3. Periodontal surgery (modified Widman flap) + ibuprofen<br>4. Periodontal surgery (modified Widman flap) + placebo | Subjects receiving antibiotics exhibited significantly more attachment level “gain” ( $0.57 \pm 0.15$ mm) than subjects receiving either ibuprofen or a placebo ( $0.02 \pm 0.10$ mm). The differences between AMO/CLAV and TET groups were not significant, nor were the differences between ibuprofen and placebo. Subjects receiving systemically administered antibiotics had a greater decrease in the number of sites colonized by <i>P. gingivalis</i> , <i>B. forsythus</i> , <i>P. intermedia</i> , and <i>P. micros</i> post therapy than subjects not receiving antibiotics. In summary, the results of this investigation indicate that adjunctive systemic antibiotics increase periodontal attachment “gain” and decrease the levels of some suspected periodontal pathogens in subjects with evidence of current disease progression. |

(continued)

**Table 3.2** (continued)

| Study                   | No. patients | Periodontal condition   | Study period | Periodontal treatment  | Outcome  |
|-------------------------|--------------|---|--------------|--|--|
| Kulkarni et al. [92]    | 27           | Recurrent periodontitis on maintenance  | 7 months     | 1. SRP every 2 months+DOXY (200 mg to start and 100 mg/day for 3 weeks)<br>2. SRP every 2 months+placebo | Based on presence or absence analysis of the sum scores of the six pathogens (Aa, Pg, Bi, Ec, Fn and Sp) in dental plaque, both the placebo and the DOXY groups exhibited similar scores at the time of detection of active disease (mean placebo: $2.38 \pm 0.32$ ; mean DOX: $2.95 \pm 0.27$ , $P < 0.05$ ). One week after treatment, the probability of detection was unchanged in the placebo group ( $3.14 \pm 0.47$ ), but was significantly reduced in the doxycycline group ( $1.77 \pm 0.26$ ; $P = 0.0002$ ).   |
| Lindhe et al. [96]      | 14           | Advanced adult periodontitis  | 50 weeks     | 1. SRP+placebo<br>2. TET (250 mg, 4x, 14d and after that 250 mg, 1x, 48 weeks)                           | Long-term TET therapy in the absence of SRP resulted in the establishment of a subgingival microbiota almost devoid of motile bacteria and in markedly reduced signs of gingivitis, PPD, and CAL. In summary, the alterations observed as a result of tetracycline administration to patients with excellent self-performed plaque control were similar to those obtained by conventional SRP in the control group.  |
| Matisko & Bissada [118] | 11           | Recurrent/progressive periodontitis demonstrating subgingival infection with A.a. and/or P.g. | 25 weeks     | 1. SRP+DOXY (200 mg the first day and 100 mg for 4d)+AMOX/CLAV (500, 3x, 5d)<br>2. SRP+DOXY (10d)        | The DOXY+AMOX/CLAV groups produced significant reduction in PPD at 4, 12, and 25 weeks (1.1, 1.3, and 1.1 mm, respectively). The DOXY group produced significant reduction in PPD only at 4 and 12 weeks (0.8 and 0.8 mm); the DOXY + AMOX/CLAV group produced significant CAL gain of 0.8 mm at 4 and 12 weeks; and the DOXY + AMOX/CLAV group produced the most sustained reduction in PPD and gain in PAL. In summary, these findings suggest that the sequential use of multiple antibiotic agents may offer greater promise as an adjunctive treatment approach for the management of recurrent and/or progressive periodontitis than a single antibiotic regimen |
| McCulloch et al. [119]  | 82           | Recurrent periodontitis   | 12 months    | 1. SRP+DOXY (100, 1x, 21d)<br>2. SRP+placebo   | Within 7 months, 15 out of 19 patients on placebo exhibited recurrent disease compared to 13 out of 29 patients on DOXY, a relative risk reduction of 43% ( $P < 0.05$ ) for DOXY compared to placebo. MIC of DOXY for subgingival plaque samples from active sites ranged between 25 and 100 $\mu\text{g}/\text{mL}$ , which is several fold higher than reported GCF concentrations for this drug. However, GCF collagenase was inhibited in vitro at concentrations of 5–10 $\mu\text{g}/\text{mL}$ DOXY. In summary, these data indicate that doxycycline provides significant risk reduction of recurrent periodontitis in patients with active disease.          |

|                     |    |  |           |   |  |
|---------------------|----|--|-----------|---|--|
| Müller et al. [132] | 33 | Juvenile and severe generalized periodontitis      | 2 years   | 1. SRP + MIN (200, 1x, 21d)<br>2. Surgery (modified Widman flap) + MIN (200, 1x, 21d)   | Regarding the GP group, 6 months after active treatment, <i>Aa</i> was present in 6/9 patients and 50% of sites in severe cases and 3/6 patients and 46% of sites in moderate periodontitis patients. In contrast, the organism was virtually eliminated by scaling and flap procedures in the localized periodontitis clusters, and did not reappear after 6 months ( $P < 0.05$ ). Combined antibiotic, mechanical, and surgical therapy resulted in a persistence of 20% of sites with residual periodontal probing depth of $\geq 4$ mm in moderate periodontitis patients after active therapy. In summary, it was suggested that the applied therapy would be appropriate in localized forms of <i>Aa</i> periodontitis, but inappropriate in more severe and generalized forms to predictably eliminate <i>Aa</i> . |
| Ng & Bissada [135]  | 32 | Generalized moderate periodontitis                 | 24 weeks  | 1. SRP + DOXY (200 mg the first day followed by 100 mg/day, 6 weeks)<br>2. SRP + ibuprofen (800 mg/day, 6 weeks)<br>3. SRP + DOXY + ibuprofen<br>4. SRP + placebo | A statistical significance ( $P < 0.05$ ) from baseline data in: (1) gains of 0.4 mm and 0.5 mm of CAL for groups 1 and 3, respectively; (2) reduction of 0.7 mm PPD for group 3; (3) reduction of 0.4 and 0.1 GI scores for groups 1 and 3, respectively; and (4) gain of 0.5 mm CAL and reductions of 0.4 mm PPD and 0.2 GI score for the SRP group when compared to the no SRP group at 24 weeks. In summary, it was concluded that the adjunctive use of systemic doxycycline alone or in combination with ibuprofen results in a statistically significant, yet modest clinical, improvement beyond that obtained by SRP.   |
| Palmer et al. [147] | 38 | Localized or generalized early onset periodontitis | 12 months | 1. OHI + SRP + TET(250, 4x, 14d) followed by SURG+CHX + TET(250, 4x, 14d)<br>2. OHI + SRP + placebo followed by SURG+CHX + placebo                                | Improvements in PPD, CAL, and BOP were significantly better in the group treated with adjunctive tetracycline, at 3 months post treatment. At 12 months, in the test group, 58% of the originally affected teeth required surgery (modified Widman flap) compared to 75% in the control group. In summary, there were no further statistically significant differences between test and control groups for any of the clinical measures, although the tetracycline group appeared to maintain an advantage. In summary, systemically administered tetracycline is a useful adjunct in the management of early-onset periodontitis, particularly in nonsurgical treatment.  |

(continued)

**Table 3.2** (continued)

| Study                   | No. patients | Periodontal condition  | Study period | Periodontal treatment   | Outcome  |
|-------------------------|--------------|--|--------------|---|--|
| Ramberg et al. [160]    | 115          | Advanced periodontitis   | 13 years     | 1. SRP+TET (250, 4x, 21d) (N=35)<br>2. SRP (N=80)   | At the re-examination 1 year after active therapy, both treatments produced a statistically significant reduction (1) in the percentage of plaque harboring surfaces (-26% and -23%) and BOP sites (-49% and -38%), (2) in the mean PPD values (-1.0 mm and -0.7 mm); and (3) in the percentage of deep pockets (-11% and -8%). In addition, in both groups there was a significant gain of the CAL; test group=0.47 mm, control group = 0.16 mm. In this time interval, the reduction of the individual mean CAL values was more pronounced in the test group than in the controls ( $P<0.001$ ). Re-examinations after 3, 5, and 13 years of supportive periodontal therapy disclosed that this short-term benefit was not maintained in the longer perspective. In summary, the beneficial effect of systemically administered tetracycline on CAL occurred in the first year post therapy. |
| Rams & Keyes [162]      | 21           | Progressive periodontitis in adults  | 11 months    | 1. SRP+TET (250, 4x, 14d)<br>2. SRP   | After 2 weeks tetracycline HCl significantly reduced elevated levels of spirochetes, motile rods, and crevicular leukocytes to low or undetectable levels, whereas levels in the placebo subjects remained generally unchanged. In summary, the results clearly demonstrate the value of tetracycline HCl as an adjunct to periodontal therapy in reducing remaining suspected periodontopathic bacterial populations in advanced lesions after local therapy of scaling, root planning, and topically applied chemotherapy.   |
| Saxén et al. [177]      | 14           | Localized juvenile periodontitis   | 20 months    | 1. SRP+DOXY (200 mg for the first day, then 100 mg once daily for 14 days)<br>2. SRP  | The only significant difference between the groups was a greater reduction in the prevalence of <i>A. actinomycetemcomitans</i> 8 months after treatment with doxycycline as compared with the placebo.  |
| Saxén & Asikainen [179] | 27           | <i>A. actinomycetemcomitans</i> -positive localized juvenile periodontitis | 18 months    | 1. SRP+OHI+periodontal surgery+MET (200, 3x, 10d) (N=9)<br>2. SRP+OHI+periodontal surgery+TET (250, 4x, 12d) (N=9)<br>3. SRP+OHI+periodontal surgery. No medication (N=9) | By the end of the study, <i>A. actinomycetemcomitans</i> was suppressed to below detection level at all test sites only in the metronidazole group, at 17/26 sites (four patients) in the tetracycline group, and at 19/26 sites (six patients) in the control group. In summary, clinically, all groups showed improvement. In summary, metronidazole was more effective than tetracycline in the suppression of <i>Aa</i> and the suppression of <i>Aa</i> appeared to produce better clinical results.  |

|                           |    |   |           |   |   |
|---------------------------|----|---|-----------|---|---|
| Sigusch et al. [182]      | 48 | Generalized rapidly progressive periodontitis | 24 months | 1. 2-step SRP+DOXY (200 mg, 1x, 8d) (N=12)<br>2. 2-step SRP+MET (500 mg, 2x, 8d) (N=15)<br>3. 2-step SRP+CLIN (150 mg, 4x, 8d) (N=11)<br>4. 2-step SRP; no antibiotic treatment (N=10)  | 6 months after the second step SRP, the clindamycin group showed a significantly great reduction of PPD from baseline ( $3.5 \pm 0.96$ mm from $5.7 \pm 1.06$ mm) compared with control ( $4.6 \pm 1.0$ mm from $5.9 \pm 0.70$ mm). In PPD site categories 6–9 mm and >9 mm the clindamycin treatment induced a PPD reduction of 4.2 mm ( $4.2 \pm 1.06$ mm from $8.4 \pm 0.76$ mm) compared with control ( $5.9 \pm 1.19$ mm from $8.2 \pm 1.03$ mm). A significantly high CAL gain was also noted in the clindamycin group ( $4.4 \pm 1.0$ mm from $6.1 \pm 0.96$ mm) compared with controls ( $5.7 \pm 0.96$ mm from $6.3 \pm 0.77$ mm). SBI decreased most in the metronidazole and clindamycin groups. <i>P. gingivalis</i> and <i>A. a.</i> were almost completely eradicated in these 2 groups 24 months after SRP. In addition, the phagocytotic capacity of crevicular polymorphonuclear neutrophils was increased in groups 2 and 3 after the second step. In summary, the present results show that metronidazole and clindamycin are effective antibiotics when used adjunctively in a two-step nonsurgical procedure of scaling and root planning (SRP) in RPP patients.   |
| Xajigeorgiou et al. [226] | 43 | Generalized aggressive periodontitis          | 6 months  | 1. SRP+debridement (ultrasonics and polishing with a rubber cup)+A/M (AMO 500, 3x, 7d+MET 500, 3x, 7d) (N=10)<br>2. SRP+debridement (ultrasonics and polishing with a rubber cup)++DOXY (200 mg of doxycycline as a loading dose and 100 mg/day 14 days) (N=10)<br>3. SRP+debridement (ultrasonics and polishing with a rubber cup)++MET (500, 3x, 7d) (N=12)<br>4. Only SRP+debridement (ultrasonics and polishing with a rubber cup) (N=11) | No differences were observed between the four groups at any time point regarding PPD, CAL, and BOP reduction. Subjects who received adjunctive metronidazole displayed a statistically significant reduction in mean PPD after antibiotic intake (MET): baseline: $4.71 \pm 0.57$ ; 6 weeks after SRP: $3.47 \pm 0.51$ ; 6 months: $2.86 \pm 0.65$ , $P < 0.05$ . The reduction in mean PPD for the DOXY group was: baseline: $4.24 \pm 0.57$ ; 6 weeks after SRP: $3.48 \pm 0.67$ ; 6 months: $3.35 \pm 0.76$ , $P < 0.05$ . CAL change for the DOXY group was: baseline: $5.03 \pm 1.42$ ; 6 weeks after SRP: $4.43 \pm 1.73$ ; 6 months: $4.22 \pm 1.9$ , $P < 0.05$ . The BOP of the DOXY group during the experimental period was baseline: $0.81 \pm 0.25$ , 6 weeks after SRP: $0.24 \pm 0.23$ , 6 months: $0.14 \pm 0.22$ , $P < 0.05$ . The intake of A/M and MET, resulted in significant reduction of the percentage of sites $\geq 6$ mm compared with the control group at 6 months (80% reduction for A/M, 87.7% for MET, 57.7% for controls). No differences were observed between the subjects who received adjunctive doxycycline compared with control subjects (64.2% change from baseline vs 57.7% in controls). A/M or MET alone (when <i>A.actinomycetemcomitans</i> is not involved) was effective in deep pockets of aggressive periodontitis patients. In summary, adjunctive metronidazole plus amoxicillin or metronidazole alone (when <i>A.actinomycetemcomitans</i> is not involved) is effective in deep pockets of aggressive periodontitis patients. |

(continued)

**Table 3.2** (continued)

| Study               | No. patients | Periodontal condition                | Study period | Periodontal treatment   | Outcome  |
|---------------------|--------------|--------------------------------------|--------------|---|--|
| Rooney et al. [171] | 62           | Advanced chronic periodontal disease | 6 months     | 1. AM: SRP+AMO 250, 3x, 7d and MET 200, 3x, 7d (N=15)<br>2. PM: lactate capsules and MET (MET 200, 3x, 7d) (N=16)<br>3. AP: amoxicillin and calcium lactate (N=16)<br>4. PP: lactate and calcium lactate (N=15) | <p>PPD improved in all groups. Treatment effects were highly significantly different and always greatest in the AM and least in the PP groups. Benefits of PM and AP over PP were also noted. The mean percentage of sites with high (&gt;6 mm) PPD reduction in the four treatment groups at 6 months compared with baseline were: AM: 1.3% from 15.9%, PM: 4.8% from 15.6%, AP: 3.8% from 14.6%, PP: 12.4% from 19.3%). CAL improved in all groups and showed the same highly significant treatment differences, again favoring AM. The mean percentage of sites with high (&gt;6 mm) attachment loss reduction in the four treatment groups at 6 months compared with baseline: AM 6.5% from 17.1%, PM: 8.8% from 18.2%, AP 10.0% from 18.7%, and PP 18.2% from 24.3%). BOP improved in all groups, particularly in AM compared to the other groups: AM 22.8% from 62.6%, PM 32.5% from 61.8%, AP 33.9% from 61.8%, PP 44.9% from 65.6%. Regarding total anaerobic and aerobic counts, the only significant difference between treatments was at 1 month where the combined treatment was significantly more effective against total anaerobic counts than the double placebo and MET and placebo. <i>P. intermedia</i> counts were always lower in active groups compared to PP and reached significance for AM and AP at 1 month and AM and PM at 3 months. In summary, the significant differences among treatment groups and the overall trend in the data, in line with other studies, support the considerable adjunctive benefits to SRP of amoxicillin and MET combined in the treatment of advanced chronic periodontal disease.</p> |

SRP scaling and root planning, PPD periodontal probing depth, CAL clinical attachment level, BOP bleeding on probing, BI gingival index, GCF gingival crevicular fluid, Aa *Aggregibacter actinomycetemcomitans* (previously *Actinobacillus actinomycetemcomitans*), Pg *P. gingivalis*, Pi *Prevotella intermedia*, OHl oral hygiene instruction, SBI Sulcus Bleeding Index, CHX chlorhexidine, MET Metronidazole, AMO Amoxicillin, AM Amoxicillin and Metronidazole, DOXY Doxycycline, AZ Azithromycin, CLIN Clindamycin, MIN Minocycline, 3x three times per day, 7d 7 days, Ec *Eikenella corrodens*, Fn *Fusobacterium nucleatum*

*P. gingivalis* and *P. intermedia* were significantly reduced in number and proportions during the follow-up period of 6 months [74].

Checchi et al. [27] suggested that there is no reason for **routine tetracycline prophylaxis in periodontal surgery** (ostectomy, osteoplasty, or tooth extraction) as no statistically significant difference in the incidence of post-operative infection could be detected between patients who were given tetracycline and those who were not.

### 3.6.1.2 Collagenase Inhibitors (Modulation of MMPs)

The matrix metalloproteinases (MMPs) are a family of structurally related but genetically distinct enzymes that degrade extracellular matrix (ECM) and basement membrane (BM) components. This group of 23 human enzymes is classified into collagenases, gelatinases, stromelysins, membrane-type MMPs, and other MMPs, mainly based on the substrate specificity and molecular structure. MMPs are involved in physiological processes such as tissue development, remodeling, and wound healing [197, 203], and play important roles in the regulation of cellular communication, molecular shedding, and immune functions by processing bioactive molecules including cell surface receptors, cytokines, hormones, defensins, adhesion molecules, and growth factors. MMP activity is controlled by changes in the delicate balance between the expression and synthesis of MMPs and their major endogenous inhibitors, tissue inhibitors of matrix metalloproteinases (TIMPs). The catalytic competence of MMPs is controlled through the activation of proenzymes, and the inhibition of the activation or activity by TIMPs [196, 197, 203].

As the roles of MMPs in tissue-degenerative diseases have become evident, attempts to control their activities by pharmacological means have gained much attention. Although the exact roles of individual MMPs in various diseases are not fully understood, it is clear that MMPs are often up-regulated in groups forming activation cascades both in the inflammatory and malignant diseases [196, 197, 203].

MMP activation and activity can be controlled by inhibition in several ways: proteolytic degradation and inactivation, nonspecific endogenous inhibitors such as

$\alpha_2$ -macroglobulin, and especially specific tissue inhibitors of MMPs, TIMPs. Currently, four TIMPs (TIMP 1–4) are known to be expressed in vertebrates. TIMPs inhibit MMPs by forming 1:1 stoichiometric enzyme–inhibitor complexes. TIMP-1, -2, and -4 are secreted, while TIMP-3 is sequestered to the ECM. The substrate specificity of TIMPs varies [17, 18, 196, 197].

Synthetic inhibition of MMPs offers an interesting possibility to control MMP-related diseases in which extensive tissue destruction is involved. One approach in MMP inhibition is aimed at chelation of the enzyme's active site,  $Zn^{2+}$  ion. The first MMP inhibitors to enter clinical trials in tumor treatment, batimastat and marimastat, base their MMP inhibitory effect on chelation. Tetracyclines and their non-antimicrobial analogs, chemically modified tetracyclines (CMTs), inhibit MMPs through several mechanisms. In addition to  $Zn^{2+}$  chelation, they can downregulate MMP mRNA expression, interfere with the protein processing during activation, and render the MMPs more susceptible for degradation [196, 197].

Minocycline, doxycycline, and tetracycline were all shown to inhibit collagenolytic activity, whereas non-tetracycline antibiotics had no effect on collagenase levels. **Doxycycline downregulates collagenolytic activity by several synergistic mechanisms.**

The first mechanism proposed was the ability of tetracyclines to inhibit already active MMPs (*collagenase and gelatinase*) in the ECM, a mechanism found to be associated with the  $Zn^{2+}$  or  $Ca^{2+}$  binding properties of the tetracycline molecule. It was suggested that the tetracyclines may bind to the secondary  $Zn^{2+}$  (and to a lesser extent,  $Ca^{2+}$ ) in collagenase, thus altering the conformation of the enzyme molecule and blocking its catalytic activity in the ECM. Additional inhibitory mechanisms of these drugs include their ability to prevent the conversion of pro-MMPs in the ECM into active MMPs. In addition, tetracyclines are known to scavenge for, and inhibit, the production of polymorphonuclear (PMN)-derived reactive oxygen metabolites, including hypochlorous acid (HOCl). This ability may further contribute to the non-antimicrobial, anti-inflammatory properties of doxycycline by inhibiting HOCl from activating latent pro-MMPs. Furthermore, HOCl oxidizes and inactivates host-derived proteinase inhibitors  $\alpha_1$ -PI and  $\alpha_2$ -macroglobulin (inhibitors of MMPs). Thus, the ability of tetracyclines to directly inhibit MMP activity and also scavenge for, and inhibit, reactive oxygen metabolites such as HOCl, represents

an important pathway for modulation of the destructive connective tissue events that occur in periodontitis [152, 153].

*Tetracyclines appear to inhibit ECM breakdown by indirect mechanisms as well.* In this regard, the serum protein  $\alpha$ 1-antitrypsin (also called  $\alpha$ 1-proteinase inhibitor) is the host's major defense against another family of tissue-destructive proteinases, the serine proteinases (particularly PMN leukocyte elastase). MMPs are now known to degrade and inactivate  $\alpha$ 1-antitrypsin, so that tetracycline inhibition of the MMPs could protect elastase-susceptible substrates (such as elastic fibers, fibronectin, proteoglycans, and TIMPs) from proteolytic attack as well. Another potential indirect mechanism by which the tetracyclines may inhibit ECM breakdown could be through *inhibition of activation of pro-tumor necrosis factor- $\alpha$* , thereby leading to a decrease in the formation of the powerful cytokine, tumor necrosis factor- $\alpha$  [152]. Doxycycline also contributes to decreased connective tissue breakdown by *downregulating the expression of pro-inflammatory mediators and cytokines* (including IL-1 and TNF- $\alpha$ ), and increasing collagen production, osteoblast activity, and bone formation. Tetracyclines have also been shown to normalize collagen formation in diabetic rats with previously suppressed collagen synthesis [172].

In 1998 the Food and Drug Administration approved Periostat® (from CollaGenex) for use in the USA. Periostat® contains 20 mg of doxycycline in tablets to be taken orally twice a day for at least 3 months as adjunctive therapy in patients with chronic periodontitis (CP) undergoing the initial phase of periodontal treatment (SRP) [64].

The rationale for using doxycycline at sub-antimicrobial doses as a host response modulator is that it inhibits the activity of MMPs by a variety of synergistic mechanisms independent of any antibiotic properties, as summarized by Walker (2008):

- Direct inhibition of active MMPs by cation chelation (dependent on  $\text{Ca}^{2+}$  and  $\text{Zn}^{2+}$ -binding properties).
- Inhibits oxidative activation of latent MMPs (independent of cation-binding properties).
- Downregulates expression of key inflammatory cytokines (interleukin-1, interleukin-6, and tumor necrosis factor- $\alpha$ ) and prostaglandin E<sub>2</sub>.
- Scavenges and inhibits production of reactive oxygen species produced by neutrophils.

- Inhibits MMPs and reactive oxygen species, thereby protecting  $\alpha$ 1-proteinase inhibitor, and thus indirectly reducing tissue proteinase activity.
- Stimulates fibroblast collagen production.
- Reduces osteoclast activity and bone resorption.
- Inhibits osteoclast MMPs.

### Animal Studies

Recent studies using the experimentally induced periodontitis model in rats and hamsters have investigated the effects of MMP inhibition with sub-antimicrobial doses of tetracyclines [14, 21, 22, 98, 106, 159]. A summary of the findings of these experiments is provided in Table 3.3.

### Clinical Studies

A series of double-blind, placebo-controlled clinical trials has demonstrated that sub-antimicrobial dose doxycycline (SDD), 20 mg twice a day (Periostat), produces an improvement in clinical indices (Table 3.4), in both smokers and nonsmokers [155]. A post-treatment SDD effectiveness was demonstrated, reflected by sustained improvements in periodontal health arising from reductions in the bacterial load (due to SRP) and the suppression of tissue-destructive host responses (due to SDD). Additionally, the substantive property of doxycycline (i.e., tendency to bind to the calcified surfaces of tooth roots and alveolar bone) and its gradual release in active form also may contribute to the maintained effectiveness of SDD post treatment [25].

Two of the larger clinical trials that investigated the efficacy of adjunctive SDD in the treatment of chronic periodontitis had similar study design and patient inclusion/exclusion criteria [26, 153]. This permitted Preshaw et al. [152] to perform a meta-analysis of the data gathered in the two studies. The adjusted least-square mean changes in probing depths and attachment levels from baseline for moderately diseased and severely diseased tooth sites are shown in Figs. 3.1–3.3. Smoking status was included as a factor in the analyses. It is clear from this meta-analysis that at each time point, adjunctive SDD resulted in statistically significantly greater mean probing depth reductions and attachment levels gains compared with SRP alone ( $P < 0.05$  in all cases). The

**Table 3.3** Animal studies on the effects of LDD on MMP production and ABL

| References              | Experimental periodontitis model   | Animal | Study treatment  | Observation period | Results   |
|-------------------------|--|--------|--|--------------------|---|
| Llavaneras et al. [98]  | Intra-gingival LPS-induced experimental periodontitis                              | Rats   | Oral administration of doxycycline (CMT-8) (1 mg/day, 7d) alone or in combination with a bisphosphonate (Clodronate) (1 mg/week, 7d) | 7 days             | Treatment of the LPS-injected rats with suboptimal CMT-8 alone or suboptimal clodronate alone produced slight reductions in the tissue-destructive proteinases (MMP-8, MMP-9, elastase) and no significant reductions in ABL. However, a combination of suboptimal CMT-8 and clodronate “normalized” the pathologically elevated levels of MMPs, elastase, and ABL, indicating synergistic inhibition of tissue breakdown in this animal model of periodontitis.  |
| Bezerra et al. [14]     | Ligature-induced experimental periodontitis  | Rats   | Daily subcutaneous doxycycline (2.5, 5 or 10 mg/kg)  | 7 days             | Nontreated rats displayed significant ABL, severe mononuclear cell influx and increase in osteoclast numbers, which were significantly reduced by 5 or 10 mg/kg/day-1 doxycycline.  |
| Ramamurthy et al. [159] | Intra-gingival LPS-induced experimental periodontitis                              | Rats   | Doxycycline and 5 different CMTs (2 mg/day orally)   | 6 days             | All six tetracyclines inhibited periodontal breakdown in the following order of efficacy: CMT-8 > CMT-1 > CMT-3 > doxycycline > CMT4 > CMT-7. Immunohistochemistry was positive for TNF, IL-1, and IL-6 in the inflammatory cells from untreated endotoxin rat tissues, whereas treatment with CMTs decreased the number of immuno-positive stained cells for cytokines and MMPs.   |
| Buduneli et al. [21]    | Intra-gingival LPS-induced experimental periodontitis                              | Rats   | Systemic administration of LDD alone or in combination with alendronate  | 7 days             | Significant reduction of gingival tissue levels of PGE <sub>2</sub> , LT <sub>B4</sub> , and PAF by low-dose doxycycline administration alone. Combined administration of doxycycline and alendronate exhibited the most prominent inhibition on gingival tissue levels of PGE <sub>2</sub> and PGF <sub>2</sub> alpha ( $P < 0.05$ ). Doxycycline + alendronate + LPS group also significantly reduced LT <sub>B4</sub> and PAF levels, although doxycycline provided the most reduction in the levels of these mediators ( $P < 0.05$ ).  |
| Buduneli et al. [20]    | Repeated injection of purified LPS derived from <i>Escherichia coli</i> endotoxin. | Rats   | Systemic administration of LDD and a bisphosphonate, alendronate   | 7 days             | Morphometric measurements revealed significantly more ABL in the LPS group compared to the saline control group ( $P < 0.05$ ). Alendronate revealed slight inhibition on ABL either alone or in combination with doxycycline (0.41 mm in alendronate and combined drug treatment groups versus 0.45 mm in LPS and doxycycline groups). Significantly higher IL-1beta levels were observed with alendronate either alone or in combination with doxycycline than in the LPS group ( $P < 0.05$ ). Combined administration of doxycycline and alendronate showed significantly higher levels of osteocalcin than all of the other groups ( $P < 0.01$ ). |

(continued)

**Table 3.3** (continued)

| References           | Experimental periodontitis model                                      | Animal | Study treatment   | Observation period | Results   |
|----------------------|---|--------|---|--------------------|---|
| Buduneli et al. [22] | Intra-gingival LPS-induced experimental periodontitis                 | Rats   | Systemic administration of saline control, LPS, LPS + doxycycline, LPS + alendronate, and LPS + doxycycline + alendronate | 7 days             | Individual administration of doxycycline or alendronate significantly decreased the expression of MMP-8 compared to LPS ( $P = 0.01$ ). Combined drug administration reduced MMP-14 significantly compared to doxycycline ( $P = 0.004$ ). MMP-14 significantly correlated with the amount of ABL in the LPS + doxycycline + alendronate group ( $P = 0.03$ ).  |
| Madan et al. [106]   | Weekly inoculation with <i>Porphyromonas gingivalis</i> ( <i>Pg</i> ) | Mice   | Intrapitoneal injection of LDD (1.5 µg/kg) performed right after injecting <i>Pg</i> (once a week)                        | 14 weeks           | Doxycycline treatment resulted in a reduction of mean lesions from 10.5 ± 0.49 to 1.09 ± 0.102% ( $P < 0.05$ ) at 14 weeks and a reduction from 21.5 ± 6.49 to 8.26 ± 0.162% ( $P = 0.106$ ) at 24 weeks. Chow-fed <i>Pg</i> mice treated with doxycycline also resulted in a reduction from 0.62 ± 0.128 to 0.0 ± 0.0% ( $P < 0.05$ ) at 14 weeks and a reduction from 0.92 ± 0.23 to 0.0 ± 0.0% ( $P < 0.05$ ) at 24 weeks. Administration of doxycycline to mice fed a high fat diet and <i>Pg</i> -inoculated resulted in a reduction of mean percentage of atheromatous lesions from 16.46 ± 1.69 to 1.141 ± 0.23% ( $P < 0.05$ ) at 14 weeks and a reduction from 25.27 ± 1.734 to 0.428 ± 0.033% ( $P < 0.05$ ) at 24 weeks. Consistent with the role of doxycycline on matrix proteases, at 24 weeks MMP-9 serum levels were markedly reduced by 60% ( $P < 0.05$ ) and 30% ( $P < 0.05$ ) with doxycycline treatment in <i>Pg</i> -infected high-fat and chow-diet groups, respectively. |

LDD low-dose doxycycline, MMP matrix metalloproteinases, TIMP endogenous tissue inhibitors of metalloproteinases, IL interleukin, LPS lipopolysaccharide, ABL alveolar bone loss

**Table 3.4** Clinical studies of sub-antimicrobial dose doxycycline (SDD) therapy in periodontal disease

| Study               | No. patients | Periodontal condition | Study period | Periodontal treatment   | Outcome  |
|---------------------|--------------|-----------------------|--------------|---|--|
| Caton et al. [26]   | 183          | Adult periodontitis   | 9 months     | 1. SRP + SDD (20, 2x, 9 months) ( $N=90$ )<br>2. SRP + placebo ( $N=93$ ) | The percentage of tooth sites with CAL gain $\geq 2$ mm was greater in the adjunctive SDD group than in the adjunctive placebo group at all time points. At 9 months, 34.3% of tooth sites receiving SDD attained this threshold level of improvement compared with 30.7% of sites receiving placebo. In diseased tooth sites with a baseline of PPD $\geq 4$ mm, the percentage of tooth sites with CAL gains $\geq 3$ mm from baseline was greater with adjunctive SDD than with placebo. The percentage of tooth sites with CAL gains $\geq 3$ mm was 60%, 30%, and 25% greater with adjunctive SDD than with adjunctive placebo at 3, 6, and 9 months, respectively. At 9 months, 29.9% of tooth sites had reductions of PPD $\geq 2$ mm or more in the SDD group compared with 22% of tooth sites in the placebo group. In summary, the adjunctive use of SDD with SRP is more effective than SRP alone and may represent a new approach in the long-term management of AP.   |
| Choi et al. [30]    | 32           | Chronic periodontitis | 4 months     | 1. SRP + SDD (20 mg 2x, 120d) ( $N=15$ )<br>2. SRP + placebo ( $N=17$ )   | After 120d, PPD (3.8 $\pm$ 1.5 from 5.4 $\pm$ 1.2) and CAL (4.2 $\pm$ 1.5 from 6.4 $\pm$ 0.9) improved significantly in the SRP + SDD group ( $P < 0.05$ ). Initial MMP-8 levels for the SRP + SDD group and the SRP + placebo group were 407.13 $\pm$ 114.45 ng/ml and 378.71 $\pm$ 189.39 ng/ml, respectively, with no statistical difference between the two groups. MMP-8 levels for the SRP + SDD group and the SRP + placebo group were: 235.35 $\pm$ 134.58 ng/mL and 364.04 $\pm$ 219.27 ng/mL at 30d; 157.50 $\pm$ 95.95 ng/mL and 236.60 $\pm$ 186.16 ng/mL at 60d; 102.70 $\pm$ 67.64 ng/mL and 208.56 $\pm$ 124.54 ng/mL at 90d; and 63.77 $\pm$ 53.33 ng/ml and 229.13 $\pm$ 168.09 ng/ml at 120d, respectively. The amount of decrease in MMP-8 levels for the SRP + SDD group was statistically significant compared to that for the SRP + placebo group, especially apparent at 120d ( $P < 0.05$ ). TIMP-1 levels in both groups increased from the baseline to 120d with statistical significance ( $P < 0.05$ ), but there was no significant difference between the two groups. Changes in MMP-9 and IL-6 levels were not statistically significant. In summary, adjunctive SDD therapy can improve the clinical parameters and this clinical improvement is reflected by controlled level of MMP-8 in chronic adult periodontitis after the therapy.  |
| Emingil et al. [51] | 20           | Chronic periodontitis | 12 months    | 1. SRP + SDD (20 mg, 2x, 3 months)<br>2. SRP + placebo ( $N=10$ )         | Significant improvements were observed in all clinical parameters in both groups over the 12-month period ( $P < 0.0125$ ). The SDD group showed a significantly greater reduction in mean PPD scores at 9 and 12 months (SDD group – baseline: 3.62 $\pm$ 0.20; 9 months: 2.09 $\pm$ 0.12; 12 months: 2.03 $\pm$ 0.13; vs placebo group – baseline: 3.97 $\pm$ 0.16; 9 months: 2.64 $\pm$ 0.18; 12 months: 2.65 $\pm$ 0.17) and in mean GI scores at all time points than the placebo group (baseline: 1.74 $\pm$ 0.15; 3 months: 0.54 $\pm$ 0.09; 6 months: 0.68 $\pm$ 0.08; 9 months: 0.49 $\pm$ 0.07; 12 months: 0.46 $\pm$ 0.10) ( $P < 0.05$ ). From baseline to 12 months, GCF scores and GCF MMP-8 levels were significantly reduced in both groups (SDD group: from 0.58 $\pm$ 0.04 mL at baseline to 0.15 $\pm$ 0.04 mL at 12 months; placebo group: from 0.50 $\pm$ 0.05 mL at baseline to 0.29 $\pm$ 0.06 mL at 12 months) ( $P < 0.0125$ ). The total amount of GCF MMP-8 in the LDD group was found to be about 50% lower than that of the placebo group at 3 months and about 70% at 6 months ( $P < 0.05$ ). After 6 months, adjunctive LDD therapy did not seem to provide any additional effect on GCF levels. In summary, the present results indicate that LDD therapy in combination with SRP can reduce GCF MMP-8 levels and improve clinical periodontal parameters in patients with chronic periodontitis. |

(continued)

Table 3.4 (continued)

| Study               | No. patients | Periodontal condition | Study period | Periodontal treatment  | Outcome   |
|---------------------|--------------|-----------------------|--------------|--|---|
| Emingil et al. [52] | 20           | Chronic periodontitis | 12 months    | 1. SRP+SDD (20 mg, 2x, 3 months)<br>(N=10)<br>2. SRP+placebo<br>(N=10) | Except for CAL, periodontal conditions of both SDD and placebo groups markedly improved between baseline and examinations at 3, 6, 9, and 12 months ( $P < 0.0125$ ). The decreased PPD and a substantial reduction in GI and PI scores support this observation. Both groups showed a similar reduction in PPD at 3 months and 6 months. However, the SDD group demonstrated a significantly greater reduction in PPD at 9 months and 12 months ( $P < 0.05$ ). The SDD improvements in the full-mouth CAL scores were similar for both study groups for all time points ( $P > 0.05$ ). The SDD group showed statistically significant improvement in GI scores compared to the placebo group at all time points ( $P < 0.05$ ). There was a significant reduction in the PI scores in both groups, with no significant differences between them over the study period ( $P > 0.05$ ). In the SDD group, GCF Ln-5 γ2 chain fragment levels were significantly reduced at 3 months ( $P < 0.0125$ ) and then slightly increased during the rest of the study period. In the placebo group, GCF 45 and 70 kDa Ln-5 γ2 chain fragments tended to decrease at 3 months compared to baseline, but did not reach significance; these levels continued to increase throughout the remainder of the study period. GCF Ln-5 γ2 chain fragment levels in the SDD group were significantly lower than those of the placebo group during the study period ( $P < 0.05$ ). In summary, the present data indicate that SDD therapy in combination with SRP therapy can reduce GCF Ln-5 γ2 chain fragment levels and improve clinical periodontal parameters in patients with chronic periodontitis.   |
| Emingil et al. [54] | 46           | Chronic periodontitis | 12 months    | 1. SRP+SDD (20 mg, 2x, 3 months)<br>(N=23)<br>2. SRP+placebo<br>(N=23) | The periodontal conditions of both SDD and placebo groups markedly improved between baseline and the re-examinations at 3, 6, 9, and 12 months ( $P < 0.0125$ ). The decreased PPD and a substantial reduction in PPD at 3 months ( $P > 0.05$ ). However, the SDD group demonstrated a significantly greater reduction in PPD than the placebo group at 6, 9, and 12 months (SDD group – baseline: $4.03 \pm 0.82$ , 3 months: $2.65 \pm 0.40$ , 6 months: $2.53 \pm 0.39$ , 9 months: $2.45 \pm 0.52$ , 12 months: $2.42 \pm 0.55$ vs placebo group – baseline: $4.15 \pm 0.80$ , 3 months: $2.86 \pm 0.46$ , 6 months: $2.85 \pm 0.57$ , 9 months: $2.88 \pm 0.61$ , 12 months: $2.89 \pm 0.62$ ( $P = 0.04$ , $P = 0.02$ , and $P = 0.04$ , respectively). The full-mouth mean CAL of SDD group significantly decreased during the study period ( $P < 0.025$ ). The CAL of the SDD group had greater improvement than the placebo group at all time points, but the difference was found to be significant only at 6 and 9 months (SDD group – baseline: $4.71 \pm 1.14$ , 3 months: $4.13 \pm 1.03$ , 6 months: $4.16 \pm 0.98$ , 9 months: $4.09 \pm 1.03$ , 12 months: $4.16 \pm 1.00$ vs placebo group – baseline: $5.19 \pm 1.20$ , 3 months: $4.73 \pm 1.01$ , 6 months: $4.86 \pm 1.05$ , 9 months: $4.84 \pm 1.06$ , 12 months: $4.84 \pm 1.10$ ( $P = 0.04$ , $P = 0.04$ , respectively). The SDD group showed statistically significant improvement in GI scores compared to the placebo group at 3, 6, and 9 months (LDD group – baseline: $1.82 \pm 0.40$ , 3 months: $0.76 \pm 0.33$ , 6 months: $0.83 \pm 0.25^*$ , 9 months: $0.70 \pm 0.29$ , 12 months: $0.70 \pm 0.35$ vs placebo group – baseline: $1.89 \pm 0.29$ , 3 months: $1.00 \pm 0.30$ , 6 months: $1.02 \pm 0.28$ , 9 months: $0.97 \pm 0.33$ , 12 months: $0.92 \pm 0.33$ ( $P = 0.01$ , $P = 0.01$ , and $P = 0.01$ , respectively) ( $P = 0.01$ , $P = 0.01$ , and $P = 0.01$ , respectively). There was a significant reduction in the PI scores of the sampling sites in both groups, with no significant differences detectable between them over the entire study period ( $P > 0.05$ ). GCF t-PA levels reduced in both groups over the 12-month period ( $P < 0.01$ ). The SDD group had lower GCF t-PA levels than the placebo group at 6 and 9 months ( $P < 0.05$ ). In summary, these results provide additional information about usefulness of SDD therapy as an adjunct to nonsurgical therapy in long-term management of periodontitis. |

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|--------------------------|----|---|-----------|--|---|
| Emingil et al. [53]      | 24 | Chronic periodontitis                     | 6 months  | 1. SRP+SDD (20 mg, 2x, 3 months)<br>(N=12)                       | In both study groups, the improvement in the full-mouth CAL scores was not significant at 3 and 6 months compared to baseline ( $P>0.025$ ). The SDD group showed a significantly greater reduction in PPD than the placebo group at 6 months ( $2.3 \pm 0.3$ from $3.7 \pm 0.6$ vs $2.6 \pm 0.5$ from $3.9 \pm 0.5$ ) ( $P=0.0433$ ) and in mean GI scores at 3 and 6 months (baseline: $1.8 \pm 0.5$ ; 3 months: $0.6 \pm 0.4$ 6 months: $0.7 \pm 0.2$ ) than the placebo group (baseline: $1.8 \pm 0.4$ ; 3 months: $1.0 \pm 0.4$ ; 9 months: $1.1 \pm 0.5$ ) ( $P<0.05$ ). There was no significant difference between the groups in the PI scores over the 6-month study period ( $2.2 \pm 0.8$ vs $2.2 \pm 0.8$ ) ( $P>0.05$ ). From baseline to 6 months, the GCF EMMPRIN levels were reduced significantly in the SDD group ( $P<0.025$ ). The GCF EMMPRIN level in the SDD group was significantly lower than that of the placebo group at 3 and 6 months ( $P<0.05$ ). In summary, SDD therapy in combination with SRP reduced GCF EMMPRIN levels and improved clinical periodontal parameters in subjects with chronic periodontitis.  |
| Górská & Nedzi-Góra [64] | 66 | Chronic periodontitis                     | 12 months | 1. SRP+SDD (20, 2x, 3 months) (N=33)<br>2. SRP (N=33)            | The BI was decreased by treatment in both groups. The reduction in doxycycline-treated patients was higher and the difference between the groups was found to be statistically significant ( $P=0.0034$ ) (test group: from $39.71 \pm 27.31$ to $11.92 \pm 9.36$ , $P<0.000001$ ) compared with controls: from $45.87 \pm 29.56$ to $33.82 \pm 23.79$ , $P=0.00026$ ). In patients treated with LDD, the mean PPD significantly decreased. After conventional treatment alone, the decrease in this parameter was not as marked, but the difference between the groups was significant ( $P=0.0030$ ) (test group: from $2.21 \pm 0.65$ to $1.92 \pm 0.45$ , $P=0.0002$ compared with controls: from $2.26 \pm 0.64$ to $2.18 \pm 0.5$ , $P=0.1830$ ). A similar tendency, i.e., a more marked reduction in the doxycycline group, was observed in the proportion of pockets $\geq 4$ mm (%PPD $> 4$ mm) ( $P=0.0383$ ) and maximum PPD (PPDmax). %PPD $> 4$ mm in the test group from $10.87 \pm 14.88$ to $4.81 \pm 6.32$ , $P=0.00348$ , controls: from $11.05 \pm 15.87$ , to $8.51 \pm 10.78$ , $P=0.0736$ ). On the other hand, the mean CAL loss was significantly reduced only in patients treated with LDD, and the difference between the two groups was statistically significant ( $P<0.05$ ) (from $3.27 \pm 1.61$ to $2.94 \pm 1.51$ , $P=0.00001$ ). Doxycycline did not produce significant reductions in MMP-8 and MMP-9 levels in saliva observed after the conventional treatment. The study revealed increases in the TIMP-1 concentration and the MMP-8/TIMP-1 and MMP-9/TIMP-1 ratios in saliva and blood after treatment with doxycycline. In summary, the study confirmed the modulating effect of doxycycline on the host response in chronic periodontitis   |
| Gürkan et al. [66]       | 26 | Severe, generalized chronic periodontitis | 12 months | 1. SRP+SDD (20, 2x, 3 months)<br>(N=13)<br>2. SRP+placebo (N=13) | No statistically significant changes were observed at sites with a baseline PPD 0–3 mm in both groups ( $P>0.05$ ). Significant PPD reductions were observed at sites with a baseline PPD 4–6 mm and $\geq 7$ mm as would be predicted and this reduction was maintained over the entire study period ( $P=0.025$ ). Although the mean PPD reduction for sites with a baseline PPD 4–6 mm and $\geq 7$ mm were greater in test than in control at 6 months (1.80 versus 1.46 mm for mild-to-moderate pockets; 3.38 versus 2.57 mm for deep pockets) the differences did not reach to significance ( $P>0.05$ ). Analysis of sites with baseline PPD $\geq 7$ mm revealed that higher percentage of sites were reduced by at least 3 mm following adjunctive SDD therapy (66.4%) than following adjunctive placebo therapy (55.1%) at 3 months, with no significant differences detectable between groups ( $P>0.05$ ). However, at 6 months percentage of sites that exhibited improved PPD by $\geq 3$ mm were significantly higher ( $P=0.011$ ) in adjunctive SDD group (73.4%) than adjunctive placebo group (49.7%). None of the deep pockets exhibited a PPD $\geq 3$ mm during the study period. The full-mouth PBI and PI scores of both SDD and placebo groups markedly improved between baseline and the re-examinations at 3 and 6 months ( $P=0.025$ ). The reduction in PBI and PI was similar for both groups at all time points ( $P>0.05$ ). At baseline there were no significant differences in GCF TGF- $\beta$ 1 levels between three groups. Both total amount and concentration of GCF TGF- $\beta$ 1 in SDD and placebo groups increased when compared with baseline at 3 months. However, only GCF TGF- $\beta$ 1 levels of SDD group was significantly higher than baseline ( $P<0.025$ ) and placebo group ( $P<0.017$ ) at 3 months. At 6 months GCF TGF- $\beta$ 1 levels of both groups were similar to baseline levels ( $P<0.025$ ). In summary, these data indicate that combination of SDD with nonsurgical therapy improves clinical parameters of periodontal disease and increases GCF TGF-beta levels together with a decrease in prevalence of residual pockets in patients with severe, generalized chronic periodontitis. |

(continued)

Table 3.4 (continued)

| Study                | No. patients | Periodontal condition                     | Study period | Periodontal treatment  | Outcome  |
|----------------------|--------------|---|--------------|--|--|
| Gürkan et al. [67]   | 26           | Severe, generalized chronic periodontitis | 12 months    | 1. SRP+SDD (20, 2x, 3 months)<br>(N=13)<br>2. SRP+placebo<br>(N=13)  | The full-mouth clinical parameters markedly improved between baseline and the re-examinations at 3, 6, 9, and 12 months in both groups ( $P < 0.0125$ ). Full-mouth clinical parameters were similar in both groups at all time points ( $P > 0.05$ ). Significant PPD reductions were observed at sites with a baseline PPD 4–6 mm and $\geq 7$ mm and this reduction was maintained over the entire study period in both groups ( $P < 0.01$ ). Mean PPD reduction for sites with a baseline PPD 4–6 mm and $\geq 7$ mm were higher in test group than in control group at 3, 9, and 12 months; however, the differences did not reach significance ( $P > 0.05$ ). At 6 months the SDD group had exhibited significantly higher PPD reduction in deep sites compared with the placebo group ( $P < 0.05$ ). Sites with moderate and deep pockets initially exhibited significant clinical attachment gain at all time intervals when compared with baseline ( $P < 0.01$ ). Sites with an initial PPD 0–3 mm did not exhibit statistical significant CAL chances over the study period ( $P > 0.01$ ). Although the mean clinical attachment gain for sites with a baseline PPD 4–6 mm and $\geq 7$ mm were greater in SRP plus SDD group when compared with SRP plus placebo group at all time points, differences were not statistically significant ( $P > 0.05$ ). Analysis of sites with a baseline PPD $\geq 7$ mm revealed that higher percentage of sites was reduced by at least 3 mm following adjunctive SDD therapy than following adjunctive placebo therapy at all time points. These effects were significant at 6 months (73.4% versus 49.7%, $P < 0.05$ ; NNT=4.2, 95% CI: 2.70–5.71) and at 9 months (79.9% versus 50.6%, $P < 0.05$ , NNT=3.4, 95% CI: 2.22–7.34). In addition percentage of initially deep pockets exhibiting $\geq 4$ mm or more PPD reduction from baseline was significantly higher in the SDD group than in the placebo group at 6 months (53.4% versus 36.3%, $P < 0.05$ ; NNT=5.9, 95% CI: 3.47–18.73). None of the deep pockets exhibited a PPD increase more than or equal to 3 mm during the study period. In summary, these results ensure further data for beneficial effects of adjunctive SDD therapy in the management of severe chronic periodontitis. |
| Haffajee et al. [72] | 92           | Chronic periodontitis                     | 12 months    | 1. SRP+AZ (500 mg once daily for 3 days) (N=25)<br>2. SRP+MET (250, 3x, 14d) (N=24)<br>3. SRP+doxycycline (SDD, Periostat, 20 mg for 12 weeks) (N=20)<br>4. SRP (N=23) | All groups showed clinical improvements at 12 months, with subjects receiving adjunctive agents showing a somewhat better response. Some subjects showed attachment loss at 12 months in each group ranging from 15% to 39% of subjects in the SDD and SRP only groups respectively. Subjects receiving AZ exhibited the largest percentage of sites showing attachment gain $> 2$ mm at 12 months (5.3%), while the MET group showed the lowest percentage of sites showing loss of attachment $> 2$ mm (0.41%). All groups exhibited a greater percentage of sites gaining attachment than losing attachment $> 2$ mm. Subjects receiving systematically administered AZ or MET showed greater reductions in mean PPD and AL at sites with initially deeper pockets ( $> 6$ mm) compared with the subjects receiving SRP alone or SDD. In summary, this study demonstrated that periodontal therapy provides clinical benefits and that antibiotics provide a clinical benefit over SRP alone, particularly at initially deeper periodontal pockets.   |
| Lee et al. [94]      | 51           | Moderate to chronic periodontitis         | 9 months     | 1. SRP+SDD (20 mg, 2x, 90d) (N=24)<br>2. SRP+placebo<br>(N=17)   | During the treatment period, per-patient reductions in PPD (at 9 months: $1.63 \pm 0.07$ vs $1.19 \pm 0.06$ in controls) and CAL (at 9 months: $1.56 \pm 0.06$ vs $0.80 \pm 0.05$ ) ( $P < 0.05$ ) were demonstrated for both treatment groups, with a significantly greater reduction for the SDD group. The mean value of per-patient change in the GCF was much greater for the SDD group (at 9 months: $80.20 \pm 0.12$ vs $61.50 \pm 9.76$ ). Microbial analysis showed there was a general tendency for cocci, nonmotile rods, and aerobes to increase with increasing treatment duration and a general decreasing tendency for spirochetes, motile rods, and anaerobes and black pigmented bacteria in both treatment groups, but no significant difference between the groups. The MMP-8 and -13 levels of the SDD group decreased gradually from $3.53 \pm 0.34$ and $2.42 \pm 0.17$ at the baseline to $1.99 \pm 0.15$ and $1.40 \pm 0.04$ after 9 months respectively. The MMP-8 and -13 levels in the placebo group decreased gradually from $3.22 \pm 0.32$ and $2.41 \pm 0.27$ at baseline, to $2.66 \pm 0.23$ and $1.84 \pm 0.18$ after 9 months, respectively. In summary, this study suggests that SDD as an adjunctive therapy with SRP might be safe and effective in the long-term management of chronic periodontitis.  |

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| Mohammadi et al. [123] | 48  | Moderate to severe chronic periodontitis | 9 months | 1. SRP+SDD (20, 2x, 3 months) ( <i>N</i> =24)<br>2. SRP+placebo ( <i>N</i> =24)  | At all time points and in both moderate and deep sites, SRP+SDD resulted in significantly greater PPD reductions relative to baseline than SRP+placebo. At month 9, in moderate sites, mean PPD reductions of $1.57 \pm 0.11$ mm were reported in the adjunctive SDD group, compared with $0.63 \pm 0.11$ mm in the adjunctive placebo group. In deep sites at month 9, mean PPD reductions of $3.22 \pm 0.29$ mm were reported in the adjunctive SDD group, compared with $0.98 \pm 0.31$ mm in the adjunctive placebo group. In summary, SDD used as an adjunct to SRP provides significant benefit for elderly patients with CP compared with SRP alone.   |
| Needleman et al. [134] | 34  | Chronic periodontitis                    | 6 months | 1. SRP+SDD (20, 2x, 3 months) ( <i>N</i> =16)<br>2. SRP+placebo ( <i>N</i> =18)  | The velocity of change was statistically greater for the test group for CAL – 0.19 mm/month (95% CI: –0.34, 0.04; $P=0.012$ ) and PDD 0.30 mm/month (95% CI: –0.42, –0.17; $P<0.001$ ). The final improvements were no different between the test and control groups regarding the PPD [control: –0.98 (–2.17, 0.21); test: –1.40 (–2.69, –0.11)], CAL [control: –0.40 (–1.20, 0.39); test: –0.65 (–2.13, 0.84)], BOP [control: –27.2 (–64.8, 10.4) and test: –21.8 (–59.7, 16.1)], and Plaque [control: –19.3 (–67.0, 28.4) and test: –21.7 (–60.1, 16.7)] changes ( $P>0.05$ ), but multilevel modeling revealed different trajectories for clinical changes. In other words, the end point of clinical healing was similar for both experimental groups; however, the rate of improvement was greater for the test group. No differences regarding absolute CAL change or terminal carboxyteleopeptide of type 1 collagen in gingival crevicular fluid were evident. In summary, this study does not provide evidence of a benefit of using SDD as an adjunct to nonsurgical periodontal therapy in smokers.   |
| Novak et al. [137]     | 20  | Severe generalized chronic periodontitis | 9 months | 1. SRP+SDD (20, 2x, 6 months) ( <i>N</i> =10)<br>2. SRP+placebo ( <i>N</i> =10)  | Subgingival debridement plus adjunctive SDD reduced deep pockets ( $\geq 7$ mm at baseline) by an average of 3.02 mm after 9 months versus 1.42 mm for the placebo group. A significant clinical response was seen in both groups as soon as 1 month, but the response was always clinically and statistically greater in the SDD group. In the SDD group, nearly 40% of 237 pockets $\geq 7$ mm were reduced by $\geq 4$ mm, and 55% were reduced by $\geq 3$ mm. In addition, only two pockets deepened by $\geq 4$ mm in the SDD group versus 10 in the placebo group. In summary, the supplementation of full-mouth subgingival and supragingival debridement with a host-modulating agent, SDD, provides clinically and statistically significant benefits in the reduction of deep pockets in patients with severe, generalized periodontitis. In addition, adjunctive SDD is more effective than a placebo in preventing further increases in periodontal probing depth.   |
| Novak et al. [136]     | 171 | Chronic periodontitis                    | 6 months | 1. SRP+SDD (20 mg, 2x, 6 months) +locally delivered doxycycline hyclate gel (TAT, 10%, in pockets $\geq 5$ mm)<br>2. SRP+placebo ( <i>N</i> =83) | Combination therapy showed significantly greater reductions in PPD at 3 and 6 months than were seen with SRP+placebo. For PPDs that were 4–6 mm at baseline, there were reductions in PPD of 1.5 mm for the experimental group versus 0.9 mm for controls by 3 months ( $P<0.01$ ) and 1.7 versus 1.2 mm, respectively, by 6 months ( $P<0.01$ ). For PPDs $\geq 7$ mm at baseline, significant reductions of 2.1 mm were noted in the experimental group versus 1.4 mm for controls. For PPDs that were 4–6 mm at baseline, there were gains in CAL of 1.5 mm for the experimental group versus 0.9 mm for controls by 3 months ( $P<0.01$ ) and 1.5 mm versus 1.2 mm, respectively, by 6 months ( $P<0.04$ ). For pockets $\geq 7$ mm at baseline, gains in CAL were 2.0 mm for the experimental group versus 1.3 mm for controls at 3 months ( $P<0.01$ ) and 2.2 mm versus 1.6 mm, respectively, at 6 months ( $P<0.02$ ). For sites with baseline PPDs of 4–6 mm, mean reductions in BOP were 34% for experimental subjects versus 24% for controls at 3 months ( $P<0.02$ ), and 43% versus 32%, respectively, at 6 months ( $P<0.05$ ). For PPDs $\geq 7$ mm at baseline, BOP was reduced by 27% for the experimental group versus 13% for controls at 3 months ( $P<0.01$ ), and 29% versus 19%, respectively, at 6 months ( $P>0.05$ ). Similar reductions in GI were observed for experimental and control subjects, with baseline PPDs of 4–6 mm showing a reduction in GI of 39% for the experimental group versus 26% for controls at 3 months ( $P<0.01$ ) and 49% versus 37%, respectively, at 6 months ( $P<0.04$ ). In summary, combination therapy, including SRP, HMT, and TAT, provided significantly greater clinical benefits than SRP alone in the treatment of moderate to severe CP. |

(continued)

Table 3.4 (continued)

| Study                 | No. patients | Periodontal condition                                 | Study period | Periodontal treatment   | Outcome   |
|-----------------------|--------------|---|--------------|---|---|
| Payne et al. [149]    | 128          | Postmeno pausal osteopenic women with periodontitis   | 2 years      | 1. SRP+SDD (20 mg, 2x, 2 years)<br>2. SRP+placebo (N=64)              | The vast majority of sites for oral radiographic outcomes did not show significant change at 1 or 2 years, either improvement or disease progression (81–95% depending on the time point and measurement: RA, CADIA, and alveolar bone height). Based on regressing modelling, the odds of more progressive disease did not differ significantly between groups based on the categorical RA measure [OR=1.04 (SDD relative to placebo), 95% CI: 0.80–1.34, $P=0.8$ ] or the categorical CADIA measure [OR=0.84 (SDD relative to placebo), 95% CI: 0.65–1.08, $P=0.21$ ]. SDD did not differ overall from placebo on alveolar bone loss. Based on the continuous measure of CADIA change, there was a significant interaction among study drug, time and baseline periodontal probing depth ( $P=0.03$ ). Among sites with a baseline periodontal probing depth of $\geq 5$ mm, SDD was associated with reduced alveolar bone density loss relative to placebo (difference in mean change = 2.45, 95% CI: 0.85–4.04, $P=0.003$ ). There was significant evidence that the effect of study drug differed by time after menopause for alveolar bone height change (drug by menopause interaction, $P=0.04$ ). Among subjects who were beyond 5 years of menopause, SDD was associated with a 29% reduction in the odds of more progressive disease (bone height loss) (SDD/placebo OR=0.71, 95%CI: 0.50–0.9, $P=0.05$ ). In summary, in postmenopausal osteoporotic women with periodontitis, SDD did not differ overall from placebo. Based on exploratory subgroup analyses, additional research is needed to determine the usefulness of SDD in nonsmokers, subjects 45 years post menopause and in deeper pockets. |
| Presshaw et al. [153] | 209          | Chronic periodontitis                                 | 9 months     | 1. SRP+SDD (20 mg, 2x, 9 months)<br>2. SRP+placebo (N=102)            | In periodontal sites with PPD 4–6 mm, mean improvements in CAL and PPD were greater in the test group than in controls, achieving statistical significance in all baseline disease categories at month 9 (CAL gains: 1.27 ± 0.05 vs 0.94 ± 0.05; PPD reduction: 1.29 ± 0.05 vs 0.96 ± 0.06, $P<0.05$ ). In periodontal sites with PPD $\geq 7$ mm, mean improvements in CAL and PPD were greater following SRP+SDD than SRP+placebo, achieving statistical significance in all baseline disease categories at month 9 (CAL gains: 2.09 ± 0.13 vs 1.60 ± 0.15; PPD reduction: 2.31 ± 0.12 vs 1.77 ± 0.13) ( $P<0.05$ ). At month 9, 42.3% of sites in the SDD group demonstrated CAL gain $\geq 2$ mm compared to 32.0% of sites in the placebo group ( $P<0.01$ ). CAL gain $\geq 3$ mm was seen in 15.4% of sites in the SDD group compared to 10.6% of sites in the placebo group ( $P<0.05$ ). When considering the same thresholds of change in PPD, 42.9% of sites in the SDD group compared to 31.1% of sites in the placebo group demonstrated PPD reduction $\geq 2$ mm ( $P<0.01$ ), and 15.4% of sites in the SDD group compared to 9.1% of sites in the placebo group demonstrated PPD reduction $\geq 3$ mm ( $P<0.01$ ). In summary, adjunctive SDD enhances SRP. It results in statistically significant attachment gains and periodontal probing depth reductions over and above those achieved by SRP with placebo.   |
| Presshaw et al. [154] | 266          | Untreated periodontitis in a population over 18 years | 9 months     | 1. SRP+SDD-40 (40 mg, 1x, 9 months) (N=133)<br>2. SRP+placebo (N=133) | Adjunctive SDD-40 provided significantly greater clinical benefits than placebo at all time points. At month 9, at sites with baseline PPD $\geq 6$ mm, 72–76% of sites in the SDD-40 group demonstrated clinically significant PPD reductions and CAL gains $\geq 2$ mm compared to 56–58% of sites in the placebo group ( $P<0.0001$ ): 48–52% of sites in the SDD-40 group demonstrated PPD reductions and CAL gains $\geq 3$ mm compared to 32% of sites in the placebo group ( $P<0.0001$ ). In moderate sites (baseline PPD 4–6 mm), adjunctive SDD-40 provided significant clinical benefits compared to placebo for mean CAL (all time points; 3 months: 1.48 ± 0.10 vs 1.33 ± 0.10; 6 months: 1.67 ± 0.16 vs 1.50 ± 0.16; 9 months: 1.66 ± 0.16 vs 1.49 ± 0.17, $P<0.05$ ), PPD (3 months: 1.49 ± 0.08 vs 1.28 ± 0.08; 6 months: 1.61 ± 0.12 vs 1.40 ± 0.12; 9 months: 1.63 ± 0.13 vs 1.41 ± 0.13, $P<0.001$ ), and BOP (3 months: $P<0.01$ ; 6 months: $P<0.02$ ; 9 months: $P<0.05$ ). In deep sites (baseline PPD $\geq 7$ mm), SDD-40 provided significant benefits over control for mean CAL (3 months: 2.38 ± 0.07, $P<0.05$ ; 6 months: 2.71 ± 0.32 vs 2.21 ± 0.32; 9 months: 2.84 ± 0.29 vs 2.25 ± 0.29; 9 months: 2.74 ± 0.24 vs 2.25 ± 0.24, $F<0.001$ ), and BOP (3 months: $P<0.05$ ; 6 months: not statistically significant; 9 months: $P<0.05$ ). In summary, SDD-40 used as an adjunct to SRP resulted in significantly greater clinical benefits than SRP alone in the treatment of periodontitis.  |

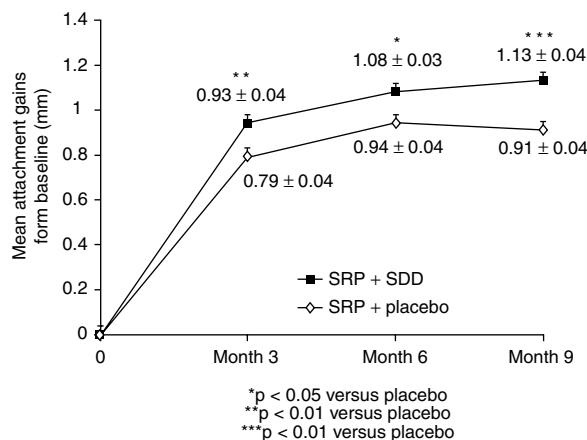
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|-----------------------|-----|---|---------|---|--|
| Reinhardt et al. [64] | 128 | Postmeno pausal osteopenic women with periodontitis | 2 years | <p>1. SRP+SDD (20 mg, 2x, 2 years)<br/>(N=64)</p> <p>2. SRP+placebo<br/>(N=64)</p> <p>All subjects received calcium and vitamin D supplements twice daily (a total of 1200 mg of calcium and 400 IU of vitamin D daily)</p> | <p>The vast majority of sites (93–95% in placebo and 92–94% in SDD as means over four post-baseline visits) did not manifest significant CAL change across the 2-year clinical trial. Based on regression modeling, the odds of more disease progression continuum from improvement to no change to progression were 19% lower (2.1% absolute difference in number of sites) for subjects receiving SDD relative to placebo (OR=0.81%, 95% CI: 0.67–0.97, <math>P=0.03</math>). A 20% (relative) reduction in the percentage of sites showing significant CAL progression and a 46% (relative) increase in the percentage of sites showing significant CAL improvement were seen in the SDD group compared with the placebo group over 2 years. Most sites showed no change in PPD across time (90–93%). The odds of more progressive PPD change were 15% lower in the SDD group relative to the placebo group, but this was not statistically significant (OR=0.85%, 95% CI: 0.58–1.24, <math>P=0.4</math>). The odds of BOP were 18% lower for subjects receiving SDD relative to placebo, which was not statistically significant (OR = 0.82%, 95% CI: 0.61–1.10, <math>P=0.2</math>). However, based on logistic regression modeling after adjustment for the treatment effect, BOP was significantly reduced in both groups (<math>P&lt;0.0001</math>) following the baseline visit. Exploratory subgroup analyses showed that the effect of study drug differed by smoking status (<math>P=0.01</math>), where SDD was associated with a reduction in the odds of BOP relative to placebo for nonsmokers (OR=1.58%, 95% CI: 0.96–2.62, (OR=0.70%, 95% CI: 0.50–1.00, <math>P=0.05</math>) and a marginal increase for smokers (OR=1.58%, 95% CI: 0.96–2.62, <math>P=0.07</math>). In summary, analyses of secondary outcomes of this clinical trial indicated that SDD may be of benefit in reducing progressive attachment loss in postmenopausal females; additional research is needed to confirm these findings.</p>   |
| Golub et al. [58]     | 128 | Postmeno pausal osteopenic women with periodontitis | 2 years | <p>1. SRP+SDD (20 mg, 2x, 2 years)<br/>(N=64)</p> <p>2. SRP+placebo<br/>(N=64)</p> <p>All subjects received calcium and vitamin D supplements twice daily (a total of 1200 mg of calcium and 400 IU of vitamin D daily)</p> | <p>The SDD-treated postmenopausal women showed ~50% reduction in GCF collagenase activity over the 2 years compared to their own baseline values. In contrast, the placebo values appeared to decrease only slightly. Moreover, based on linear regression analysis, the SDD-treated group showed a statistically significant 22% reduction in median GCF collagenase activity compared to placebo-treated subjects over the study period, based on intent-to-treat analysis (95% CI: 37% lower to 5% lower; <math>P=0.01</math>), and a 29% reduction in median GCF collagenase activity compared to placebo subjects based on the per-protocol analysis (95% CI: 48% lower to 4% lower; <math>P=0.02</math>) after adjusting for baseline values. For subgroup analyses, the effect of SDD seemed to depend on smoking status (<math>P=0.05</math>), and there was a significant interaction between time and treatment for nonsmokers (<math>P=0.02</math>). At 1 year, median levels of collagenase activity per pool of GCF were 40% lower for SDD subjects compared to placebo subjects in the nonsmoking group, which was statistically significant (95% CI: 53% lower to 22% lower; <math>P&lt;0.0001</math>). In contrast, SDD therapy over the study period seemed to reduce the median ICTP levels per pool of GCF by ~30% compared to this group's own baseline values. Using linear regression analysis, the SDD-treated group showed a 16% reduction in median GCF ICTP levels compared to placebo-treated subjects, after adjusting for baseline values (<math>P=0.08</math>). Focusing on changes in the dominant type of collagenase, MMP-8, in the GCF of these postmenopausal women, and based on intent-to-treat analysis, SDD therapy reduced the odds of elevated MMP-8 values (across the ordered categories of 0–1.00, 1.001–2.5, and &gt;2.5 units) by 60% compared to placebo during the 2-year study period. This treatment effect was highly statistically significant (OR = 0.40, 95% CI: 0.21–0.77; <math>P=0.006</math>). Consistent with this pattern, SDD therapy increased the odds of lower values (among the ordered categories of 0–1.00, 1.001–2.5, and &gt;2.5 units) for this type of collagenase, compared to placebo therapy, over the study period. Based on per-protocol analysis, this effect was even more dramatic because the odds of higher values for MMP-8 in SDD-treated subjects were 78% lower than in those receiving placebo tablets (OR=0.22, 95% CI: 0.07–0.66, <math>P=0.007</math>).</p> |

(continued)

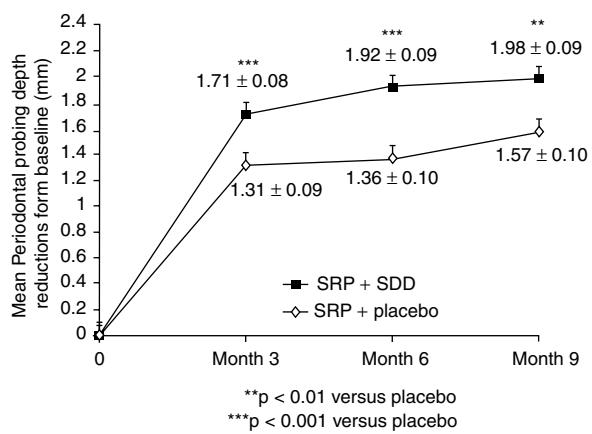
Table 3.4 (continued)

| Study               | No. patients | Periodontal condition  | Study period | Periodontal treatment   | Outcome   |
|---------------------|--------------|--|--------------|---|---|
| Tütter et al. [202] | 36           | Patients with both chronic periodontitis and coronary artery disease | 6 weeks      | 1. SRP+SDD (20, 2x, 6 weeks) (N=18)<br>2. SRP+placebo (N=18)      | There were statistically significant improvements for all clinical parameters, GCF volumes, GCF MMPs, and serum levels of HsCRP, apolipoprotein-A (APO-A), high-density lipoprotein (HDL), and lipoprotein-a between pre- and post-treatment in both groups. Between groups, there were statistically significantly greater improvements in PPD (controls: from 4.59 (4.18–4.72) to 3.78 (3.52–4.2); test: from 4.44 (4.19–4.8) to 3.45 (3.24–3.69), GI (controls: from 1.90 (1.83–2.08) to 1.27 (0.97–1.52); test from 1.99 (1.86–2.21) to 1.0 (0.77–1.16)). APO-A and HDL, favoring the group receiving SDD adjunctive to SRP ( $P < 0.05$ ). In summary, greater improvement was detected for PPD and GI, and for serum levels of APO-A and HDL cholesterol when using SRP+SDD compared with SRP+placebo in this study. An investigation with larger numbers of patients and a longer duration of drug treatment is needed to confirm these preliminary findings.  |
| Walker et al. [216] | 128          | Postmeno pausal osteopenic women with periodontitis                  | 2 years      | 1. SRP+SDD (20 mg, 2x, 24 months) (N=64)<br>2. SRP+placebo (N=64) | Based on ITT analysis, the odds of periodontitis progression for relative clinical attachment level, measured using an electronic probe, over 2 years were 19% lower in the SDD group than in the placebo group (OR = 0.81; 95% CI: 0.67–0.97; $P = 0.03$ ). There were no significant differences between the treatment groups for the change in the odds of detection over time for the more common pathogens ( $P = 0.4$ for <i>Candida</i> and enterics). For <i>Prevotella intermedia</i> , <i>Eikenella corrodens</i> , and <i>Fusobacterium nucleatum</i> , there was no evidence of a change within either treatment group over time. Based on logistic regression modeling, the odds of resistance decreased over the treatment period by 51% (relative change) for the placebo group and by 14% (relative change) for the SDD group. These changes in resistance over the treatment period did not differ significantly between the groups ( $P = 0.2$ ). These data indicated that treatment with SDD for 24 months did not result in a significant increase in susceptibilities to doxycycline (as determined by change in MICs) or a significant increase in the proportion of the isolates recovered that were characterized as resistant to therapeutic levels. In summary, no antimicrobial effect on the subgingival flora was detected following treatment with SDD for 24 months, relative to baseline or to placebo. The increase in initial resistance (at 4 $\mu\text{g/mL}$ ) did not translate into a significant increase in the percent resistant to doxycycline ( $\text{MIC} \geq 16 \mu\text{g/mL}$ ) for patients in the SDD group. |

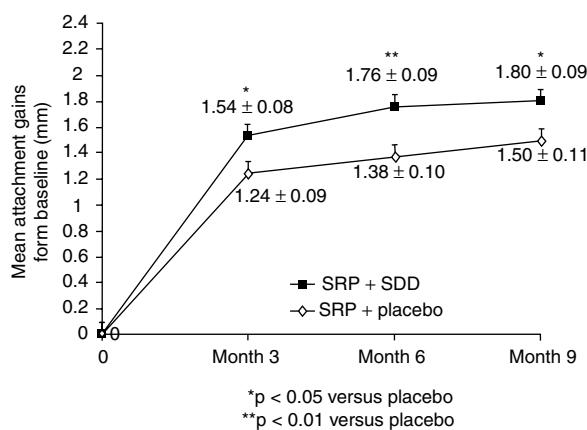
SRP scaling and root planning, MMP matrix metalloproteinases, TIMP tissue inhibitors of metalloproteinases, SDD sub-antimicrobials dose doxycycline (20 mg orally administered twice daily), PPD periodontal probing depth, CAL clinical attachment loss, GCF gingival crevicular fluid, LDD low-dose doxycycline, TGF-1 transforming growth factor-beta 1, ITT intent-to-treat, OR odds ratio, APO-A apolipoprotein-A, HDL high-density lipoprotein



**Fig. 3.1** Meta-analysis of data from Caton et al. [26] and Preshaw et al. [151]. Effect of sub-antimicrobial dose doxycycline (SDD) on attachment gains in tooth sites with mild to moderate disease (baseline periodontal probing depths 4–6 mm). Patients received scaling and root planning (SRP) at baseline, and then received SDD 20 mg BID (■, n = 197) or placebo BID (◊, n = 195) for 9 months. The mean per patient changes from baseline, adjusted for smoking status, and standard errors are presented [152, 153] (Reprinted with permission John Wiley & Sons)



**Fig. 3.3** Meta-analysis of data from Caton et al. [26] and Preshaw et al. [151]. Effect of sub-antimicrobial dose doxycycline (SDD) on periodontal probing depth (PPD) reductions in tooth sites with severe disease (baseline PPD ≥ 7 mm). Patients received scaling and root planning (SRP) at baseline, and then received SDD 20 mg BID (■, n = 197) or placebo BID (◊, n = 195) for 9 months. The mean per patient changes from baseline, adjusted for smoking status, and standard errors are presented [152, 153] (Reprinted with permission John Wiley & Sons)



**Fig. 3.2** Meta-analysis of data from Caton et al. [26] and Preshaw et al. [151]. Effect of sub-antimicrobial dose doxycycline (SDD) on attachment gains in tooth sites with severe disease (baseline periodontal probing depths (PPDs) ≥ 7 mm). Patients received scaling and root planning (SRP) at baseline, and then received SDD 20 mg BID (■, n = 197) or placebo BID (◊, n = 195) for 9 months. The mean per patient changes from baseline, adjusted for smoking status, and standard errors are presented [152, 153] (Reprinted with permission John Wiley & Sons)

benefits of adjunctive SDD were apparent as early as 3 months after commencing treatment, and were maintained for the 9 months of the study.

Another meta-analysis was performed by Reddy et al. [163] based on the evaluation of seven papers

[7, 26, 39, 59, 137, 151, 153]. For sites with pretreatment PPD of 4–6 mm and ≥ 7 mm, a statistically significant adjunctive benefit of clinical attachment level (CAL) was found when SDD was used in combination with SRP. For PPD changes, a significant adjunctive benefit was noted after a combination of SRP and SDD for pretreatment PPDs of 4–6 mm and ≥ 7 mm [163].

## Safety Data

Doxycycline at antibiotic doses (≥ 100 mg) is associated with adverse events including, among others, photosensitivity, hypersensitivity reactions, nausea, vomiting, and esophageal irritation. During active treatment, SDD was well tolerated, with no differences between the treatment groups in the incidence of adverse events, including those events related to infection or associated with the gastrointestinal and urogenital tract [25, 26]. The most frequently reported adverse events were headache, common cold, and influenza symptoms, and there were no significant differences in the incidence of these adverse events between the treatment groups. The types of adverse events did not differ significantly between treatment groups and the typical side effects of the tetracycline class of antibiotics were not observed [26]. Furthermore, adjunctive SDD did not exert

a detrimental antimicrobial effect on the oral, large intestine or vaginal microflora [199, 200, 215, 220], nor did its use contribute to changes in the antibiotic susceptibility of the periodontal microflora [198, 215].

### 3.6.1.3 Metronidazole

Metronidazole, a 5-nitroimidazole compound, specifically targets anaerobic microorganisms but has essentially no activity against aerobic or microaerophilic bacteria. Initially, metronidazole was thought to interact with biochemical pathways present only in obligate anaerobes. It is now known that cytotoxic metabolites of metronidazole directly interact with bacterial DNA, and possibly other macromolecules, resulting in cell death. Upon entry into an anaerobic organism, metronidazole is reduced at the 5-nitro position by electron transport proteins that are part of anaerobic metabolic energy-yielding pathways. Alteration of the metronidazole molecule creates a continuous concentration gradient favoring diffusion of additional metronidazole into the cell. Reduction of the parent compound yields many short-lived cytotoxic free radicals. These free radicals react with macromolecules, particularly DNA, resulting in cell death. Although resistance to metronidazole occurs in some anaerobic bacteria, e.g., *Fusobacterium* sp., it is relatively rare and appears to be due to a decrease in the ability of the bacterium to actively reduce the 5-nitro position [219].

The most common adverse reactions associated with metronidazole involve the gastrointestinal tract. About 12% of the patients experience nausea, which may be accompanied by headache, anorexia, and vomiting. Drowsiness, depression, skin rashes, and vaginal and/or urethral burning have been reported. Metronidazole affects the activity of hepatic enzymes involved with the metabolism of ethanol, producing unpleasant symptoms due to the accumulation of acetaldehyde in the blood. Alcohol ingestion is strictly contraindicated in patients receiving metronidazole. Metronidazole crosses the placenta barrier, entering the fetal circulation system. It is also secreted in breast milk. Because of the association of metronidazole with tumorigenicity in some animals, the drug is contraindicated in pregnant women or nursing mothers [219].

Several studies summarized in Table 3.5 have investigated the efficacy of metronidazole in

conjunction with and without SRP or periodontal surgery. Controversial results were obtained in patients with chronic severe periodontal disease, with both positive and nonsignificant benefits of the adjuvant metronidazole therapy. No clear trend was found, although it seems that dosage was more relevant in the results than factors related to debridement. More recent studies, prescribing higher dosages and including patients with more aggressive forms of periodontitis, tended to report better results [78].

In a meta-analysis, Elter et al. [50] provided a quantitative overview of clinical trials assessing the use of systemic metronidazole (MET) as an adjunct to SRP in the treatment of adult periodontitis. Eight clinical trials were chosen based upon a priori selection criteria, and two outcomes, “reduction in periodontal probing depth” (PPD) and “gain in clinical attachment level” (CAL), were analyzed. Results for each outcome were stratified by initial PPD 1–3 mm, 4–6 mm, or  $\geq 7$  mm, and length of follow-up 4–6 weeks, 9–13 weeks, or 14–26 weeks. MET in conjunction with SRP was superior to SRP alone in reducing PPD where initial PPD was 4–6 mm and follow-up was 9–13 weeks (0.43 mm; 99% CI: 0.12, 0.73). No significant advantage was observed for MET for reducing PPD where initial PPD was less than 4 mm or follow-up was longer than 13 weeks. MET in conjunction with SRP was superior to SRP alone in reducing CAL where initial PPD was 4–6 mm and where follow-up was 4–6 weeks (0.29; 99% CI: 0.01, 0.58) and 9–13 weeks (weighted mean difference 0.32; 99% CI: 0.03, 0.61). Significant heterogeneity of effect was not seen for PPD or CAL at any level of initial PPD or length of follow-up. No significant dose-response relationship was observed. This meta-analysis was limited due to diversity of data presentation and the small number of trials in each stratum. These results suggest that MET in conjunction with SRP may offer a benefit over SRP alone in the treatment of adult periodontitis patients in managing pockets of 4 mm or greater, but the additional benefit was not evident if initial PPD was less than 4 mm or follow-up was beyond 13 weeks [50]. Another meta-analysis performed by Herrera et al. [80] revealed that for CAL change in initially deep pockets, the effect of metronidazole was close to the level of statistical significance ( $P=0.057$ ; Fig. 3.5).

**Table 3.5** Clinical studies of systemic metronidazole therapy in periodontal disease

| Study                | No. patients | Periodontal condition | Study period | Periodontal treatment   | Outcome  |
|----------------------|--------------|-----------------------|--------------|---|--|
| Carvalho et al. [24] | 44           | Chronic periodontitis | 3 months     | 1. Control (C): SRP + placebo ( $N=10$ )<br>2. Test 1 (T1): SRP + systemically MET (400, 3x, 10d) ( $N=12$ )<br>3. Test 2 (T2): SRP + weekly PC + placebo; ( $N=12$ )<br>4. Test 3 (T3): SRP + MET (400, 3x, 10d) + PC ( $N=10$ ) | The percentage of sites exhibiting plaque accumulation, BOP, suppuration, as well as mean full-mouth PPD and AL were significantly reduced at 90 days post therapy in all treatment groups. There was a statistically significant difference among groups at 90 days for plaque and BOP with groups T2 and T3 showing greater reductions than groups C and T1. Although not significantly from the other treatment groups, the greatest reduction in mean AL was seen in group T3, which received the combination of therapies. The baseline shallow pockets ( $<4$ mm), showed an increase in the mean PPD in the control group and in mean CAL in all treatment groups. At pockets with an initial PPD of 4–6 mm, the changes observed for mean PPD and CAL did not differ significantly among groups. However, subjects in group T3 showed the greatest improvement in mean CAL post therapy, followed by the subjects in group T2. The percentage of sites with BOP was significantly different among groups at the initially 4–6 mm sites. Subjects in groups T2 and T3 showed a more marked reduction in this parameter when compared with groups C and T1. The three test groups (T1, T2, and T3) also showed a somewhat greater reduction in PPD when compared with the control group. At pockets with baseline PPD $>6$ mm, there was a significant difference among groups for PPD reduction. Sites in subjects receiving adjunctive MET, with or without PC (T1 and T3), exhibited a greater mean reduction in PPD than groups C and T2. In summary, the data suggest a significant clinical benefit in combining SRP, systemic metronidazole, and weekly professional supragingival plaque removal for the treatment of chronic periodontitis. |
| Carvalho et al. [23] | 44           | Chronic periodontitis | 3 months     | 1. Control (C): SRP + placebo ( $N=10$ )<br>2. Test 1 (T1): SRP + systemically MET (400, 3x, 10d) ( $N=12$ )<br>3. Test 2 (T2): SRP + weekly PC + placebo; ( $N=12$ )<br>4. Test 3 (T3): SRP + MET (400, 3x, 10d) + PC ( $N=10$ ) | There was a reduction in mean counts of a number of bacterial species, particularly those of the red complex, <i>Tannerella forsythensis</i> , <i>Porphyromonas gingivalis</i> , and <i>Treponema denticola</i> . All three species in this complex were significantly reduced in the subjects in groups T2 and T3. Subjects in group T2 also showed a reduction in mean counts of five of the orange complex species, <i>Campylobacter gracilis</i> , <i>Campylobacter rectus</i> , <i>Eubacterium nodatum</i> , <i>Fusobacterium periodonticum</i> , and <i>Prevotella nigrescens</i> . Of interest was the minimal change in mean counts of <i>Actinomyces</i> , purple, yellow, and green complex species in group T3. There were no significant differences in changes in mean counts of each species among treatment groups. Mean proportions of red complex species decreased from 18.4% at baseline to 3% at 90 days post therapy in group T3 ( $P < 0.01$ ), from 25.8% to 2.3% in group T2 ( $P < 0.01$ ), from 17.7% to 5.6% in group T1 ( $P < 0.05$ ) and from 19.4% to 8.8% in group C (NS). Proportions of the suspected periodontal pathogens from the orange complex were also markedly reduced in groups T2 and T3. In summary, all treatments reduced counts and proportions of red complex species. Adjunctive therapy appeared to have a greater effect and also affected members of the orange complex.  |

(continued)

Table 3.5 (continued)

| Study                      | No. patients | Periodontal condition  | Study period | Periodontal treatment   | Outcome   |
|----------------------------|--------------|--|--------------|---|---|
| Haffajee et al. [72]       | 92           | Chronic periodontitis  | 1 year       | 1. SRP + AZ (500 mg once daily for 3 days) ( $N=25$ )<br>2. SRP + MET (250, 3x, 14d) ( $N=24$ )<br>3. SRP + doxycycline (SDD, Peristat, 20 mg for 12 weeks) ( $N=20$ )<br>4. SRP ( $N=23$ ) | All groups showed clinical improvements at 12 months, with subjects receiving adjunctive agents showing a somewhat better response. Some subjects showed attachment loss at 12 months in each group ranging from 1.5% to 39% of subjects in the SDD and SRP only groups respectively. Subjects receiving AZ exhibited the largest percentage of sites showing attachment gain > 2 mm at 12 months (5.3%), while the MET group showed the lowest percentage of sites showing loss of attachment > 2 mm (0.41%). All groups exhibited a greater percentage of sites gaining attachment than losing attachment > 2 mm. Subjects receiving systemically administered AZ or MET showed greater reductions in mean PPD and AL at sites with initially deeper pockets (>6 mm) compared with the subjects receiving SRP alone or SDD. In summary, this study demonstrated that periodontal therapy provides clinical benefits and that antibiotics provide a clinical benefit over SRP alone, particularly at initially deeper periodontal pockets.                     |
| Joyston-Bechal et al. [84] | 45           | Moderate and severe chronic periodontal disease  | 22 weeks     | 1. SRP + OHI +topically CHX gel for the first 10 weeks + MET (200, 3 x, 5d)<br>2. SRP + OHI +topically CHX gel for the first 10 weeks + placebo   | The results showed that MET had no effect on plaque levels and gingival bleeding beyond the effect of OHI, scaling, and chlorhexidine gel. On the other hand, significantly greater reductions in PPD were achieved with the use of MET. These reductions were apparent only in the subjects with severe periodontal disease ( $P=4.0-6.0$ ).   |
| Joyston-Bechal et al. [83] | 28           | Moderate and severe chronic periodontal disease  | 3 years      | 1. SRP + OHI +topically CHX gel for the first 10 weeks + MET (200, 3 x, 5d)<br>2. SRP + OHI +topically CHX gel for the first 10 weeks + placebo   | No significant differences in any of the parameters between test and control groups. Moreover, the significantly greater reductions in mean PPD, achieved with the use of MET in the severe group at the end of the trial, had disappeared after 3 years. However, in subjects with mild disease, statistically significant reductions in PPD, not originally apparent, were observed 3 years later.  |
| Loesche et al. [99]        | 33           | patients with elevated proportions or levels of spirochetes in 2 or more plaque samples, i.e., 60% spirochetes | 6 weeks      | 1. SRP + MET (250, 3x, 7d) ( $N=15$ )<br>2. SRP + placebo ( $N=18$ )  | The test group exhibited a highly significant ( $P<0.01$ ) reduction in PPD and apparent gain in CAL relative to control group about those teeth that initially had PPD 4–6 mm. This pattern was also observed about teeth that initially had PPD $\geq 7$ mm. This reduction in PPD and apparent gain in CAL was associated with a significant reduction in the need for periodontal surgery in the MET-treated patients (difference 8.4 teeth per patient) compared to the positive-control patients (2.6 teeth per patient). These clinical improvements in the metronidazole group were associated with significantly lower proportions of spirochetes, selenomonads, motile rods, and <i>Prevotella intermedia</i> , and a significantly higher proportion of cocci in the plaques. In summary, these findings indicate that systemic metronidazole, when given after all the root surface debridement is completed, leads to additional treatment benefits, including a reduced need for surgery, beyond that which can be achieved by debridement alone. |

|                       |    |                                      |          |   |  |
|-----------------------|----|--------------------------------------|----------|---|--|
| Mahmood & Dolby [110] | 15 | Moderate and advanced periodontitis  | 6 months | 1. Modified Widman flap+metronidazole (200, 3x, 7d)<br>2. Modified Widman flap+placebo  | PPDs and SBIs were reduced significantly at all stages, in both groups. Probing attachment levels increased at 7 days, to significant levels only in the MET group, and subsequently PALs decreased in both groups with no significant differences between the groups. Although the differential bacterial count altered markedly in both groups at all times, only the straight rod count at 1 month was significantly ( $P<0.05$ ) lower in the MET group. In summary, MET with surgery did not exert a significantly greater beneficial effect than placebo with surgery.   |
| Palmer et al. [145]   | 90 | Moderate to advanced periodontitis   | 6 months | 1. SRP (subgingival scaling using ultrasonic scalers) (N=27)<br>2. SRP+systemic MET (200, 3x, 7d) (N=31)<br>3. SRP+local MET gel (two applications of 25% MET gel 1 week apart) (N=27)                                | Mean PPDs were reduced following treatment by greater than 1.6 mm (Group 1=1.68 mm, Group 2=1.62 mm, Group 3=1.74 mm at 6 months post treatment) but no significant differences were detected between treatment groups at any time point. Similarly, no significant differences were detectable between treatments for changes in mean CAL, BOP, PI, or proportions of bacterial morphotypes. In summary, this study does not support the routine use of adjunctive metronidazole in the nonsurgical treatment of periodontitis.   |
| Palmer et al. [146]   | 85 | Moderate to advanced periodontitis   | 6 months | 1. SRP (subgingival scaling using ultrasonic scalers) (N=27)<br>2. SRP+systemic MET (200, 3x, 7d) (N=31)<br>3. SRP+local MET gel (two applications of 25% MET gel 1 week apart) (N=27)                                | There were no differences in any clinical measure in response to the three treatment regimens at 2 or 6 months for either smokers or nonsmokers. Multiple linear regression analysis on periodontal probing depth at 6 months demonstrated that smoking was a significant explanatory factor ( $P<0.001$ ) for poor treatment outcome, whilst the presence or absence of adjunctive MET was not ( $P=0.620$ ). In summary, this study confirms that smokers have a poorer treatment response to SRP, regardless of the application of either systemic or locally applied adjunctive metronidazole.   |
| Rooney et al. [171]   | 62 | Advanced chronic periodontal disease | 6 months | 1. AM: SRP+A/M (AMO 250, 3x, 7d and MET 200, 3x, 7d) (N=15)<br>2. PM: lactate capsules and MET (MET 200, 3x, 7d) (N=16)<br>3. AP: amoxycillin and calcium lactate (N=16)<br>4. PP: lactate and calcium lactate (N=15) | PPD improved in all groups. Treatment effects were highly significantly different and always greatest in the AM and least in the PP groups. Benefits of PM and AP over PP were also noted. The mean percentage of sites with high (>6 mm) PPD reduction in the four treatment groups at 6 months compared with baseline were: AM: 1.3% from 15.9%, PM: 4.8% from 15.6%, AP: 3.8% from 14.6%, PP: 12.4% from 19.3%. CAL improved in all groups and showed the same highly significant treatment differences, again favoring AM. The mean percentage of sites with high (>6 mm) attachment loss reduction in the four treatment groups at 6 months compared with baseline: AM 6.5% from 17.1%, PM: 8.8% from 18.2%, AP 10.0% from 18.7% and PP 18.2% from 24.3%. BOP improved in all groups, particularly in AM compared to the other groups: AM 22.8% from 62.6%, PM 32.5% from 61.8%, AP 33.9% from 61.8%, PP 44.9% from 65.6%. Regarding total anaerobic and aerobic counts, the only significant difference between treatments was at 1 month where the combined treatment was significantly more effective against total anaerobic counts than the double placebo and MET and placebo. <i>P. intermedia</i> counts were always lower in active groups compared to PP and reached significance for AM and AP at 1 month and AM and PM at 3 months. In summary, the significant differences among treatment groups and the overall trend in the data, in line with other studies, support the considerable adjunctive benefits to SRP of amoxycillin and MET combined in the treatment of advanced chronic periodontal disease. |

(continued)

Table 3.5 (continued)

| Study                   | No. patients                        | Periodontal condition               | Study period | Periodontal treatment   | Outcome  |
|-------------------------|-------------------------------------|-------------------------------------|--------------|---|--|
| Saxen & Asikainen [179] | 27                                  | Localized juvenile periodontitis    | 18 months    | 1. SRP + OHI + periodontal surgery + MET (200, 3 ×, 10 d) (N=9)<br>2. SRP + OHI + periodontal surgery + tetracycline (250, 4×, 12 d) (N=9)<br>3. SRP + OHI + periodontal surgery. No medication (N=9) | By the end of the study, <i>Aggregatibacter actinomycetemcomitans</i> was suppressed to below detection level at all test sites only in the MET group, at 17/26 sites (4 patients) in the tetracycline group and at 19/26 sites (6 patients) in the control group. Clinically, all groups showed improvement. In summary, MET was more effective than tetracycline in the suppression of Aa and the suppression of <i>A. actinomycetemcomitans</i> appeared to produce better clinical results.  |
| Söder et al. [195]      | 98                                  | Moderate and advanced periodontitis | 6 months     | 1. SRP + MET (400, 3×, 7d)<br>2. SRP + placebo  | Reassessment showed statistically significant clinical improvement, with a reduction in the number of sites ≥ 5 mm in both test and control groups. Complete healing, with no PPD ≥ 5 mm, was noted in 30% of the test group and 9% of the control group. In summary, the difference is statistically significant and shows the supplementary effect of adjunctive MET in nonsurgical treatment of moderate and advanced periodontitis.  |
| Söder et al. [194]      | 64 (37 smokers and 27 non-smokers), | Severe chronic periodontal disease  | 5 years      | 1. SRP + MET (400, 3×, 7d) (N=32)<br>2. SRP + placebo (N=32)  | Nonsmoking patients who required only nonsurgical therapy in the intervention group showed statistically significant improvement in the clinical parameters after 5 years (PPD at baseline 2.8±0.2, at 7 years 2.1±0.4; CAL at baseline 3.8±0.4, at 5 years 3.1±0.4; bone height percentage at baseline 79.7±3.2, at 5 years 82.3±2.7). Patients with complete healing, defined as the absence of inflamed sites ≥ 5 mm, after 5 years were found only in the intervention group (percent of teeth with pockets ≥ 5 mm in nonsmokers with intervention: at baseline 25.9±12.9 vs at 5 years 4.7±8.4; no sites with pockets ≥ 5 mm in nonsmokers with intervention baseline: 9.6±5.6 vs at 5 years 1.6±2.8, $P<0.01$ ). Smokers responded less favorably to periodontal therapy than nonsmokers. The number of patients infected with <i>Aa</i> , <i>Pg</i> , <i>Pi</i> , and spirochetes decreased during the study. Most patients who harbored spirochetes at the end of the study had these microorganisms at the beginning. The patients considered healthy after 5 years were the same patients found to be healthy after 6 months. In summary, decisive factors in the sustained long-term improvement of patients who respond satisfactorily to treatment are probably initial SRP; a brief course of metronidazole; and regular follow-up examinations at 6-month intervals for oral hygiene and SRP. |

|                           |    |                                      |          |  |  |
|---------------------------|----|--------------------------------------|----------|--|--|
| Xajigeorgiou et al. [226] | 43 | Generalized aggressive periodontitis | 6 months | 1. SRP+debridement (ultrasomics and polishing with a rubber cup)+A/M (AMO 500, 3x, 7d+MET 500, 3x, 7d) (N=10)<br>2. SRP+debridement (ultrasomics and polishing with a rubber cup) + DOXY (200 mg of doxycycline as a loading dose and 1.00 mg/day 14 days) (N=10)<br>3. SRP+debridement (ultrasomics and polishing with a rubber cup) + MET (500, 3x, 7d) (N=12)<br>4. only SRP+debridement (ultrasomics and polishing with a rubber cup) (N=11) | No differences were observed between the four groups at any time point regarding PPD, CAL, and BOP reduction. Subjects who received adjunctive metronidazole displayed a statistically significant reduction in mean PPD (mm) after antibiotic intake (MET: baseline: 4.71±0.57; 6 weeks after SRP: 3.47±0.51; 6 months: 2.86±0.65, $P<0.05$ ). The CAL change (mm) in the MET group was baseline: 5.35±1.27, 6 weeks after SRP: 4.61±1.13; 6 months: 4.11±1.34. The BOP of the MET group during the experimental period was baseline: 0.80±0.36, 6 weeks after SRP: 0.29±0.15, 6 months: 0.21±0.31. The intake of A/M and MET, resulted in significant reduction of the percentage of sites ≥ 6 mm compared with the control group at 6 months (80% reduction for A/M, 87.7% for MET, 57.7% for controls). Adjunctive metronidazole plus amoxicillin or metronidazole alone (when <i>A.actinomycetemcomitans</i> is not involved) was effective in deep pockets of aggressive periodontitis patients. In summary, adjunctive metronidazole plus amoxicillin or metronidazole alone (when <i>A.actinomycetemcomitans</i> is not involved) is effective in deep pockets of aggressive periodontitis patients. |
| Matarazzo et al. [116]    | 43 | Chronic periodontitis in smokers     | 3 months | 1. SRP+A/M (AMO 500, 3x, 14d and MET, 400, 3x, 14d) (N=14)<br>2. SRP+MET (400, 3x, 14d) (N=14)<br>3. SRP+placebo (N=15)  | Subjects receiving A/M showed the greatest improvements in mean PPD and CAL. Both antibiotic therapies led to additional clinical benefits over SRP alone in initially shallow, intermediate, and deep sites. The SRP+A/M therapy led to the most beneficial changes in the subgingival microbial profile. These subjects showed significant reductions in the mean counts and proportions of periodontal pathogens such as <i>T.forsythia</i> , <i>P.gingivalis</i> , and <i>T.denticola</i> , and the greatest increase in proportions of host-compatible species. In summary, significant advantages are observed when systemic antibiotics are combined with SRP in the treatment of smokers with chronic periodontitis. The greatest benefits in clinical and microbiological parameters are achieved with the use of SRP + A/M.  |

SRP scaling and root planning, *PPD* periodontal probing depth, *CAL* clinical attachment level, *BOP* bleeding on probing, *GI* gingival index, *GCF* gingival crevicular fluid, *Aa* *A.actinomycetemcomitans*, *Pg* *P.gingivalis*, *Pi* *Prevotella intermedia*, *OHI* oral hygiene instruction, *SBt* Sulcus Bleeding Index, *CHX* chlorhexidine, *MET* metronidazole, *AMO* amoxicillin, *DOXY* doxycycline, *AZ* azithromycin, *CLIN* clindamycin

### 3.6.1.4 Clindamycin

Clindamycin is bacteriostatic and inhibits bacterial protein synthesis by binding to the 50 S ribosomal subunit. The drug is active against most gram-positive bacteria, including both facultative and anaerobic species. It is particularly active against gram-negative anaerobes and is very active against the gram-negative anaerobes associated with the periodontal flora [1, 219].

Clindamycin is effective as an adjuvant to SRP in the treatment of **refractory periodontitis** [108] and **generalized rapidly progressive periodontitis** [182] (Table 3.6).

Gordon et al. [63] has evaluated **the use of clindamycin hydrochloride in the treatment of patients who had previously been unsuccessfully treated with scaling, surgery, and the adjunctive use of tetracyclines** and in some instances a penicillin derivative. It was shown that clindamycin hydrochloride had an effect on the subgingival flora as demonstrated by darkfield microscopy and influenced the progression of periodontitis, by reducing gingival inflammation as measured by improvements in BOP, redness, suppuration, and PPDs. The percentage of pockets with PPDs greater than 6 mm, 4–6 mm, and 1–3 mm changed from 11% to 2%, 38–24%, and 51–74%, respectively, following clindamycin therapy as compared to scaling alone. The percentage of sites that bleed on probing decreased from 33% after scaling alone to 8% following clindamycin and scaling. Gingival redness decreased from 36% to 1% of sites.

**The use of clindamycin hydrochloride in the treatment of adult refractory periodontitis** was also evaluated by Gordon et al. [62]. Thirty patients with a history of unsuccessful treatment with scaling, periodontal surgery, and the use of tetracyclines were entered into the study. Upon entry, the suspected refractory patients were scaled several times and then monitored for the presence of active disease by probing AL measurements performed in duplicate. Active disease was defined as a 3.0 mm or greater loss in attachment from the baseline examination or the occurrence of a periodontal abscess. Scaling and clindamycin treatment **decreased the incidence of active disease** from an annual rate of 8.0–0.5% of sites per patient ( $P < 0.001$ ). The mean time required to detect the first active site increased from  $4.9 \pm 3.7$  months following scaling alone to  $16.7 \pm 7.6$  months following

scaling and clindamycin ( $P < 0.001$ ). Active sites lost an average of 3.1 mm of probing attachment following scaling alone but “gained” back 2.0 mm at 6 months and 1.5 mm at 24 months post antibiotic and scaling treatment. BOP was significantly reduced ( $P < 0.05$ ) from 31.8% of sites pre-clindamycin treatment to 12.3% at 12 months and 17.9% of sites at 24 months post clindamycin treatment.

Walker and Gordon [214] investigated **the effect of clindamycin hydrochloride, as an adjunct to scaling, on the microbiota associated with refractory periodontitis** to elucidate the probable causative bacteria associated with the disease. Microbial samples were collected from a subset of nine patients with severe adult periodontitis who had not responded to conventional treatment modalities including the use of tetracycline and other antibiotics. Microbial samples were collected from a relatively deep site determined to be actively losing attachment and a comparably deep, but quiescent, control site in each patient prior to clindamycin therapy. Samples continued to be collected from the same sites for up to 1 year post therapy. The microbial flora of each sample was enumerated by darkfield microscopy and predominant cultivable methods. Prior to clindamycin therapy, both active and control sites consisted on average of approximately 50% spirochetes and motile rods and 40% gram-negative anaerobic rods. *Bacteroides intermedius* and *P. gingivalis* (formerly *B. gingivalis*) were elevated in the active, as compared to control, sites and accounted for approximately 20% of the cultured microbiota in the former. Following treatment with clindamycin, the gram-negative components of the microbiota were either eliminated or severely suppressed. At 1 year post therapy, spirochetes and motile rods together accounted for about 15% of the microscopic flora. Total gram-negative anaerobic rods accounted for approximately 20%, and *B. intermedius* and *P. gingivalis* combined accounted for less than 2% of the cultured microbiota from historical active sites.

Unfortunately, a number of undesirable adverse effects have been associated with the use of clindamycin. Due to its acidic nature and its effect on the gram-negative intestinal bacteria, adverse effects such as diarrhea, abdominal cramping, esophagitis, and stomach irritation are relatively common. Although more frequently associated with the use of clindamycin phosphate, pseudomembranous colitis has also occurred following oral administration of clindamycin-HCl [181, 219].

**Table 3.6** Clinical studies of clindamycin in periodontal disease therapy

| Study                  | No. patients | Periodontal condition                         | Study period | Periodontal treatment  | Outcome   |
|------------------------|--------------|---|--------------|--|---|
| Walker et al. [217]    | 30           | Refractory periodontitis                      | 9 months     | 1. SRP+placebo<br>2. SRP+Augmentin (AMO 250, CLAV 125, 3x, 14d)<br>3. SRP+clindamycin (150, 4x, 10d)   | At 3 months post treatment, the clindamycin-treated group showed an average gain of 2.1 mm, the Augmentin-treated group gained 1.9 mm, and the SRP group gained 1.4 mm in attachment. The clindamycin group remained relatively stable for up to 21 months and the Augmentin group remained stable for about 15 months without additional treatment. Five of the six subjects treated with scaling alone required additional treatment within 9 months.   |
| Magnusson et al. [108] | 21           | Refractory periodontitis                      | 2 years      | 1. SRP+Augmentin (AMO 250, CLAV 125, 3x, 14d)<br>2. SRP+clindamycin (4x, 10d)<br>3. SRP  | There was no significant difference in the proportion of sites losing attachment before and after treatment (11.3% and 12.4%, respectively). However, the proportion of sites showing gain of attachment increased from 0.9% before therapy to 5.1% ( $P=0.029$ ) following selective antibiotic therapy + SRP. The remainder of sites showed no change between pre- and post-therapy monitoring periods. The progression of attachment loss in the active sites could not be completely stopped over the entire 2-year period. Although the proportion of sites losing attachment decreased from 5.1% to 2.3% ( $P>0.05$ ), the proportion of sites gaining attachment also decreased from 2.0% to 1.0% (NS). In summary, the results suggested that selected antibiotic therapy + SRP repeated every 12–15 months may be beneficial for subjects with refractory periodontitis, although it may not completely stop progressive attachment loss.  |
| Sigusch et al. [182]   | 48           | Generalized rapidly progressive periodontitis | 24 months    | 1. 2-step SRP+doxycycline (200 mg, 1x, 8d) ( $N=12$ )<br>2. 2-step SRP+MET (500 mg, 2x, 8d) ( $N=15$ )<br>3. 2-step SRP+clindamycin (150 mg, 4x, 8 days) ( $N=11$ )<br>4. 2-step SRP; no antibiotic treatment ( $N=10$ ) | 6 months after the second-step SRP, the clindamycin group showed a significantly great reduction of PPD from baseline ( $3.5 \pm 0.96$ mm from $5.7 \pm 1.06$ mm) compared with control ( $4.6 \pm 1.0$ mm from $5.9 \pm 0.70$ mm). In PPD site categories 6–9 mm and >9 mm the clindamycin treatment induced a PPD reduction of $4.2 \text{ mm}$ ( $4.2 \pm 1.06$ mm from $8.4 \pm 0.76$ mm) compared with controls ( $5.9 \pm 1.19$ mm from $8.2 \pm 1.03$ mm). A significantly high CAL gain was also noted in the clindamycin group ( $4.4 \pm 1.0$ mm from $6.1 \pm 0.96$ mm) compared with controls ( $5.7 \pm 0.96$ mm from $6.3 \pm 0.77$ mm). SBI decreased most in the metronidazole and clindamycin groups. <i>Porphyromonas gingivalis</i> and <i>Aggregatibacter actinomycetemcomitans</i> were almost completely eradicated in these two groups 24 months after SRP. In addition, the phagocytotic capacity of crevicular polymorphonuclear neutrophils was increased in groups 2 and 3 after the second step. In summary, the present results show that metronidazole and clindamycin are effective antibiotics when used adjunctively in a two-step nonsurgical procedure of SRP in RPP patients. |

PPD periodontal probing depth, CAL clinical attachment level, SBI Sulcus Bleeding Index, SRP scaling and root planning

### 3.6.1.5 Azithromycin

Azithromycin belongs to the same general class of macrolide antibiotics as erythromycin but differs in several important aspects. Unlike erythromycin, it has broad-spectrum activity against a number of bacteria including gram-negative anaerobes and provides excellent and prolonged drug concentrations in tissue and serum. Convenient dosing is a major advantage. Azithromycin has conventionally been prescribed as a 500 mg initial loading dose followed by 250 mg/day once daily for 4 days. This schedule provides therapeutic concentrations for 10 days [219]. More recently, a single dose of extended-release azithromycin (2 g) administration was introduced. Although there are several reports about its effect in sinusitis, pneumonia, and rhinosinusitis, no report for periodontitis has been shown at the moment.

In addition, azithromycin is preferentially taken up by phagocytes; therefore, its level in infected tissues is greater than in noninfected sites [121]. It has been reported that in patients receiving azithromycin (500 mg/day for 3 days), the concentrations of azithromycin in plasma, saliva, normal gingiva, and pathological tissues reached the highest values at 12 h after the last dose ( $0.37 \pm 0.05$  mg/L,  $2.12 \pm 0.30$  mg/L,  $6.30 \pm 0.68$  mg/kg, and  $11.60 \pm 1.50$  mg/kg, respectively) and then declined gradually [15]. Consistent levels of the drug have been detected in normal gingiva and pathological tissues, however, up to 6.5 days; azithromycin has been retained in target tissues for a long time after administration (saliva:  $0.52 \pm 0.09$  mg/L; gingival:  $2.87 \pm 0.63$  mg/kg; and alveolar bone:  $0.44 \pm 0.05$  mg/kg, respectively) [15, 111]. Moreover, azithromycin levels in both normal gingiva and pathological tissues exceeded the minimum inhibitory concentrations (MIC) of most pathogens involved in the pathophysiology of chronic periodontal diseases. Notably, azithromycin levels in pathological tissues were significantly higher than those in normal gingiva at 0.5, 2.5, and 4.5 days after the last dose [15]. Gomi et al. [61] have reported that after the administration of 500 mg once daily for 3 days, the azithromycin concentrations in inflamed periodontal tissues on days 4, 7, and 14 were  $2.92 \pm 1.88$ ,  $1.47 \pm 0.71$ , and  $0.54 \pm 0.27$  mg/g, respectively. Azithromycin concentration on day 7 was approximately half of that on day 4 but still remained at an effective concentration. On day 14, the drug concentration had fallen to 20% of that on day 4.

The azithromycin concentration on day 4 was more than the MIC90 for *P. gingivalis* and an effective concentration for *P. intermedia* and *A. actinomycetemcomitans* was maintained even on day 14 [61].

Azithromycin demonstrates good in vitro activity against a number of gram-negative periodontal pathogens including all serotypes of *A. actinomycetemcomitans* and *P. gingivalis* [131, 142, 143, 219]. Azithromycin is relatively nontoxic and has only a few adverse side effects, including nausea, vomiting, abdominal pain or cramping, and diarrhea. Azithromycin is excreted in human breast milk, and is therefore contraindicated in nursing mothers [219].

Azithromycin improved clinical parameters when used as an adjunct to SRP in patients with chronic [60, 72, 193], aggressive [68], and refractory periodontitis [115] (Table 3.7). It has been also revealed that azithromycin reduced gingival overgrowth in animal models [141] and in humans [37, 133, 158].

### 3.6.1.6 Spiramycin

Only two studies evaluated the additional effect of spiramycin as an adjunct to scaling and root planning in the treatment of advanced chronic periodontitis. Bain et al. [10] treated 193 patients with advanced periodontitis with SRP. All patients randomly received either spiramycin, 1,500,000 IU, twice per day (IU, bid) for 14 days (96 patients), or a visually identical placebo capsule (97 patients). Statistically significant differences in PPD, favoring spiramycin, were seen at 2 weeks ( $P < 0.0125$ ), 8 weeks ( $P < 0.0020$ ), 12 weeks ( $P < 0.0032$ ), and 24 weeks ( $P < 0.0075$ ). Spiramycin also produced a significant improvement in AL at 12 weeks ( $P < 0.0146$ ). All other clinical parameters showed no difference between drug and placebo. This study shows that spiramycin, as an adjunct to thorough SRP, provides a statistically significant improvement in PPDs for up to 24 weeks when compared with SRP alone.

In contrast, no significant intergroup differences in any of the clinical parameters measured were observed in a study performed by Al-Joburi et al. [5]. The 96 patients with advanced adult chronic periodontitis were randomly assigned to one of three groups. All groups were scaled and root-planed, with each respective group receiving spiramycin, tetracycline, or a placebo for 2 weeks. At the end of 24 weeks, although all three groups had shown clinical improvement when

**Table 3.7** Clinical studies of azithromycin in periodontal disease therapy

| Study               | No. patients | Periodontal condition                                 | Study period | Periodontal treatment  | Outcome   |
|---------------------|--------------|---|--------------|--|---|
| Dastoor et al. [41] | 30           | Moderate to advanced chronic periodontitis in smokers | 6 months     | 1. Periodontal surgery (apically positioned flap with osseous recontouring) + AZ (500, 1x, 3d) (N=15)<br>2. Periodontal surgery + placebo (N=15) | At 6 months following surgical therapy, test and control groups of surgically treated sites demonstrated statistically significant reductions in PPD ( $-0.83$ and $-0.98$ mm for test and control groups, respectively). At 6 months, only the test group had statistically significant gains in CAL ( $-0.27$ mm) compared to baseline. Nonetheless, there were no significant differences between the groups at any time point for PPD or CAL. The additional use of AZM did not enhance this improvement, nor did it promote reduction of cross-linked telopeptide of type I collagen levels in GCF. Compared to the control group, the test group had significantly better WHI scores at 1 month, significantly less GI at 2 weeks, and sustained reductions of red-complex bacteria with trypsin-like enzyme activity at 3 months. For nonsurgery teeth, only the test group showed significant gains in overall CAL compared to baseline. In summary, the findings of this pilot study demonstrated that in heavy smokers, adjunctive systemic AZM in combination with pocket reduction surgery did not significantly enhance PPD reduction or CAL gain. However, the clinical value of adjunctive AZM may be appreciated by more rapid wound healing, less short-term gingival inflammation, and sustained reductions of peripathogenic bacteria. |
| Gomi et al. [61]    | 34           | Severe chronic periodontitis                          | 25 weeks     | 1. Toothbrushing instructions + FM-SRP + AZ (500, 1x, 3d) (N=17)<br>2. Toothbrushing instructions + FM-SRP (N=17)                                | A statistically significant difference ( $P<0.001$ ) in PPD was observed between the test and control groups at 13 and 25 weeks (test: PPD at 13 and 25 weeks: $2.21 \pm 0.33$ and $2.36 \pm 0.76$ mm, respectively; controls: $3.28 \pm 0.41$ and $3.30 \pm 0.36$ mm, respectively). There was a statistically significant difference ( $P<0.01$ ) in BOP rate between test and control groups at 13 and 25 weeks from baseline ( $4.46\% \pm 3.27\%$ and $5.35\% \pm 2.91\%$ in the test, $12.08\% \pm 7.15\%$ and $12.91\% \pm 7.62\%$ in controls). In the test group, no periodontopathogenic bacteria could be detected 1 week after full-mouth SRP to 13 weeks after baseline. In the control group, <i>Porphyromonas gingivalis</i> , <i>Tannerella forsythensis</i> , and <i>Prevotella intermedia</i> were detected 5 weeks after baseline. In summary, azithromycin is detectable in inflamed periodontal tissues $\geq 14$ days after systemic administration; it is associated with clinical and microbiologic improvement.  |

(continued)

**Table 3.7** (continued)

| Study                | No. patients | Periodontal condition    | Study period | Periodontal treatment   | Outcome  |
|----------------------|--------------|--------------------------|--------------|---|--|
| Haas et al. [68]     | 24           | Aggressive periodontitis | 1 year       | 1. SRP+AZ (500, 1x, 3d) ( <i>N</i> =12)<br>2. SRP+placebo ( <i>N</i> =12)   | <p>The use of AZ resulted in a significantly higher reduction in mean PPD of approximately 1 mm, compared with placebo (<math>2.88 \pm 0.23</math> vs <math>1.85 \pm 0.36</math>, <math>P=0.025</math>). The additional reduction in PPD was similar in sites that at baseline had moderate (4–6 mm) (0.73 mm: <math>2.02 \pm 0.14</math> vs <math>1.25 \pm 0.17</math>, <math>P=0.003</math>) or deep PPD depths (<math>\geq 7</math> mm) (0.77 mm: <math>3.49 \pm 0.23</math> vs <math>2.76 \pm 0.51</math>, <math>P=0.21</math>). The AZ group showed higher CAL improvement than the control group when all eligible sites (<math>\geq 4</math> mm) were considered in the analysis, with a borderline <math>P</math> value (<math>1.68 \pm 0.20</math> vs <math>0.97 \pm 0.29</math>, <math>P=0.05</math>). There were no significant differences in mean CAL gain between groups in sites with moderate or deep PPD at baseline. The two treatment groups showed similar changes in BOP, with both groups showing a decrease of approximately 45% of bleeding sites (<math>45.04 \pm 3.32</math> in test and <math>44.46 \pm 3.89</math> in control; <math>P=0.91</math>). Compared with the placebo group, the AZ group had significantly higher percentage of teeth with a PPD reduction of <math>\geq 1</math> and <math>\geq 2</math> mm from baseline to 12 months post-operatively (<math>\geq 1</math> mm: <math>97.01 \pm 1.81</math> vs <math>82.64 \pm 4.76</math>, <math>P=0.01</math>; <math>\geq 2</math> mm: <math>81.34 \pm 4.00</math> vs <math>57.85 \pm 8.21</math>, <math>P=0.017</math>). Additionally, patients taking AZ had a significantly higher percentage of teeth with <math>\geq 1</math> mm decrease in CAL than those taking the placebo (<math>81.34 \pm 3.28</math> vs <math>63.63 \pm 7.29</math>, <math>P=0.037</math>). Patients in the placebo group had an increase in the percentage of teeth showing loss of attachment <math>\geq 1</math> mm during the study period (<math>11.57 \pm 3.43</math> vs <math>2.24 \pm 0.97</math>, <math>P=0.015</math>). In summary, the adjunctive use of azithromycin has the potential to improve periodontal health of young patients with AgP.</p> |
| Haffajee et al. [72] | 92           | Chronic periodontitis    | 1 year       | 1. SRP+AZ (500, 1x, 3d) ( <i>N</i> =24)<br>2. SRP+MET (250, 3x, 14d) ( <i>N</i> =25)<br>3. SRP+doxycycline (SDD, Periostat, 20 mg for 12 weeks) ( <i>N</i> =20)<br>4. SRP alone ( <i>N</i> =23) | <p>All groups showed clinical improvements at 12 months, with subjects receiving adjunctive agents showing a somewhat better response. Some subjects showed attachment loss at 12 months in each group ranging from 15% to 39% of subjects in the SDD and SRP only groups respectively. Subjects receiving AZ exhibited the largest percentage of sites showing attachment gain <math>&gt; 2</math> mm at 12 months (5.3%), while the MET group showed the lowest percentage of sites showing loss of attachment <math>&gt; 2</math> mm (0.41%). All groups exhibited a greater percentage of sites gaining attachment than losing attachment <math>&gt; 2</math> mm. Subjects receiving systemically administered AZ or MET showed greater reductions in mean PPD and AL at sites with initially deeper pockets (<math>&gt; 6</math> mm) compared with the subjects receiving SRP alone or SDD. In summary, this study, demonstrated that periodontal therapy provides clinical benefits and that antibiotics provide a clinical benefit over SRP alone, particularly at initially deeper periodontal pockets.</p>  |
| Haffajee et al. [70] | 92           | Chronic periodontitis    | 1 year       | 1. SRP+AZ (500, 1x, 3d) ( <i>N</i> =25)<br>2. SRP+MET (250, 3x, 14d) ( <i>N</i> =24)<br>3. SRP+doxycycline (SDD, Periostat, 20 mg for 12 weeks) ( <i>N</i> =20)<br>4. SRP ( <i>N</i> =23)       | <p>All treatments reduced counts of red-complex species at 12 months, although no significant differences were detected among treatment groups for most species at all time points. Both antibiotics significantly reduced counts of red-complex species by 2 weeks. Percentage of resistant isolates increased in plaque samples in all adjunctive treatment groups, peaking at the end of administration, but returned to pretreatment levels by 12 months. In summary, the significant reduction of red- and orange-complex species at 2 weeks in the subjects receiving SRP plus azithromycin or metronidazole may have contributed to a better clinical response in these treatment groups. Therapy did not appear to create lasting changes in the percentage of resistant isolates or sites harboring resistant species</p>   |

|                          |    |  |          |   |  |
|--------------------------|----|--|----------|---|--|
| Mascarenhas et al. [115] | 30 | Refractory moderate to advanced chronic periodontitis in smokers | 6 months | 1. SRP+AZ (two 250 mg tablets the first day and one 250 mg tablet for each of the next 4 days) (N=15)<br>2. SRP(N=15) | In shallow sites (<4 mm), the data demonstrate that both control and test groups showed a reduction in PPD compared to baseline (0.02 and 0.43 mm, respectively). A difference that was statistically significant for the AZM group at both the 3 and 6 months time points compared to baseline ( $P < 0.05$ ). The test group showed a statistically significant CAL gain (0.55 versus 0.11 mm). A statistically significant difference ( $P < 0.05$ ) between groups was only identified at baseline. In moderate sites (PPD=4–6 mm), there was a statistically significant difference between groups for the reduction of PPD at 6 months (1.0 and 1.7 mm for control and test groups, respectively, $P < 0.05$ ). CAL levels were statistically different between groups at baseline (4.95 and 5.47 mm, $P < 0.05$ ) but were identical at 6 months. Drug therapy resulted in a greater reduction of PPD in deep sites (>6 mm) compared to controls (3.52 versus 1.98 mm, $P < 0.05$ ), and this difference was sustained for the duration of the study. CAL changes were larger for the test group compared to the control and the differences were significantly different at 6 months (2.56 versus 1.32 mm, $P < 0.05$ ). In addition, patients receiving the drug showed a trend increase CAL gain at 6 months while control group showed almost no further gain after 3 months. No statistically significant differences between groups on mean BOP and ICIP levels during the course of the study were noted. In summary, the utilization of AZ in combination with SRP improves the efficacy of nonsurgical periodontal therapy in reducing PPD and improving ALs in smokers with moderate to advanced attachment loss. |
| Smith et al. [193]       | 44 | Adult periodontitis  | 22 weeks | 1. SRP+AZ (500, 1×, 3d at week) (N=23)<br>2. SRP+placebo (N=21)   | The mean PPD of initially deep pockets in the AZ and placebo groups demonstrated improvements between baseline and 22 weeks (adjusted means for both groups initially being 6.76 mm and then at 22 weeks 3.67 mm for the AZ group and 4.54 mm for the control group). The mean periodontal probing depths were statistically significantly ( $P < 0.05$ ) shallower in the AZ group than in the placebo group at all time intervals after the drug had been administered. The clinical data showed that by week 22, 5.6% pockets initially >5 mm deep remained above that level in the 23 patients taking AZ, compared with 23.3% in the 21 patients taking the placebo. Also at week 22, for pockets initially 4 mm or more, the test group had 26.1% pockets >3 mm deep compared with 44.3% in the control group. There were fewer pockets failing to improve in periodontal probing depth (AZ group 6.6%, control group 21.6%). Fewer pockets of initial PPD ≥ 4 mm continued to bleed on probing in the test group (46.9%) when compared with the control group (55.6%). PPDs initially 4–5 mm or 6–9 mm analyzed by analysis of covariance showed lower mean PPDs in the patients on AZ, at weeks 6, 10 and 22 ( $P < 0.01$ ). For pockets initially 4–9 mm deep, the percentage of pockets bleeding was statistically significantly lower in the AZ group as compared with the control at weeks 6, 10 ( $P = 0.02$ ), but the effect was not seen at week 22 ( $P = 0.69$ ). In summary, azithromycin may be a useful adjunct in the treatment of adult periodontitis, particularly where deep pockets are present.  |

WHI wound healing indices, SDD sub-antimicrobial dose doxycycline, GCF gingival crevicular fluid, FM-SRP one-stage full-mouth scaling and root planning, PM-SRP partial-mouth scaling and root planning, AZ Azithromycin, SRP scaling and root planning, PPD periodontal probing depth, CAL clinical attachment level, BOP bleeding on probing, BI bleeding index, GI gingival index, GCF gingival crevicular fluid, CHX chip chlorhexidine controlled-delivery device, A/M amoxicillin/metronidazole

compared to the baseline data, there were no significant intergroup differences in any of the clinical parameters measured. While the proportion of spirochetes was significantly decreased ( $P < 0.05$ ) at 2- and 8-week intervals in both tetracycline and spiramycin groups (26% to 0.04% and 28% to 0.04%, respectively), compared to the placebo group (30% to 7%), only in the spiramycin group was the proportion of spirochetes significantly lower than the placebo group at the 24-week interval (3% and 11%, respectively).

### 3.6.1.7 Penicillins

The penicillins are a broad class of antibiotics that inhibit bacterial cell wall synthesis and directly result in the death of the cell. All penicillins consist of a  $\beta$ -lactam ring, a thiazolidine ring, and an acyl side chain. Substitutions on the acyl side chain have yielded a wide variety of penicillin compounds with vastly different properties. These include improved stability to gastric acid, improved absorption, and higher serum concentrations, and activity against gram-negative as well as gram-positive bacteria. Allergic hypersensitivity is the most common adverse reaction. Exposure to any of the penicillins may precipitate an allergic reaction in a susceptible individual. Reported allergies to penicillins are relatively common and caution is advised [219].

Amoxicillin, a semisynthetic penicillin, has excellent activity against both gram-negative and gram-positive bacteria, is absorbed well following oral administration, and penetrates into the GCF. Unfortunately, amoxicillin is also highly susceptible to bacterial  $\beta$ -lactamases.  $\beta$ -Lactamase is an enzyme produced by a number of different bacteria which hydrolyzes the  $\beta$ -lactam ring. Hydrolysis of this ring destroys all antimicrobial activity of the penicillin [219].

Only one study assessed amoxicillin alone. Rooney et al. [171] compared the adjunctive benefits to SRP of amoxicillin and metronidazole alone and combined. Sixty-six subjects less than 46 years of age with advanced chronic periodontal disease participated in this randomized, double-blind, four parallel treatment group designed study. All subjects received quadrant SRP and then were prescribed amoxicillin capsules (250 mg) and metronidazole tablets (200 mg), lactate capsules and metronidazole, amoxicillin and calcium lactate tablets, or lactate and calcium lactate. All

medication was three of each per day for 7 days. The clinical examinations were performed pretreatment, and 1, 3, and 6 months post treatment. For the amoxicillin group compared to the placebo group differences reached significance for high sites ( $PD \geq 6$  mm) at all times ( $P$  ranged  $< 0.05$ –0.001) but at low sites (PPD 0–3 mm) differences were significant at 3 and 6 months ( $P < 0.01$ –0.001) but not 1 month ( $P < 0.05$ ). Regarding percentage of BOP, differences between the amoxicillin group compared to the placebo group were mainly nonsignificant.

Amoxicillin-clavulanate was the first  $\beta$ -lactam- $\beta$ -lactamase inhibitor combination introduced into clinical practice, in 1981 in the UK and in 1984 in the USA, and remains the only combination available for oral use. The activity of amoxicillin against susceptible bacteria that do not possess a  $\beta$ -lactamase (e.g., streptococci, enterococci, *Escherichia coli*, and *Listeria* spp.) is not improved by the use of clavulanate. However, the addition of clavulanate significantly expands amoxicillin's spectrum to include penicillinase-producing *Staphylococcus aureus*, *Haemophilus influenzae*, *Moraxella catarrhalis*, *Bacteroides* spp., *Neisseria gonorrhoeae*, *E. coli*, *Klebsiella* spp., and *Proteus mirabilis*. The oral availability of amoxicillin-clavulanate makes it well suited for the outpatient clinic, and this  $\beta$ -lactam- $\beta$ -lactamase inhibitor combination exerted its greatest impact on the treatment of community-acquired respiratory infections. Additionally, amoxicillin-clavulanate is sometimes used as an oral equivalent ("step-down therapy") of ampicillin-sulbactam or ticarcillin-clavulanate in the treatment of skin, soft tissue, abdominal, and gynecological infections [45].

In theory, one would expect amoxicillin-clavulanate in combination with mechanical debridement to be relatively effective in the eradication of both gram-negative and gram-positive periodontal pathogens. Although the data are limited, clinical studies do not support the use of Augmentin as a particularly effective adjunctive antibiotic. It may provide some benefit over mechanical therapy for certain periodontitis patients; however, other antibiotics used adjunctively appear more effective [219]. It had been shown that the four most frequently occurring microorganisms in a group of 56 advanced periodontitis patients were *Fusobacterium nucleatum* (38/56), *Peptostreptococcus micros* (34/56), *P. gingivalis* (32/56), and *Prevotella oralis* (32/56). These four bacterial species had been

subjected to a susceptibility testing towards amoxicillin/clavulanate. All isolates of *P. gingivalis* and *P. oralis* were shown to be susceptible, whereas *F. nucleatum* revealed isolates being nonsusceptible to amoxicillin-clavulanate in 29% of the investigated bacterial cultures and *P. micros* in 12%. When performing SRP in those 56 subjects, some of them could be identified as not responding to this therapeutic approach [88]. More recently, Kleinfelder et al. [87, 89] compared antibiotic susceptibility of putative periodontal pathogens before and after systemic therapy with amoxicillin-clavulanate as adjunct to periodontal surgery. The results showed that even if pretreatment susceptibility testing reveals putative periodontal pathogens such as *F. nucleatum* or *P. micros* as being susceptible to amoxicillin-clavulanate, these microorganisms might not in all cases be targeted by the antibiotic agent tested. Furthermore, the outcomes of conventional methods of susceptibility testing have to be interpreted very carefully when used for treatment of plaque-related diseases. The biofilm phenomena might be in part responsible for the discrepancy between susceptibility in vitro and clinical efficacy of systemic antimicrobial therapy in vivo. In addition, if the end point of systemic antibiotic treatment as adjunct to conventional therapy is elimination of *F. nucleatum* or *P. micros* in patients harboring these organisms, the use of amoxicillin-clavulanate appears not to be justified [87, 89] (Table 3.8).

In **adult periodontitis patients**, initial periodontal therapy in conjunction with systemic amoxicillin plus clavulanic acid was no more effective than initial periodontal therapy alone [224]. In a subsequent study, performed in patients with advanced chronic periodontal disease, benefits of amoxicillin treatment over placebo (lactate and calcium lactate) were noted in terms of mean percentage of sites with high (>6 mm) PPD and attachment loss reduction. No differences were noted regarding total anaerobic and aerobic counts between the two groups [171].

Among others, amoxicillin with potassium clavulanate, a potent  $\beta$ -lactamase inhibitor, has been used as adjunct to conventional periodontal therapy in **refractory periodontitis patients** [108, 217]. Magnusson et al. [108] evaluated the effect of non-surgical periodontal therapy with the adjunct of a selected antibiotic in subjects diagnosed with refractory periodontal disease. The study group was formed by 21 patients with a history of periodontal surgery,

tetracycline therapy, and regular maintenance. After detection of disease activity and whole plaque susceptibility assessment, all subjects received SRP in addition to antibiotic or placebo therapy. Based on the result of the susceptibility test, subjects in the antibiotic group were treated either with Augmentin or with clindamycin. The results demonstrated that in subjects with refractory periodontal disease there was no significant difference in the proportion of sites losing attachment before and after treatment (11.3% and 12.4%, respectively) over a 2-year post-therapy observation period. However, the proportion of sites showing gain of attachment increased from 0.9% before therapy to 5.1% ( $P=0.029$ ) following selective antibiotic therapy when combined with SRP. The remainder of sites showed no change between pre- and post-therapy monitoring periods. The progression of attachment loss in the active sites could not be completely stopped over the entire 2-year period. After 12–15 months following therapy, there was a tendency towards new loss of attachment and an increase of PPD. It was suggested that SRP together with selected antibiotic therapy repeated every 12–15 months may be beneficial for these subjects although it may not completely stop progressive attachment loss [108].

It was reported that Augmentin may enhance the results of guided tissue regeneration, patients on Augmentin therapy experiencing a significantly higher mean probing attachment gain (36.5% of potential gain to the cemento-enamel junction) than patients who did not receive antibiotic therapy following surgery (3.3 vs 2.4 mm). At the time of membrane removal the whole membrane in the Augmentin group demonstrated significantly fewer organisms than the whole membrane in the control group [139].

The amoxicillin-clavulanate regime was also effective in the short-term treatment of **periodontal abscesses** in periodontitis patients [79].

### 3.6.2 Combination Antimicrobial Therapy

Since the subgingival microbiota in destructive periodontal disease consists of various putative pathogens that may differ in antimicrobial susceptibility, the use of a combination of two or more antibiotics may represent

**Table 3.8** Clinical studies of amoxicillin and amoxicillin plus clavulanate in periodontal disease therapy

| Study                  | No. patients | Periodontal condition        | Study period | Periodontal treatment  | Outcome  |
|------------------------|--------------|------------------------------|--------------|--|--|
| Demolon et al. [43]    | 15           | Class II furcation invasions | 4 weeks      | 1. SRP + GTR + CHX (N=7)<br>2. SRP + GTR + CHX + Augmentin (AMO 250, CLAV 125, 3x, 10d) (N=8)    | At baseline no parameter showed statistical differences between groups or sites. At week 1 significantly greater levels of <i>Prevotella intermedia</i> type I ( $P < 0.05$ ) and <i>Fusobacterium nucleatum</i> ( $P < 0.01$ ) were found in group 1. At week 4, paper-point samples from test sites ( $P < 0.05$ ) and e-PTFE materials ( $P < 0.001$ ) showed significantly higher presence of <i>Bacteroides forsythus</i> in group 1. No significant microbial changes were found for control sites over time or between groups. The total bacterial load at test sites over time increased similarly for patients administered or not administered the antibiotic. Clinical signs of inflammation were significantly greater in group 1 and associated with the presence of <i>B. forsythus</i> ( $P < 0.01$ ).  |
| Demolon et al. [42]    | 15           | Class II furcation invasions | 1 year       | 1. SRP + GTR + CHX (N=7)<br>2. SRP + GTR + CHX + Augmentin (AMO 250, CLAV 125, 3x, 10d) (N=8)    | After 1 year, the reduction in mean periodontal probing depth of the furcation invasions was $2.0 \pm 1.2$ mm for group 1 and $1.8 \pm 1.1$ mm for group 2. An overall gain of 0.8 mm of clinical attachment was found. Twenty-two of the 24 sites were re-entered. Wide individual variations were found but the changes between pretreatment and 1-year data for any of six linear measurements of hard tissue landmarks did not differ between groups or between pretreatment and re-entry. A combination of an overall ABL of 0.4 mm at the crest and 0.3 mm gain of bone at the bottom of the furcation defects was found. Volumetric analysis indicated an average 32% bone fill for both groups, ranging from a decrease in defect volume by 84% (gain) to an increase of the size of the furcation invasion by 66% (loss). A decrease in defect volume >30% was found at seven sites from each group. In summary, the antibiotic may have controlled initial inflammation, but 12 months later it had no direct effect on bone regeneration or soft tissue attachment. |
| Magnusson et al. [108] | 21           | Refractory periodontitis     | 2 years      | 1. SRP + Augmentin (AMO 250, CLAV 125, 3x, 14d)<br>2. SRP + clindamycin (CLIN 4x, 10d)<br>3. SRP | There was no significant difference in the proportion of sites losing attachment before and after treatment (11.3% and 12.4%, respectively). However, the proportion of sites showing gain of attachment increased from 0.9% before therapy to 5.1% ( $P=0.029$ ) following selective antibiotic therapy + SRP. The remainder of sites showed no change between pre- and post-therapy monitoring periods. The progression of attachment loss in the active sites could not be completely stopped over the entire 2-year period. Although the proportion of sites losing attachment decreased from 5.1% to 2.3% ( $P>0.05$ ), the proportion of sites gaining attachment also decreased from 2.0% to 1.0% (NS). In summary, the results suggested that selected antibiotic therapy + SRP repeated every 12–15 months may be beneficial for subjects with refractory periodontitis, although it may not completely stop progressive attachment loss.   |

|                     |    |                                      |           |   |  |
|---------------------|----|--------------------------------------|-----------|---|--|
| Rooney et al. [171] | 62 | Advanced chronic periodontal disease | 6 months  | 1. AM: SRP+A/M (AMO 250, 3x, 7d and MET 200, 3x, 7d) (N=15)<br>2. PM: lactate capsules and MET (MET 200, 3x, 7d) (N=16)<br>3. AP: amoxycillin and calcium lactate (N=16)<br>4. PP: lactate and calcium lactate (N=15) | PPD improved in all groups. Treatment effects were highly significantly different and always greatest in the AM and least in the PP groups. Benefits of PM and AP over PP were also noted. The mean percentage of sites with high (>6 mm) PPD reduction in the four treatment groups at 6 months compared with baseline were: AM: 1.3% from 15.9%, PM: 4.8% from 15.6%, AP: 3.8% from 14.6%, PP: 12.4% from 19.3%. CAL improved in all groups and showed the same highly significant treatment differences, again favoring AM. The mean percentage of sites with high (>6 mm) AL reduction in the four treatment groups at 6 months compared with baseline: AM 6.5% from 17.1%, PM: 8.8% from 18.2%, AP 10.0% from 18.7%, and PP 18.2% from 24.3%. BOP improved in all groups, particularly in AM compared to the other groups: AM 22.8% from 62.6%, PM 32.5% from 61.8%, AP 33.9% from 61.8%, and PP 44.9% from 65.6%. Regarding total anaerobic and aerobic counts, the only significant difference between treatments was at 1 month where the combined treatment was significantly more effective against total anaerobic counts than the double placebo and metronidazole and placebo. <i>P. intermedia</i> counts were always lower in active groups compared to PP and reached significance for AM and AP at 1 month and AM and PM at 3 months. In summary, the significant differences among treatment groups and the overall trend in the data, in line with other studies, support the considerable adjunctive benefits to SRP of amoxycillin and metronidazole combined in the treatment of advanced chronic periodontal disease. |
| Walker et al. [217] | 30 | Refractory periodontitis             | 9 months  | 1. SRP+placebo<br>2. SRP+Augmentin (AMO 250, CLAV 125, 3x, 14d)<br>3. SRP+clindamycin (150, 4x, 10d)  | At 3 months post treatment, the clindamycin-treated group showed an average gain of 2.1 mm, the Augmentin-treated group gained 1.9 mm, and the SRP group gained 1.4 mm in attachment. The clindamycin group remained relatively stable for up to 21 months and the Augmentin group remained stable for about 15 months without additional treatment. Five of the six subjects treated with scaling alone required additional treatment within 9 months.  |
| Winkel et al. [224] | 21 | Adult periodontitis                  | 12 months | 1. SRP+amoxicillin (AMO 500+CLAV 125, 3x, 10d) (Augmentin) (N=10)<br>2. SRP+placebo (N=11)  | The changes in BOP and GI in the course of the study were similar in both groups. The mean full-mouth PPD reduction of 1.0 mm in the placebo group and 0.9 mm in the test group was observed during the first 3 months. No further reduction in PPD was noticed during the study period in either group. There was no statistically significant difference in the PPD reduction between the two groups. The change in CAL from baseline to 3 months amounted to 0.5 mm in both groups. Between 3 and 12 months, CAL did not change in either group. In both groups, treatment resulted in a decrease in the number of spirochetes and motile rods in positive patients, but no significant differences between either group were noted in any of the dark field microscopy observations. In summary, in adult periodontitis patients, initial periodontal therapy in conjunction with systemic amoxicillin plus clavulanic acid was no more effective than initial periodontal therapy alone.  |

SRP scaling and root planning, *ABL* alveolar bone loss, *PPD* periodontal probing depth, *CAL* clinical attachment level, *BOP* bleeding on probing, *BI* bleeding index, *GI* gingival index, *GCF* gingival crevicular fluid, *CHX chip* chlorhexidine controlled-delivery device, *A/M* amoxicillin/metronidazole

a valuable approach in periodontal chemotherapy. Combination therapy may help to broaden the antimicrobial range of the therapeutic regimen beyond that attained by any single antibiotic, to prevent or forestall the emergence of bacterial resistance by using agents with overlapping antimicrobial spectra and to lower the dose of individual antibiotics by exploiting possible synergy between two drugs against targeted organisms. The disadvantages of combination drug therapy are increased drug interactions with improperly selected antibiotics. A bactericidal antibiotic ( $\beta$ -lactam drug or metronidazole) should not be used simultaneously with a bacteriostatic agent (tetracyclines) because the bactericidal agent exerts activity during cell division that is impaired by the bacteriostatic drug [207].

Antibiotics that are bacteriostatic (e.g., tetracycline) generally require rapidly dividing microorganisms to be effective. They do not function well if a bactericidal antibiotic (e.g., amoxicillin) is given concurrently. When both types of drugs are required, they are best given serially, not in combination [85].

Valuable combination therapies include metronidazole–amoxicillin (250–375 mg of each 3 $\times$  daily for 8 days) for *A. actinomycetemcomitans* and various anaerobic periodontal infections, and metronidazole–ciprofloxacin (500 mg of each 2 $\times$  daily for 8 days) for mixed anaerobic and enteric rod/pseudomonas periodontal infections [187].

The combination of mechanical therapy and **systemic application of amoxicillin and metronidazole combined** has been shown to resolve periodontal inflammation effectively in generalized aggressive periodontitis patients [65], with stability of the improved clinical attachment documented for up to 5 years [19]. Primarily, amoxicillin–metronidazole had been introduced as a specific treatment for periodontal infections with a detected presence of the periodontal pathogen *A. actinomycetemcomitans* (previously *Actinobacillus actinomycetemcomitans*) [209]. However, this drug regimen is more efficacious than the respective single drugs or placebo, even if empirically prescribed without diagnostic identification of detectable pathogens in patients exhibiting advanced periodontal disease [171]. Accordingly, amoxicillin–metronidazole is considered to be an antibiotic regimen of first choice and is used widely [2, 86, 188] (Table 3.9). The meta-analysis performed by Herrera et al. [80] showed a statistically significant additional effect of amoxicillin plus metronidazole

with regard to CAL change ( $P=0.001$ ) for initial PPD>6 mm.

Another combination antibiotic therapy of periodontal disease that was proposed is the association of **metronidazole and ciprofloxacin** (Slots, 2000; Beikler et al., 2004; van Winkelhoff et al., 2005).

Slots et al. [185] examined the sensitivity of isolates of enteric rods and pseudomonads in 844 adult periodontitis patients. These organisms were recovered from 13.5% of the study subjects. Ciprofloxacin exhibited the highest inhibitory activity of the 14 oral antimicrobial agents tested, virtually all enteric rods and pseudomonas, as well as against *A. actinomycetemcomitans*, *Eikenella corrodens*, *Capnocytophaga*, and *Staphylococcus*, but was largely inactive against most obligate anaerobes. It was suggested that in treating mixed anaerobic/facultative infections like periodontitis, ciprofloxacin should therefore be combined with an antimicrobial agent that is active against anaerobes, such as metronidazole.

When subgingival plaque samples of patients with progressive periodontitis and pus of odontogenic abscesses were tested, it was revealed that gram-negative anaerobes (*Prevotella* spp., *Porphyromonas* spp., *Fusobacterium* spp.) were highly susceptible to clindamycin and metronidazole [47].

Also no strains of *A. actinomycetemcomitans* demonstrated resistance to amoxicillin, amoxicillin–clavulanic acid combination, pristinamycin, or ciprofloxacin. Pristinamycin and ciprofloxacin appeared as effective alternative monotherapies against *A. actinomycetemcomitans* [107, 131]. As eradication of *A. actinomycetemcomitans* from the oral cavity seems to be a prerequisite for successful therapeutic outcome in patients periodontally infected with the organism, clinical and microbiological parameters were monitored in ten adult patients with *A. actinomycetemcomitans*-associated periodontitis during successive nonsurgical and adjunctive metronidazole plus amoxicillin (or ciprofloxacin) therapy. At baseline the organism was isolated in  $47\pm29\%$  subgingival and  $64\pm31\%$  extrarevicular samples. Six weeks following subgingival scaling, *A. actinomycetemcomitans* was detected in  $37\pm30\%$  subgingival and  $55\pm38\%$  extrarevicular samples (n.s.). Three months after antibiotic therapy, the organism was recovered from only one patient, revealing that eradication of *A. actinomycetemcomitans* seems to be possible with adjunctive antibiotic treatment. Statistical analysis revealed that the

**Table 3.9** Clinical studies of combination antibiotic therapy in periodontal disease

| Study                 | No. patients | Periodontal condition              | Study period | Periodontal treatment   | Outcome   |
|-----------------------|--------------|------------------------------------|--------------|---|---|
| Berglundh et al. [13] | 16           | Advanced periodontal disease       | 2 years      | 1. A/M (AMO 375, 2x, 14d and MET 250, 3x, 14d)<br>2. SRP+A/M<br>3. Placebo<br>4. SRP  | The systemic administration of A/M resulted in (1) an improvement of the periodontal conditions, (2) elimination/suppression of putative periodontal pathogens such as <i>Aggregatibacter actinomycetemcomitans</i> , <i>Porphyromonas gingivalis</i> , <i>Prevotella intermedia</i> , and (3) reduction in the size of the inflammatory lesion. The antibiotic regimen alone, however, was less effective than mechanical therapy with respect to reduction of BOP-positive sites, PPD reduction, and CAL gain. In summary, the combined mechanical and systemic antibiotic therapy (group 2) was more effective than mechanical therapy alone in terms of improvement of clinical and microbiological features of periodontal disease.  |
| Cionca et al. [35]    | 51           | Moderate to advanced periodontitis | 6 months     | 1. Full-mouth periodontal debridement, performed within 48 hours + A/M (AMO 375, 3x, 7d, and MET 500, 3x) ( $N=25$ )<br>2. Full-mouth periodontal debridement, performed within 48 h + placebo ( $N=26$ ) | After treatment, in the placebo group, a mean of 4.4 and 3.0 pockets >4 mm with BOP were noted after 3 and 6 months, respectively, whereas 1.3 sites were detected in the test group after 3 months ( $P=0.02$ ) and only 0.4 pockets were present after 6 months ( $P=0.005$ ). <i>A. actinomycetemcomitans</i> could not be detected in the antibiotic group after treatment. However, in the placebo group, three of six subjects positive for <i>A. actinomycetemcomitans</i> continued to be positive. Lower frequencies were also noted in the test group for <i>P. gingivalis</i> ( $P=0.013$ ) and <i>Tannerella forsythia</i> (previously <i>T. forsythensis</i> ) ( $P=0.007$ ). In summary, excellent clinical results in the antibiotics group were obtained regardless of the presence or absence of six classic periodontal pathogens prior to treatment.   |
| Cionca et al. [34]    | 51           | Chronic periodontitis.             | 6 months     | 1. Full-mouth periodontal debridement, performed within 48 h + A/M (AMO 375, 3x, 7d, and MET 500, 3x) ( $N=25$ )<br>2. Full-mouth periodontal debridement, performed within 48 h + placebo ( $N=26$ )     | At 3 months, significant and clinically relevant improvements were observed in patients in both groups. In addition, significantly better results were noted in subjects treated with full-mouth SRP+A/M to those receiving placebo. The mean PPD was $3.2 \pm 0.3$ mm in the placebo group and $3.0 \pm 0.3$ mm in the test group ( $P=0.02$ ). In addition, $25.5 \pm 10.2\%$ and $19.3 \pm 8.8\%$ of sites exhibited BOP in the placebo and test groups ( $P=0.02$ ). The mean PS, GI, and clinical AL gain in the control and test subjects were not significantly different. At 6 months, the mean PPD was $3.1 \pm 0.3$ and $3.0 \pm 0.2$ mm in the placebo and test groups, respectively ( $P=0.05$ ). The most important finding related to the absolute number of residual pockets >4 mm with BOP. The overall average number of such sites was 1.8 per subject, with a 95% confidence interval of zero to seven sites. However, in the test group, only $0.4 \pm 0.8$ such pockets were still detected on average compared with controls ( $3.0 \pm 4.3$ ) ( $P=0.005$ ). The number of persisting bleeding pockets was 7.5 times greater if the subjects had not received the antibiotics after full-mouth SRP. The mean PS, GI, REC, CAL, and BOP were not significantly different. In pockets initially deeper than 4 mm, mean PPD decreased from $5.6 \pm 0.4$ mm at baseline to $3.6 \pm 0.5$ mm at 3 months and to $3.4 \pm 0.5$ mm at 6 months in the placebo group. In the test group, pockets >4 mm decreased from $5.4 \pm 0.4$ mm at baseline to $3.2 \pm 0.3$ mm at 3 months and to $3.2 \pm 0.3$ mm at 6 months. The difference in mean PPD in this subset of sites was statistically significant between the groups only at 3 months ( $P=0.02$ ). The clearest advantage of antibiotics over placebo was noted in pockets initially >6 mm, but such sites were available for evaluation for up to 6 months in only 30 subjects. Subjects receiving the antibiotics showed a decrease in mean PPD in such sites from $7.3 \pm 0.3$ mm at baseline to $3.6 \pm 0.8$ mm at 3 months and to $3.7 \pm 0.6$ mm at 6 months, whereas patients receiving placebo showed a decrease from $7.2 \pm 0.7$ mm at baseline to $5.2 \pm 1.1$ mm at 3 months and to $4.9 \pm 1.4$ mm at 6 months. The difference between the two groups was highly significant at 3 months ( $P<0.001$ ) and 6 months ( $P=0.003$ ). The protective risk of the antibiotics for having more than one pocket deeper than 4 mm and bleeding on probing per subject after 6 months was 8.85. In summary, systemic metronidazole and amoxicillin significantly improved the 6-month clinical outcomes of full-mouth nonsurgical periodontal debridement, thus significantly reducing the need for additional therapy |

(continued)

**Table 3.9** (continued)

| Study                | No. patients | Periodontal condition  | Study period | Periodontal treatment   | Outcome   |
|----------------------|--------------|--|--------------|---|---|
| Ehmke et al. [46]    | 35           | Chronic periodontitis and subgingival A. <i>actinomycetemcomitans</i> and/or P. <i>gingivalis</i>      | 24 months    | 1. SRP+A/M (MET 250, 3x, AMO, 375, 3x, 8d) + supragingival irrigation with 0.06% CHX once daily (N=18)<br>2. SRP+oral hygiene instruction (N=17)        | At sites with an initial PPD $\geq$ 7 mm or more, after 12 months, test group showed a significantly higher ( $P<0.05$ ) proportion of sites gaining attachment ( $37.3\%\pm4.6\%$ versus $7.2\%\pm3.9\%$ , respectively) and significantly ( $P<0.05$ ) lower proportions of sites losing attachment ( $8.2\%\pm3.9\%$ versus $19.1\%\pm3.1\%$ , respectively); relative risk reduction of 62% for AL. Compared to controls, the intraoral prevalence of A. <i>actinomycetemcomitans</i> (up to 18 months) and P. <i>gingivalis</i> (up to 3 months) decreased and that of <i>Eikenella corrodens</i> (at 10 days) increased in test patients ( $P<0.05$ ). In both treatment groups, the detection frequency of <i>Tannerella forsythensis</i> decreased transiently, while an overall increase was recorded for <i>Treponema</i> spp. In summary, over the 24-month period, a single course of the administered adjunctive antimicrobial therapy led to a relative risk reduction of 62% for AL at deep sites. However, with the exception of A. <i>actinomycetemcomitans</i> , it failed to induce long-term changes in the prevalence profiles of oral colonization.   |
| Flemming et al. [57] | 38           | Untreated periodontitis harboring A. <i>actinomycetemcomitans</i> and/or P. <i>s</i> <i>gingivalis</i> | 1 year       | 1. SRP+A/M (AMO, 375, 3x, 8d and MET 250, 3x, 8d) + supragingival irrigation with 0.06% CHX once daily (N=18)<br>2. SRP+oral hygiene instruction (N=20) | In test patients, the adjunctive antimicrobial therapy resulted in a significantly lower detection frequency of A. <i>actinomycetemcomitans</i> in the oral cavity at the 9 and 12 month examinations compared to controls ( $P<0.01$ ). The strongest efficacy of the adjunctive antimicrobial therapy was found in subgingival plaque, where the detection frequency of this microorganism was significantly lower in test compared to control patients at 3, 6, 9, and 12 months ( $P<0.05$ ). However, it had limited effects in reducing the detection frequency of A. <i>actinomycetemcomitans</i> on oral mucous membranes. A significant difference between the groups was found only at the 9- and 12-month examination ( $P<0.01$ ). In addition, suppression of A. <i>actinomycetemcomitans</i> in subgingival plaque below detectable levels was associated with an increased incidence of CAL gain. The intraoral detection frequency of P. <i>gingivalis</i> was significantly reduced only 10 days following therapy ( $P<0.01$ ). At any time after therapy A. <i>actinomycetemcomitans</i> was not detected intraorally in 5 of 10 (50%) test and 1 of 13 (8%) control patients harboring this pathogen at baseline; P. <i>gingivalis</i> was not detected in only 1 of 18 (6%) test and none of the 17 control patients harboring this pathogen at baseline. Although the data indicated that the assessed antimicrobial therapy may suppress A. <i>actinomycetemcomitans</i> from the entire oral cavity below detectable levels over a minimum of 12 months, P. <i>gingivalis</i> persisted or re-occurred. |
| Guerrero et al. [65] | 41           | Generalized aggressive periodontitis   | 6 months     | 1. SRP+A/M (AMO 500, 3x, 7d and MET 500, 3x, 7d) (N=20)<br>2. SRP+placebo (N=21)  | In deep pockets ( $\geq$ 7 mm), the test treatment resulted in an additional 1.4 mm (95% CI: 0.8–2.0 mm) in full-mouth PPD reduction and 1 mm (0.7, 1.3 mm) of life cumulative CAL gain at 6 months. In moderate pockets (4–6 mm), the adjunctive benefit was smaller in magnitude: PPD reduction was 0.4 mm (0.1–0.7 mm) and CAL gain was 0.5 mm (0.2–0.8 mm). In addition, the 6-month data showed CAL gains $\geq$ 2 mm at 25% of sites in test patients compared with 6% in placebo. Similarly, PPD reductions of 2 mm or more were observed in 30% of sites in test and 21% of sites in placebo patients. Seventy-four percent of pockets with PPD $\geq$ 5 mm at baseline were 4 mm or shallower at 6 months in the test group. This compared with 54% in the placebo group ( $P<0.01$ ). Disease progression at 6 months was observed at 1.5% of sites in test and 3.3% of sites in placebo, respectively ( $P=0.072$ ). In summary, these data indicate that a 7-day adjunctive course of systemic metronidazole and amoxicillin significantly improved the short-term clinical outcomes of full-mouth nonsurgical periodontal debridement in subjects with GAP.  |

|                      |    |   |   |  |   |
|----------------------|----|---|---|--|---|
| Haffajee et al. [73] | 14 | Refractory periodontitis  | 2 years   | SRP + locally delivered TET at pockets $\geq 4$ mm + systemically administered A/M (AMO 500, 3x, 14d, + MET 250, 3x, 14d)            | Major reductions in plaque accumulation, gingival redness, BOP, and suppuration occurred by the 3-month monitoring visit and were sustained to 24 months. The majority of the reduction in mean PPD and CAL had occurred by 6 months post therapy and the mean improvements continued to the 24-month monitoring visit. Significant reductions in mean PPD and BOP could be observed at sites subset into initial PPD categories of <4, 4–6, or $>6$ mm. Improvement in CAL level occurred for most subjects by 6–9 months and continued up to 24 months. Mean ( $\pm$ SEM) full-mouth PPD reduction was $0.83 \pm 0.13$ mm and mean CAL “gain” was $0.44 \pm 0.12$ at 24 months. Clinical improvement was accompanied by major reductions in multiple subgingival species during the first 3 months of active therapy that were maintained for most species to the last monitoring visit. Reductions occurred for three <i>Actinomyces</i> spp., “orange-complex” species including <i>Campylobacter showae</i> , <i>Eubacterium nodatum</i> , three <i>Fusobacterium nucleatum</i> subspecies, <i>Peptostreptococcus micros</i> , <i>Prevotella intermedia</i> , as well as the “ <i>Streptococcus milleri</i> ” group, <i>Streptococcus anginosus</i> , <i>Streptococcus constellatus</i> , and <i>Streptococcus intermedius</i> . Subjects differed in their response to therapy. The change in mean ( $\pm$ SEM) full-mouth attachment level for the modest responders from baseline to 24 months was $0.0 \pm 0.04$ mm (range: “gain” of $0.13$ to a loss of $0.08$ mm). The corresponding values for the good responders were a mean “gain” of $0.77 \pm 0.11$ mm (range: $0.39$ – $1.27$ mm). In summary, the combined antibacterial therapy was successful in controlling disease progression in 14 “refractory” periodontitis subjects for 2 years. |
| Kaner et al. [86]    | 36 | Generalized aggressive periodontitis  | 6 months  | 1. SRP+A/M (AMO 500, 3x, 10d, and MET 250, 3x, 10d) (N=18)<br>2. SRP+CHX chip into every site with a baseline PPD $\geq 5$ mm (N=18) | After 3 months, change of full-mouth PPD did not differ significantly between the two groups ( $P>0.05$ ). However, at that time change of full-mouth CAL was significantly higher in the A/M group (difference between groups: $0.23$ mm; $P<0.05$ ). Between 3 and 6 months, full-mouth PPD significantly increased again in the CHX group ( $0.10$ mm, inter-quartile ranges (IQR) $0.0$ , $0.12$ ; $P=0.013$ ). A concomitant significant decrease of full-mouth PPD was observed in A/M patients ( $-0.06$ mm; IQR $-0.20$ , $-0.01$ ; $P=0.021$ ), finally yielding in $0.66$ mm greater reduction of full-mouth PPD in the A/M group at the end of the observation period ( $P<0.001$ ). Regarding full-mouth CAL, no further significant changes were observed within the treatment groups between months 3 and 6. Six months after SRP and medication, the CHX group presented $0.34$ mm less “gain” of full mouth CAL than the A/M group ( $P<0.01$ ). After completion of the hygiene phase, both groups presented similarly low plaque levels (APL) at baseline. CHX group patients maintained their oral hygiene performance without significant changes throughout the observation period. An additional significant improvement of APL was found in the A/M group only. After 6 months, the A/M group showed a significantly decreased APL level compared with the CHX group ( $P=0.002$ ). Reduction of BOP frequency was similar in both groups. In summary, in first-line nonsurgical therapy for GAP, SRP, plus adjunctive systemic amoxicillin/metronidazole was more efficacious in clinically relevant measures of outcome than SRP plus adjunctive placement of CHX chips.   |
| Loos et al. [103]    | 25 | Treated periodontitis with nonadjacent, proximal intrasseous defects with a PPD from the buccal and/or lingual aspect $\geq 6$ mm | Treated periodontitis with nonadjacent, proximal intrasseous defects with a PPD from the buccal and/or lingual aspect $\geq 6$ mm | 1. MEM+A/M (AMO 375, 3x, 8d, and MET 250, 3x, 8d) (N=13)<br>2. MEM (N=12)  | At 12 months, adjusted means Reduction for PPD ranged from $4.16$ to $5.14$ mm and reductions in PPD from baseline ranged between $2.54$ and $3.06$ mm. The reductions in PPD at 12 months for all four treatment modalities were highly significant. However, no differential effects of the treatments on postoperative change in PPD were observed and no interaction between AB and MEM was seen. At 6 and 12 months postoperatively, for all treatment combinations, adjusted means for gain in CAL ranged between $0.56$ and $1.96$ mm; the gains in CAL were judged to be significant, as the ratios of these values and the corresponding SE were generally $>2$ . Again, overall, no main effects of MEM or AB were found for probing bone level. Explorative statistical analyses indicated that smoking and not MEM or AB is a determining factor for gain in PBL ( $P=0.0009$ ). In summary, neither the application of barrier membranes nor the use of systemic antibiotics showed an additional effect over control on both soft and hard tissue measurements in the treatment of intrasseous defects.   |

(continued)

**Table 3.9** (continued)

| Study                  | No. patients | Periodontal condition  | Study period | Periodontal treatment   | Outcome  |
|------------------------|--------------|--|--------------|---|--|
| López et al. [104]     | 39           | Moderate to advanced adult periodontitis that showed ≥2 mm CAL loss in at least two sites in the previous 2 months | 12 months    | 1. A/M (AMO 500, 3x, 7d) and MET 250, 3x, 7d (N=20)<br>2. Placebo (3x, 7d)<br>Repeated at 4 and 8 months                | After 2 months and thereafter, the A/M group showed significant clinical improvement while the placebo group showed a progressive deterioration of periodontal status. At 12 months compared to baseline, subjects of the A/M group showed: (1) a significant overall mean attachment gain of 0.43 mm ( $P=0.005$ ); (2) a significant decrease of active sites ( $P\leq 0.03$ ); (3) a significant increase of sites gaining attachment level ( $P<0.01$ ); (4) a significant reduction of periodontal probing depth ( $P<0.00006$ ); and (5) a significant decrease in percentage BOP sites ( $P\leq 0.0005$ ). Significant differences between both groups at all 2-month evaluations were found in overall mean attachment level ( $P\leq 0.00004$ ), in percent of active sites ( $P\leq 0.03$ ), and in percent of BOP sites ( $P\leq 0.02$ ). Sites exhibiting ≥2 mm of CAL loss in two successive or alternate evaluations, and periodontal abscess were noticed only in the placebo group. In summary, a 1-week course of systemic A/M every 4 months, as the only therapy, arrests the progression of adult periodontitis and significantly improves the clinical parameters of the disease.   |
| López et al. [105]     | 22           | Chronic periodontitis  | 12 months    | 1. OHI+supragingival scaling+A/M (AMO 500, 3x, 7d and MET250, 3x, 7d) (N=11)<br>2. OHI+SRP+2 placebos (N=11)            | Mean PPD was reduced from 2.80±0.45 at baseline to 1.95±0.05 at 12 months ( $P<0.001$ ) and from 2.39±0.41 to 1.95±0.10 ( $P<0.001$ ) in the A/M and SRP-treated patients, respectively. Corresponding values for relative mean CAL were 10.07 ± 1.30–9.77 ± 0.34 ( $P<0.001$ ) and 9.94 ± 0.28–9.77 ± 0.26 ( $P<0.001$ ). Percentage of sites exhibiting BOP were 40.6 ± 18.3–14.0 ± 1.4 ( $P<0.001$ ) and 38.5 ± 5.1–19.0 ± 2.8 ( $P<0.001$ ) in the A/M and SRP groups, respectively. Mean total DNA probe counts and counts of the majority of the 40 test species were significantly reduced over time in both groups, with no significant differences detected at any time point between groups. At 12 months many of the species were still present at significantly lowered levels compared with their baseline counts in both groups. In summary, changes in clinical and microbiological parameters were similar after receiving systemically administered MIA as the sole therapy or after receiving SRP only   |
| Matarazzo et al. [116] | 43           | Chronic periodontitis in smokers   | 3 months     | 1. SRP+A/M (AMO 500, 3x, 14d and MET, 400, 3x, 14d) (N=14)<br>2. SRP+MET (400, 3x, 14d) (N=14)<br>3. SRP+placebo (N=15) | Overall, subjects who received antibiotics as part of the periodontal treatment had the best clinical results, especially those from the A/M group. When the full-mouth mean values were considered, subjects receiving this combination of therapies showed the greatest reduction in mean PPD and CAL compared with the other two groups (PPD reduction (mm): controls: from 3.9±0.6 to 3.3±0.5, MET: from 3.7±0.9 to 2.9±0.6, A/M: 4.0±0.7 to 3.0±0.8; CAL (mm): controls: from 4.7±1.2 to 4.2±1.1, MET: from 4.5±0.9 to 3.9±0.9, A/M: from 4.8±0.8 to 3.9±0.8) ( $P<0.01$ ). Both antibiotic therapies were more effective in reducing the mean percentage of sites with BOP than SRP alone (MET from 65.5±25.8 to 55.1±20.2, A/M from 75.8±22.9 to 56.3±16.5) ( $P<0.01$ ). The initially shallow sites of the A/M group showed the greatest reduction in mean PPD, followed by MET and C groups ( $P<0.01$ ). The antibiotic therapies also led to more effective reductions in mean PPD and CAL in initially intermediate and deep sites compared with SRP alone ( $P<0.01$ ). The difference observed between the two test groups was a greater reduction in mean CAL in subjects receiving A/M in initially intermediate sites ( $P<0.01$ ). Therapies with antibiotics were also more effective than SRP in reducing BOP in initially intermediate sites ( $P<0.01$ ). The SRP+A/M therapy led to the most beneficial changes in the subgingival microbial profile. These subjects showed significant reductions in the mean counts and proportions of periodontal pathogens such as <i>Tannerella forsythia</i> , <i>Porphyromonas gingivalis</i> , and <i>Treponema denticola</i> , and the greatest increase in proportions of host-compatible species. In summary, significant advantages are observed when systemic antibiotics are combined with SRP in the treatment of smokers with chronic periodontitis. The greatest benefits in clinical and microbiological parameters are achieved with the use of SRP+A/M |

|                           |    |  |           |  |   |
|---------------------------|----|--|-----------|--|---|
| Moeintaghavi et al. [122] | 50 | Moderate to severe chronic periodontitis | 2 months  | 1. SRP+A/M (AMO 500, 3x, 7d and MET 250, 3x, 7d) (N = 28)<br>2. SRP+placebo (N=22)   | Reduction of the PPD in the placebo and A/M groups were $1.5 \pm 0.45$ and $2.1 \pm 0.67$ , respectively. The difference between the two groups was significant ( $P=0.00001$ ). Covariance analysis using baseline PPD as a covariate showed the final significant difference between the two groups was independent of the initial periodontal probing depth. Clinical attachment gain was significant in both placebo and A/M groups ( $P = 0.00001$ ). It was $1.15$ mm in the placebo group versus $1.92$ mm in the A/M group. The PI was reduced significantly compared to the baseline in both groups ( $P = 0.00001$ ) and between the two groups ( $P = 0.05$ ). Similar to PI, the differences in BI within groups between baseline and re-examination were significant ( $P = 0.00001$ ). The BI decreased in the A/M group more significantly than in the placebo group ( $P < 0.05$ ). Parallel with the clinical changes, the treatment of the A/M group reduced the number of <i>Aa</i> , <i>Pg</i> , and <i>Pi</i> significantly compared to baseline ( $P=0.003$ , $0.02$ , and $0.001$ , respectively). In the placebo group only the <i>Pi</i> colony count was reduced significantly ( $P = 0.001$ ) and showed a significant difference between the two groups at the end of the study ( $P = 0.026$ ). The number of patients positive for <i>Pg</i> , <i>Aa</i> , and <i>Pi</i> in the A/M group decreased significantly after therapy, but in the placebo group only patients positive for <i>Aa</i> decreased significantly. In summary, the significant differences between treatment and placebo groups are in line with other studies and support the considerable adjunctive benefits of the combination of amoxicillin and metronidazole in the treatment of chronic periodontitis. |
| Mombelli et al. [125]     | 16 | Chronic periodontitis                    | 12 months | 1. SRP+EMD+A/M (AMO 375, 3x, 7d and MET 250, 3x, 7d) (N=8)<br>2. SRP+EMD+placebo (N=8)   | The overall PPD reduction from baseline to 6 and 12 months was highly significant ( $P < 0.001$ ). The mean PPD was $4.9 \pm 1.6$ mm after 6 months and $5.1 \pm 1.6$ mm after 1 year. A significant overall gain in clinical attachment was noted over 6 months ( $1.5 \pm 1.9$ , $P < 0.001$ ) and 12 months ( $1.1 \pm 2.3$ mm, $P=0.02$ ). Subtraction radiography confirmed an increase in bone density in 28% sites at month 6, and in 38% sites at month 12. None of the sites showed a decrease in bone density. Subjects treated with A/M yielded significantly better clinical results than those treated with placebo. After 6 months PPD was reduced significantly more ( $3.0 \pm 2.1$ versus $1.6 \pm 1.4$ mm, $P < 0.05$ ), and the mean PPD was significantly smaller. Mean CAL gain was significantly greater after 6 months ( $2.3 \pm 1.9$ mm versus $0.7 \pm 1.6$ mm, $P = 0.02$ ) and 12 months ( $2.3 \pm 3.5$ mm versus $0.4 \pm 3.8$ mm, $P = 0.02$ ). On the other hand, although <i>P. gingivalis</i> was reduced considerably, this microorganism was not completely eradicated, even in the subjects treated with antibiotics. In summary, the present study supports the notion that optimal repair and regeneration of the periodontium requires suppression of the microbiota causing periodontal disease.   |
| Ribeiro Edel et al. [165] | 25 | Severe chronic periodontitis             | 6 months  | 1. Full-mouth ultrasonic debridement+A/M (AMO, 375, 3x, 7d and MET, 250, 3x, 7d) (N=13)<br>2. Full-mouth ultrasonic debridement+placebo (N=12) | At 6 months, the test treatment resulted in lower BOP (7.75% and 21.11% of the sites in the test and control groups, $P < 0.05$ ), and an additional reduction (0.83 mm) in PPD ( $3.28 \pm 0.41$ vs $2.45 \pm 0.50$ mm, $P < 0.05$ ). The mean CAL gain was $1.68 \pm 0.95$ mm in the control group and $1.88 \pm 0.89$ mm in the test group ( $P > 0.05$ ). A difference between the groups was observed with regard to the proportions of sites presenting CAL gain $\geq 2$ mm at 6 months (58.03% of sites in test vs 43.52% of sites in control patients, $P < 0.05$ ). In addition, a difference between the groups was found with regard to the proportion of sites with PPD $\geq 5$ mm at all periods of evaluation. All qualifying sites (100%) presented PPD $\geq 5$ mm at baseline. At 3 months, 21.30% of the sites in the control group still had PPD $\geq 5$ mm, whereas only 8.93% of sites in the test group did ( $P < 0.05$ ). Real-time PCR and ELISA failed to identify significant differences between the groups. In summary, both treatments resulted in significant clinical improvements; no improvement in the microbiologic or immunologic outcome was observed with the adjunctive use of systemic amoxicillin and metronidazole.   |

(continued)

**Table 3.9** (continued)

| Study                | No. patients | Periodontal condition                | Study period | Periodontal treatment   | Outcome   |
|----------------------|--------------|--------------------------------------|--------------|---|---|
| Rooney et al. [171]  | 62           | Advanced chronic periodontal disease | 6 months     | 1. AM: SRP + A/M (AMO 250, 3x, 7d and MET 200, 3x, 7d) (N=15)<br>2. PM: lactate capsules and MET (MET 200, 3x, 7d) (N=16)<br>3. AP: amoxicillin and calcium lactate (N=16)<br>4. PP: lactate and calcium lactate (N=15) | PPD improved in all groups. Treatment effects were highly significantly different and always greatest in the AM and least in the PP groups. Benefits of PM and AP over PP were also noted. The mean percentage of sites with high (>6 mm) PPD reduction in the four treatment groups at 6 months compared with baseline were: AM: 1.3% from 15.9%, PM: 4.8% from 15.6%, AP: 3.8% from 14.6%, PP: 12.4% from 19.3%. CAL improved in all groups and showed the same highly significant treatment differences, again favoring AM. The mean percentage of sites with high (>6 mm) AL reduction in the four treatment groups at 6 months compared with baseline: AM 6.5% from 17.1%, PM: 8.8% from 18.2%, AP 10.0% from 18.7% and PP 18.2% from 24.3%. BOP improved in all groups, particularly in AM compared to the other groups: AM 22.8% from 62.6%, PM 32.5% from 61.8%, AP 33.9% from 61.8%, PP 44.9% from 65.6%. Regarding total anaerobic and aerobic counts, the only significant difference between treatments was at 1 month where the combined treatment was significantly more effective against total anaerobic counts than the double placebo and metronidazole and placebo. <i>P. intermedia</i> counts were always lower in active groups compared to PP and reached significance for AM and AP at 1 month and AM and PM at 3 months. In summary, the significant differences among treatment groups and the overall trend in the data, in line with other studies, support the considerable adjunctive benefits to SRP of amoxicillin and metronidazole combined in the treatment of advanced chronic periodontal disease. |
| Sculian et al. [180] | 34           | Deep intrabony defects               | 1 year       | 1. EMD+ A/M (AMO 375, 3x, 7d and MET 250, 3x, 7d) (N=17)<br>2. EMD alone (N=17)   | The results have shown that in the EMD+A/M group the PPD decreased from $9.1 \pm 1.5$ to $4.5 \pm 1.1$ mm ( $P < 0.0001$ ) and the CAL changed from $11.0 \pm 1.6$ to $7.5 \pm 1.4$ mm ( $P < 0.0001$ ). In the EMD group the PPD decreased from $9.0 \pm 1.7$ to $4.3 \pm 1.7$ mm ( $P < 0.0001$ ) and the CAL changed from $10.6 \pm 1.6$ to $7.3 \pm 1.5$ mm ( $P < 0.0001$ ). There were no significant differences in any of the investigated parameters between the two groups. In summary, it can be concluded that the systemic administration of amoxicillin and metronidazole adjacent to the use of EMD for the surgical treatment of intrabony periodontal defects does not produce statistically superior PPD reduction and CAL gain when compared to treatment with EMD alone. Hence, the present results do not support the routine administration of amoxicillin and metronidazole following regenerative treatment with EMD.   |
| Winkel et al. [225]  | 49           | Adult periodontitis                  | 3 months     | 1. Supra- and subgingival debridement+A/M (AMO 375, 3x, 7d and MET 250, 3x, 7d) (N=23)<br>2. Supra- and subgingival debridement+placebo (N=26)  | Significant differences were observed between the placebo and A/M groups after therapy regarding change in the BI ( $0.4 \pm 0.2$ , $P < 0.05$ ), PPD ( $3.4 \pm 3.0$ , $P < 0.05$ ), and CAL ( $3.6 \pm 3.2$ , $P < 0.05$ ). The greatest reduction in PPD was found at sites with initial PPD $\geq 7$ mm, 2.5 mm in the placebo group and 3.2 mm in the A/M group. The improvement in CAL was most pronounced in the PPD category $\geq 7$ mm and amounted to 1.5 and 2.0 mm in the placebo and A/M groups, respectively. In summary, this study has shown that systemic usage of metronidazole and amoxicillin, when used in conjunction with initial periodontal treatment in adult periodontitis patients, achieves significantly better clinical and microbiological results than initial periodontal treatment alone. Moreover, this research suggests that especially patients diagnosed with <i>P. gingivalis</i> benefit from antibiotic treatment.  |

|                           |    |                                      |          |   |  |
|---------------------------|----|--------------------------------------|----------|---|--|
| Xajigeorgiou et al. [226] | 43 | Generalized aggressive periodontitis | 6 months | 1. SRP+debridement (ultrasonics and polishing with a rubber cup)+A/M (AMO 500, 3x, 7d+MET 500, 3x, 7d) (N=10)                                     | No differences were observed between the four groups at any time point regarding PPD, CAL, and BOP reduction. The reduction in mean PPD (mm) in subjects who received A/M combination was statistically significant (baseline: $4.63 \pm 0.97$ ; 6 weeks after SRP: $3.44 \pm 0.48$ ; 6 months: $3.12 \pm 0.71$ , $P < 0.05$ ). The CAL change (mm) in the A/M group: baseline: $4.97 \pm 1.01$ , 6 weeks after SRP: $4.31 \pm 0.92$ , 6 months: $4.05 \pm 1.34$ . The BOP of the A/M group during the experimental period was baseline: $0.87 \pm 0.21$ , 6 weeks after SRP: $0.22 \pm 0.18$ , 6 months: $0.15 \pm 0.14$ . The intake of A/M and MET, resulted in significant reduction of the percentage of sites $\geq 6$ mm compared with the control group at 6 months: (80% reduction for A/M, 87.7% for MET, 57.7% for controls). Adjunctive A/M or MET alone (when <i>A.actinomycetemcomitans</i> is not involved) was effective in deep pockets of aggressive periodontitis patients. In summary, adjunctive metronidazole plus amoxicillin or metronidazole alone (when <i>A.actinomycetemcomitans</i> is not involved) is effective in deep pockets of aggressive periodontitis patients. |
|                           |    |                                      |          | 2. SRP+debridement (ultrasonics and polishing with a rubber cup) + + DOXY (200 mg of doxycycline as a loading dose and 100 mg/day 14 days) (N=10) |  |
|                           |    |                                      |          | 3. SRP+debridement (ultrasonics and polishing with a rubber cup) + + MET (500, 3x, 7d) (N=12)   |  |
|                           |    |                                      |          | 4. Only SRP + debridement (ultrasonics and polishing with a rubber cup) (N=11)  |  |

PC professional cleaning, MEM guided tissue regeneration procedure with a bioresorbable membrane, PBL probing bone level, PPD periodontal probing depth, CAL clinical attachment levels, PI plaque index, BI bleeding index, EMD enamel matrix derivative

organism was strongly related, at baseline, to PPD $\geq$ 7 mm (odds ratio (OR)=9.8,  $P<0.001$ ). Six weeks after scaling, the organism was associated with CAL $\geq$ 6 mm (OR=1.8,  $P=0.02$ ). After scaling, high counts of *A. actinomycetemcomitans* in excess of 10(4) CFU/mL significantly interfered with attachment gain of  $\geq$ 2 mm (OR=0.24,  $P=0.001$ ) [130].

The effect of a combined systemic ciprofloxacin-metronidazole therapy (500 mg of each for 8 days) was examined on 17 adults with recurrent periodontitis despite prior mechanical/surgical therapy, plaque control, and systemic maintenance care [161]. Clinical and microbiological parameters were evaluated pre-therapy and from 6 to 18 months post therapy at 4–8 severe sites/patient. Ciprofloxacin-metronidazole therapy eliminated or significantly suppressed evaluated subgingival putative periodontal pathogens. *Streptococci* and occasionally *Actinomyces* spp. were the predominant cultivable subgingival microorganisms up to 6–18 months post treatment. Significant improvements in PPD, CAL, and BOP paralleled elimination or suppression of suspected periodontal pathogens in all patients were reported, with no additional periodontitis disease activity detected at any site post treatment [161].

Another antibiotic combination proposed for periodontal disease therapy was the association of **spiramycin and metronidazole** [150]. However, Isla et al. [82] reported that the spiramycin plus metronidazole combination, present in the commercial formulation Rhodogyl, did not reach satisfactory pharmacokinetic/pharmacodynamic indexes of antimicrobial treatment efficacy for orofacial infections.

Spiramycin as well as metronidazole showed good antimicrobial activity against species of *Prevotella*, *Eubacterium*, *Peptostreptococcus*, *Bacteroides*, and *Porphyromonas*, as well as the anaerobic spirochetes [157, 169]. However, 90% of the *A. actinomycetemcomitans* strains are slightly resistant to spiramycin and 72% to metronidazole [107].

In a clinical study, the commercial preparation of these two antibiotics (Rodogyl) was used adjunctively to SRP in the treatment of advanced periodontal disease. At 6-month evaluation, the Rodogyl group exhibited a greater gain in CAL (0.67 mm) and a significantly greater decline in the proportion of spirochetes compared with the placebo group [156].

### 3.6.3 Sequencing of Antibiotic Therapy

**The sequential use of drugs overcomes the potential risk of antagonism between bacteriostatic and bactericidal antibiotics.** To date, serial drug regimens studied in periodontics include systemic doxycycline administered initially, followed by either Augmentin or metronidazole [207].

Aitken et al. [3] has examined the efficacy of metronidazole and tetracycline in preventing recurrent periodontitis in 23 patients. After an initial subgingival SRP and prophylaxis, baseline measurements of plaque index (PI), gingival index (GI), crevicular fluid flow, and periodontal AL were made. Every 2 months thereafter all measurements were repeated and the teeth were again scaled subgingivally, root planed, and polished. Upon detection of periodontal AL loss exceeding 2 mm or the occurrence of a periodontal abscess at one or more sites at any examination, patients entered into the treatment phase. **Doxycycline (200 mg to start and 100 mg every 24 h for 3 weeks)** or a lactose-containing placebo of identical appearance was then administrated according to a random allocation (RA) method. Patients were recalled at 1, 3, and 7 months after the last day of doxycycline or placebo treatment for clinical measurements, subgingival SRP, and prophylaxis. Patients who continued to exhibit active destruction at these appointments were entered into a prospective study in which they were treated with **metronidazole (250 mg every 8 h for 10 days)**. After administration of metronidazole, 5 out of 12 (42%) of the placebo plus metronidazole group exhibited recurrence of disease within 3 months while only 1 out of 11 (9%) of the doxycycline plus metronidazole group exhibited recurrence ( $P<0.096$ ).

The systemic use of a combination antibiotic regimen: **doxycycline, 200 mg the first day and 100 mg for 4 days thereafter, and then Augmentine, 500 mg three times daily for 5 days**, in conjunction with root planning as treatment of *A. actinomycetemcomitans* and/or *P. gingivalis*-associated periodontitis has demonstrated sustained reduction in PPD (2.2 mm) and gain in periodontal AL (0.8 mm) at 12 weeks. The authors also revealed that the clinical results occur without any prolonged alteration in the test bacteria [117, 118].

## 3.7 Adverse Effects

Before writing a prescription, the clinician must estimate the likelihood that a given patient will experience a **serious adverse effect** with a specific drug [192].

The possibility of unique age-related adverse effects is an important consideration in prescribing antibiotics. Prescribing antibiotics to pregnant women is a cause for particular concern. Table 3.10 lists age-related safety issues of antibiotics commonly used in periodontal therapy.

Adverse drug reactions related to **amoxicillin alone and in association with clavulanic acid** include gastrointestinal tolerability (nausea, vomiting, cramping, diarrhoea), allergic reactions (Stevens–Johnson syndrome), hematological reactions (purpura, thrombocytopenia, granulocytopenia, and leucopenia), and liver tolerability (cholestatic hepatitis, hepatitis, hepatic necrosis, and hepatocellular damage) [175, 176].

The most common adverse reactions associated with **metronidazole** involve the gastrointestinal tract. About 12% of the patients experience nausea, which may be accompanied by headache, anorexia, and vomiting. Drowsiness, depression, skin rashes, and vaginal and/or urethral burning have been reported. Metronidazole affects the activity of hepatic enzymes involved with the metabolism of ethanol, producing unpleasant symptoms due to the accumulation of acetaldehyde in the blood. Alcohol ingestion is strictly contraindicated in patients receiving metronidazole. Metronidazole crosses the placenta barrier, entering the fetal circulation system. It is also secreted in breast milk. Because of the association of metronidazole with tumorigenicity in some animals, the drug is contraindicated in pregnant women or nursing mothers [219].

**Tetracyclines**, as an antibiotic class, are known to have side effects such as gastrointestinal symptoms, pediatric tooth discoloration, candidiasis, photosensitivity reactions, pigmentation changes, and central nervous system (CNS) effects (e.g., lightheadedness, dizziness) [192]. Smith and Leyden [192] summarized the available literature covering the safety profiles of oral doxycycline and minocycline reported in clinical trials of doxycycline from 1966 through August 2003 ( $N=3,833$  patients). The reported adverse effects for **doxycycline** were: body as a whole (headache, somnolence, tooth discoloration), digestive system (gastrointestinal not

otherwise specified (NOS), heartburn/gastritis, nausea/vomiting), skin and appendages (allergic skin symptoms, skin reactions, pimples/acne, photosensitivity, rash, pruritus), urogenital system (urogenital NOS, vaginitis, yeast infection, vaginal dryness), nervous system (CNS-related, dizziness), neuropsychiatric immune system (hypersensitivity), respiratory system (rhinitis), and special senses (taste perversion). The review of adverse events reported in clinical trials of **minocycline** from 1966 through August 2003 ( $N=788$  patients) revealed the most common ones as: body as a whole (headache, somnolence), digestive system (gastrointestinal NOS, nausea/vomiting, diarrhea, epigastric/abdominal pain, flatulence), skin and appendages, allergic skin symptoms, pimples/acne, rash, pruritus, fixed drug eruption), urogenital system (heavy period), nervous system (CNS NOS, dizziness/vertigo, lightheadedness, lack of concentration, disassociation, loss of balance, vestibular NOS, tinnitus), other (other NOS, euphoria, weakness/fatigue), oral disturbances (dryness, altered taste, halitosis, thrush), visual problem, chest pain, increased appetite, and frequency of adverse events [192].

Substantial evidence indicates that the adjunctive use of **SDD** consisting of 20 mg doxycycline hydiate (Periostat, CollaGenex Pharmaceuticals Inc., Newtown, PA, USA), twice per day, provides a significant benefit to SRP in the treatment of periodontitis because of the anticollagenase and anti-inflammatory activities of doxycycline. However, serious concern has been expressed that even sub-antimicrobial levels of doxycycline may exert a detrimental antimicrobial effect on the normal flora. Such an effect could result in the disruption or suppression of the normal flora and lead to its colonization or overgrowth by opportunistic pathogens as well as the development of nonsusceptible microorganisms [199, 200, 215, 220]. Several studies have documented the safety of SDD on the subgingival, large intestine, or vaginal flora and have clearly shown that SDD exerts no detectable effect on the normal oral bacteria. There is also no indication that treatment with SDD tended to promote a greater likelihood of the development of cross- or multi-antibiotic resistance with any of the several antibiotics tested (minocycline, tetracycline, amoxicillin, erythromycin, clindamycin) [215].

In two systematic reviews on the effects of systemic antimicrobial therapy in periodontitis, Herrera et al. [78, 80] observed that only few studies reported data

**Table 3.10** Considerations on dosage and adverse effects of antibiotic regimes used in periodontal therapy

| Category             | Agent                                   | Regimen (mg)  | Dose                         | Time of exposure              | Adverse drug reactions in the test group | Reference |
|----------------------|---|---|------------------------------|-------------------------------|--|-----------|
| <b>Monotherapies</b> |   |   |                              |                               |  |           |
| Penicillin           | Amoxicillin                             | 250   | 3 times per day              | 7 days                        | No                                       | [171]     |
|                      | Augmentin (amoxicillin/clavulanic acid) | 250/125   | 3 times per day              | 30 days                       | NA                                       | [69]      |
|                      | 250/125                                 | 3 times per day   | 14 days                      | NA                            | [217]                                    |           |
|                      | 250/125                                 | 3 times per day   | 14 days                      | NA                            | [108]                                    |           |
|                      | 250/125                                 | 3 times per day   | 10 days                      | NA                            | [43]                                     |           |
|                      | 250/125                                 | 3 times per day   | 10 days                      | NA                            | [42]                                     |           |
|                      | 500/125                                 | 3 times per day   | 10 days                      | Mild diarrhea 20%             | [224]                                    |           |
| <b>Tetracyclines</b> |   |   |                              |                               |  |           |
|                      | Tetracycline-HCl                        | 250   | 4 times per day/once per day | 14 days/48 weeks              | NA                                       | [96]      |
|                      | 250                                     | 4 times per day   | 12 days                      | NA                            | [179]                                    |           |
|                      | 250                                     | 4 times per day   | 14 days                      | Diarrhea 3.7%                 | [5]                                      |           |
|                      | 250                                     | 4 times per day   | 14 days                      | NA                            | [147]                                    |           |
|                      | 250                                     | 4 times per day   | 14 days                      | NA                            | [162]                                    |           |
|                      | 250                                     | 4 times per day   | 21 days                      | NA                            | [160]                                    |           |
|                      | 250                                     | 4 times per day   | 30 days                      | NA                            | [69]                                     |           |
|                      | 200                                     | Once per day  | 21 days                      | NA                            | [131]                                    |           |
|                      | 100                                     | Once a day  | 14 days                      | NA                            | [55]                                     |           |
|                      | 100                                     | Once a day  | 21 days                      | Gastrointestinal mild (10.3%) | [118]                                    |           |
|                      | 200/100                                 | 200 mg of doxycycline as a loading dose and 100 mg/day 14 days) | 1/14 days                    | NA                            | [4]                                      |           |
|                      | 200/100                                 | 200 mg of doxycycline as a loading dose and 100 mg/day 14 days) | 1/14 days                    | No                            | [177]                                    |           |
|                      | 200/100                                 | 200 mg of doxycycline as a loading dose and 100 mg/day 14 days) | 1/14 days                    | No                            | [226]                                    |           |
|                      | 200/100                                 | 200 mg of doxycycline as a loading dose and 100 mg/day 14 days) | 6 weeks                      | No                            | [135]                                    |           |
|                      | 200/100                                 | 200 mg the first day followed by 100 mg/day                     | 21 days                      | NA                            | [92]                                     |           |
|                      | 200/100                                 | 200 mg to start and 100 mg/day for 3 weeks                      | 8 days                       | [182]                         |  |           |
|                      | 200                                     | Once a day  |                              |                               |  |           |

|  |    |                 |           |   |       |
|--|----|-----------------|-----------|---|-------|
| Sub-antimicrobial dose doxycycline (SDD) | 20 | 2 times per day | 9 months  | Dizziness 1.11%   | [26]  |
|  | 20 | 2 times per day | 120 days  | NA  | [30]  |
|  | 20 | 2 times per day | 3 months  | No  | [52]  |
|  | 20 | 2 times per day | 3 months  | No  | [51]  |
|  | 20 | 2 times per day | 3 months  | No  | [54]  |
|  | 20 | 2 times per day | 3 months  | No  | [53]  |
|  | 20 | 2 times per day | 3 months  | NA  | [64]  |
|  | 20 | 2 times per day | 3 months  | No  | [66]  |
|  | 20 | 2 times per day | 3 months  | No  | [67]  |
|  | 20 | 2 times per day | 3 months  | No  | [72]  |
|  | 20 | 2 times per day | 3 months  | Dizziness/tachycardia 10%   | [94]  |
|  | 20 | 2 times per day | 3 months  | The adverse events in the SDD group were similar to those in the placebo group (headache, backache, cold, flu, periodontal abscess, nausea, sinusitis)  | [123] |
|  | 20 | 2 times per day | 3 months  | NA  | [134] |
|  | 20 | 2 times per day | 3 months  | No  | [136] |
|  | 20 | 2 times per day | 6 months  | NA  | [137] |
|  | 20 | 2 times per day | 6 months  | No  | [136] |
|  | 20 | 2 times per day | 2 years   | Similar between the two groups (11% in SDD group and 13% in controls). Fewer SDD subjects experienced a dermatologic adverse event (including rash, itchy skin, acne, rosacea, hives, and nail fungus) at some time during the clinical trial compared with placebo subjects (2 versus 17%, $P = 0.002$ )   | [149] |
|  | 20 | 2 times per day | 2 years   | Diverticulitis, appendicitis, pneumonia, kidney obstruction, recurrent breast cancer, breast cancer, colon cancer, collapsed lung resulting in hospitalization, acute pancreatitis and cholelithiasis, broken arm requiring surgery, hospitalization for hypertension, gallbladder removal, squamous cell carcinoma of skin, basal cell carcinoma of skin. Hospitalization for knee surgery, acute pancreatitis | [153] |
|  | 20 | 2 times per day | 9 months  | No  | [154] |
|  | 40 | 2 times per day | 9 months  | No  | [164] |
|  | 20 | 2 times per day | 2 years   | NA  | [202] |
|  | 20 | 2 times per day | 6 weeks   | No  | [216] |
|  | 20 | 2 times per day | 24 months | No  |       |

(continued)

**Table 3.10** (continued)

| Category              | Agent         | Regimen (mg) | Dose                   | Time of exposure | Adverse drug reactions in the test group                   | Reference |
|-----------------------|---------------|--------------|------------------------|------------------|--|-----------|
| Macrolide             | Azithromycin  | 500          | Once a day             | 3 days           | Allergic reaction 4%, difficulty swallowing the tablets 4% | [72]      |
|                       |               | 500          | Once a day             | 3 days           | No   | [41]      |
|                       |               | 500          | Once a day             | 3 days           | Mild diarrhea 5.8%   | [61]      |
|                       |               | 500          | Once a day             | 3 days           | No   | [68]      |
|                       |               | 500/250      | Twice daily/once a day | 1/4 days         | No   | [115]     |
|                       |               | 500          | Once a day             | 3 days           | No   | [193]     |
| Lincomycin derivative | Clindamycin   | 150          | 4 times per day        | 10 days          | NA   | [217]     |
|                       |               | 150          | 4 times per day        | 10 days          | NA   | [108]     |
|                       |               | 150          | 4 times per day        | 8 days           | NA   | [182]     |
| Nitroimidazole        | Metronidazole | 200          | 3 times per day        | 10 days          | NA   | [179]     |
|                       |               | 500          | 3 times per day        | 7 days           | Metallic taste 8.33%                                       | [226]     |
|                       |               | 500          | 2 times per day        | 8 days           | NA   | [182]     |
|                       |               | 200          | 3 times per day        | 7 days           | No   | [171]     |
|                       |               | 250          | 3 times per day        | 14 days          | Dizziness 4.1%, diarrhea 4.1%                              | [72]      |
|                       |               | 400          | 3 times per day        | 10 days          | No   | [24]      |
|                       |               | 400          | 3 times per day        | 10 days          | No   | [23]      |
|                       |               | 200          | 3 times per day        | 5 days           | NA   | [84]      |
|                       |               | 200          | 3 times per day        | 5 days           | NA   | [83]      |
|                       |               | 250          | 3 times per day        | 7 days           | NA   | [99]      |
|                       |               | 200          | 3 times per day        | 7 days           | NA   | [110]     |
|                       |               | 200          | 3 times per day        | 7 days           | NA   | [145]     |
|                       |               | 200          | 3 times per day        | 7 days           | NA   | [146]     |
|                       |               | 400          | 3 times per day        | 7 days           | Diarrhea, taste  | [195]     |
|                       |               | 400          | 3 times per day        | 7 days           | NA   | [194]     |
|                       |               | 400          | 3 times per day        | 14 days          | Diarrhea and vomiting (nausea) 14.3%                       | [116]     |

|                              |   |            |   |         |   |       |
|------------------------------|---|------------|---|---------|---|-------|
| <b>Combination therapies</b> | Amoxicillin/<br>metronidazole           | 500/500    | 3/3 times per day   | 7 days  | Mild gastrointestinal discomfort 20%  | [226] |
|                              |   | 250/200    | 3/3 times per day   | 7 days  | No  | [171] |
|                              |   | 375/250    | 2/3 times per day   | 14 days | NA  | [13]  |
|                              |   | 375/500    | 3/3 times per day   | 7 days  | Diarrhea 8%, nausea 8%, stomach burning 4%  | [35]  |
|                              |   | 375/500    | 3/3 times per day   | 7 days  | Stomach upset (nausea or vomiting) 20%, gastrointestinal disorder (diarrhea) 24%, headache 16%, musculoskeletal disorder (cramps) 20%, respiratory disorder (e.g., dyspnea) 4%, metallic taste 8% | [34]  |
|                              | Doxycycline<br>followed by<br>Augmentin | 375/250    | 3/3 times per day   | 8 days  | Gastrointestinal intolerance 11.11%   | [46]  |
|                              |   | 375/250    | 3/3 times per day   | 8 days  | Gastrointestinal intolerance 11.11%   | [57]  |
|                              |   | 500/500    | 3/3 times per day   | 7 days  | Stomach upset (nausea and vomiting), gastrointestinal disorder (diarrhea), headache, metallic taste, general wellness (irritability, flu, etc.) 20, 55%   | [65]  |
|                              |   | 500/250    | 3/3 times per day   | 14 days | NA  | [73]  |
|                              |   | 500/250    | 3/3 times per day   | 10 days | Diarrhea 27.8%  | [86]  |
|                              |   | 375/250    | 3/3 times per day   | 8 days  | No  | [103] |
|                              |   | 500/250    | 3/3 times per day   | 7 days  | No  | [104] |
|                              |   | 500/250    | 3/3 times per day   | 7 days  | No  | [105] |
|                              |   | 500/40     | 3/3 times per day   | 14 days | Diarrhea and vomiting (nausea) 7.14%  | [116] |
|                              |   | 500/250    | 3/3 times per day   | 7 days  | Intestinal disturbance 3.57%  | [122] |
|                              |   | 375/250    | 3/3 times per day   | 7 days  | NA  | [125] |
|                              |   | 375/250    | 3/3 times per day   | 7 days  | Nausea, diarrhea 30.77%   | [165] |
|                              |   | 250/200    | 3/3 times per day   | 7 days  | No  | [171] |
|                              |   | 375/250    | 3/3 times per day   | 7 days  | Diarrhea 29.14%   | [180] |
|                              |   | 375/250    | 3/3 times per day   | 7 days  | Constipation, softened, watery, diarrhea, rash on the face, rash on the neck  | [225] |
|                              |   | 500/500    | 3/3 times per day   | 7 days  | Mild gastrointestinal discomfort 20%  | [226] |
| <b>Sequencing</b>            | Doxycycline followed by Augmentin       | 200/500 mg | DOXY (200 mg the first day and 100 mg for 4 days)+AMOX/CLAV (500, 3x, 5d) | 5 days  | NA  | [118] |

on adverse effects. Adverse events were more frequent in test than in control groups. This was especially evident when two antibiotics were combined, mainly for the combination of amoxicillin and metronidazole. However, most adverse effects were related to gastrointestinal problems and were considered minor by the patients. Few adverse effects led to dropout from the studies. No proper evaluation of adverse microbiological effects was reported [78].

As Herrera et al. [80] revealed, only very few studies assessed the safety and tolerance of the drugs [5, 10, 26, 29] as well as the increases in bacterial resistance in the subgingival microbiota [29].

## 3.8 Discussions of Available Data Regarding Effectiveness of Systemic Antibiotics in Periodontal Therapy

### 3.8.1 Plaque Index Change

Plaque was assessed either as the mean PI (Silness and Löe) or as the mean percentage of sites with plaque [80]. Systemic antibiotic therapy does not significantly affect supragingival plaque accumulation. Reduction in dental plaque depends mostly on patients' oral hygiene efforts [188]. When considering the percentage of sites with plaque, final results were always lower or equal to 30%, with no great differences in the final level of plaque or in the improvement between groups [80, 188, 211].

### 3.8.2 Gingival Inflammation

When the change in the mean percentage of sites with BOP was evaluated, overall, SRP and SRP+AB obtained similar results, with a high variability among studies, but differences between groups ranged around 15%, in favor of either control or test groups [80, 188, 211].

However, in smokers with CP, antibiotic therapies (with MTZ [400 mg, 3× per day, 14 days] or with A/M [500 mg, 3× per day, 14 days]) were more

effective in reducing the mean percentage of sites with BOP than SRP alone: MET from  $65.5 \pm 25.8$  to  $55.1 \pm 20.2$  and A/M from  $75.8 \pm 22.9$  to  $56.3 \pm 16.5$  ( $P < 0.01$ ) [116].

Similar results were reported by Rooney et al. [171]. In advanced chronic periodontal disease subjects who received quadrant SRP and then were prescribed amoxycillin capsules (250 mg) and metronidazole tablets (200 mg) (A/M), lactate capsules and metronidazole (PM), amoxycillin and calcium lactate tablets (AP), or lactate and calcium lactate (PP), the pattern for all groups was for a reduction of BOP over time, being most marked in the A/M group. Overall differences among treatments were significant at 1, 3, and 6 months ( $P < 0.01$  to  $P < 0.001$ ). Two-way analysis of variance revealed highly significant differences in favor of A/M compared to PP ( $P < 0.001$ ). However, differences between AP and MP compared to PP were mainly nonsignificant. A/M was also significantly more effective for reductions in BOP than in AP at all the points and significantly more than PM at 1 and 3 months [171].

A statistically significant difference between CP patients taking A/M or placebo after full-mouth ultrasonic debridement was found at 6 months after therapy. At this time, 7.75% and 21.11% of the sites in the test and control groups, respectively, exhibited BOP [165].

In adult periodontitis patients there were reductions in BOP for shallow, moderate, and deep pockets with the azithromycin (500 mg, once daily for 3 days a week) and placebo groups both showing improvements until week 10 but thereafter deteriorating slightly between weeks 10 and 22 with the exception of the deep pockets in the azithromycin group. Fewer pockets of initial depth 4 mm or more continued to bleed on probing in the test group (46.9%) when compared with the control group (55.6%) [193].

In CP cases, azithromycin administration in conjunction with full-mouth SRP (SRP+AZ, 500, 1×, 3 days) significantly reduced BOP rate compared with control groups (SRP only) at 13 and 25 weeks from baseline. In the test group, the BOP rate at baseline was  $31.42 \pm 17.15\%$ , and at 5, 13, and 25 weeks after full-mouth SRP, these values were  $5.08 \pm 2.94\%$ ,  $4.46 \pm 3.27\%$ , and  $5.35 \pm 2.91\%$ , respectively. In the control group, the BOP rate at baseline was  $31.43 \pm 18.46\%$ , and at 13 and 25 weeks from baseline, it was  $12.08 \pm 7.15\%$  and  $12.91 \pm 7.62\%$ , respectively [61].

In postmenopausal, osteopenic, estrogen-deficient women on periodontal maintenance, the odds of BOP were 18% lower for subjects receiving SDD (20 mg, 2x, 2 years) relative to placebo, which was not statistically significant ( $OR=0.82\%$ , 95% CI: 0.61–1.10,  $P=0.2$ ). However, based on logistic regression modeling after adjustment for the treatment effect, BOP was significantly reduced in both groups ( $P<0.0001$ ) following the baseline visit. Exploratory subgroup analyses showed that the effect of study drug differed by smoking status ( $P=0.01$ ), where SDD was associated with a reduction in the odds of BOP relative to placebo for non-smokers ( $OR=0.70\%$ , 95% CI: 0.50–1.00,  $P=0.05$ ) and a marginal increase for smokers ( $OR=1.58\%$ , 95% CI: 0.96–2.62,  $P=0.07$ ). The effect of SDD over time differed by site location ( $P=0.01$ ), where the greatest reductions in the odds of BOP with SDD were seen between the molars after 6, 12, and 18 months ( $OR$  ranging from 0.66 to 0.74,  $P=0.1$  for each), while no significant effect was observed at 24 months. Among the subjects who were adherent to the protocol (“per-protocol” analysis), the odds of BOP were 34% lower for subjects receiving SDD relative to placebo ( $OR=0.66\%$ , 95% CI: 0.44–1.00,  $P=0.05$ ) [164].

When the mean GI change assessed by different indexes, Löe and Silness, Cowell, Lobene, and Sulcus Bleeding Index (SBI) were reviewed, minor differences between groups were detected, regardless of the selected parameter [80, 165, 180, 193, 188, 211].

In contrast, in patients with CP, the SDD group (20 mg, 2x, 3 months) showed a significantly greater reduction in mean GI [183] scores at all time points than the placebo group (baseline:  $1.74\pm0.15$ ; 3 months:  $0.54\pm0.09$ ; 6 months:  $0.68\pm0.08$ ; 9 months:  $0.49\pm0.07$ ; 12 months:  $0.46\pm0.10$ ) ( $P<0.05$ ) [51]. Similar results were reported by Emingil et al. [54, 64] and Górska and Nedzi-Góra [55]. The low-dose doxycycline (LDD) group showed statistically significant improvement in GI scores compared to placebo group at 3, 6, and 9 months (LDD group – baseline:  $1.82\pm0.40$ , 3 months:  $0.76\pm0.33$ , 6 months:  $0.83\pm0.25^*$ , 9 months:  $0.70\pm0.29$ , 12 months:  $0.70\pm0.35$  vs placebo – baseline:  $1.89\pm0.29$ , 3 months:  $1.00\pm0.30$ , 6 months:  $1.02\pm0.28$ , 9 months:  $0.97\pm0.33$ , 12 months:  $0.92\pm0.33$ ) ( $P=0.01$ ,  $P=0.01$ , and  $P=0.01$ , respectively) [54]. There was a greater improvement of GI in

the test group (SRP+AZ, 500, 1x, 3 days) than the control (SRP only) [54].

### 3.8.3 PD and CAL Change

In CP patients, differences in mean CAL change, in moderate, deep, and all-pockets categories were evaluated separately. For the all-pockets category, a higher CAL gain was seen in test groups (tetracycline, amoxicillin/metronidazole, metronidazole, doxycycline). However, the magnitude of the additional benefit was small (0.04 mm for SRP to 0.3 mm for SRP+AB). In the moderate pocket (4–6 mm) category, despite obtaining a better result for the adjunctive antimicrobial use (tetracycline, amoxycillin/metronidazole, metronidazole, doxycycline), the magnitude of the differences was again small (0.22 mm for SRP to 0.2 mm for SRP+antibiotics). For deep pockets (>6 mm), better results were revealed for several AB (tetracycline, spiramycin, amoxycillin/metronidazole, metronidazole), with an additional benefit of 0.2–0.6 mm [80].

In CP patients, for all pockets, better results for test groups were obtained (tetracycline, amoxycillin/metronidazole, metronidazole, doxycycline), with an additional benefit ranging from 0.05 to 0.6 mm. For moderate pockets, the magnitude of the differences was limited in both senses. For deep pockets, five studies reported better results for test groups (tetracycline, spiramycin, metronidazole), ranging from 0.2 to 0.8 mm. One additional study [101], using Met achieved a better outcome for SRP+metronidazole of 1.64 mm. Two other studies observed a benefit for SRP (amoxicillin/clavulanate, spiramycin), with differences around 0.3 mm, based on the best results of the control groups [5, 80, 224].

Guerrero et al. [65] revealed that in patients with generalized aggressive periodontitis after an adjunctive course of systemic antibiotic consisting of 500 mg amoxicillin and 500 mg metronidazole three times a day for 7 days, there were highly significant treatment effects for full-mouth PPD reduction, PPD reduction at 4–6 mm pockets, and PPD reduction at ≥7 mm pockets at 2 and 6 months, with the outcomes favoring the test treatment. For PPD reduction in 4–6 mm pockets, the adjusted differences between test and placebo treatment were 0.5 mm at 2 months and 0.4 mm at 6 months. In the deeper pockets (≥7 mm) this difference was

much larger: 0.9 mm at 2 months and 1.4 mm at 6 months. At sites with initial PPD 4–6 mm, there was no statistically significant benefit in terms of 2-month CAL gain ( $P=0.650$ ), a highly significant difference of 0.50 mm in favor of the test group was observed at 6 months ( $P = 0.001$ ). For sites with initial PPD $\geq 7$ , CAL gain was also significantly better in test subjects: an adjunctive benefit of 0.6 mm at 2 months and 1.0 mm at 6 months was observed.

In patients with previously untreated CP and subgingival *A. actinomycetemcomitans* and/or *P. gingivalis*, subgingival scaling without or with systemic amoxicillin plus metronidazole and CHX irrigation, significant differences were found between the two treatment regimes concerning the proportions and absolute values of sites demonstrating attachment gain or loss, in relation to the initial PPD [46]. At sites with an initial PPD of 7 mm or more, scaling plus the administration of adjunctive antimicrobial therapy compared to scaling alone led to a significantly ( $P<0.05$ ) higher proportion of sites gaining attachment ( $37.3\pm4.6\%$  vs  $7.2\pm3.9\%$ , respectively) and to significantly ( $P<0.05$ ) lower proportions of sites losing attachment ( $8.2\pm3.9\%$  vs  $19.1\pm3.1\%$ , respectively; relative risk reduction of 62% for attachment loss). However, at sites with an initial PPD of 0–3 mm and 4–6 mm, respectively, the proportions of sites with an attachment gain or loss of  $\geq 2$  mm did not differ significantly ( $P\geq0.05$ ) between the groups. In addition, a significantly higher ( $P<0.05$ ) mean clinical attachment gain ( $1.7\pm0.3$  vs  $0.3\pm0.3$  mm) was found in test group patients at sites with an initial PPD of  $\geq 7$  mm compared to controls. At sites with an initial PPD of 0–3 mm and 4–6 mm, respectively, no significant ( $P\geq0.05$ ) differences in AL changes between test and control group were detected (PD 0–3 mm:  $0.47\pm0.09$  mm vs  $0.45\pm0.15$  mm; PPD 4–6 mm:  $0.76\pm0.21$  mm vs  $1.04\pm0.26$  mm) [46].

In adult periodontitis patients, there were no statistically significant differences at any visit in average PPD between **azithromycin** (500 mg, once daily for 3 days a week) and the placebo group for pockets which were shallow at baseline. Pockets which were 4–5 mm deep at baseline demonstrated significant improvements in both the azithromycin and placebo groups by week 1 and these differences were maintained at week 22 (adjusted means of both groups was 4.58 mm at baseline, the azithromycin group 2.79 mm at week 22, and the placebo group 3.31 mm at week 22). Also

the mean of average PPD in the azithromycin group was statistically significantly lower ( $P<0.01$ ) than that of the placebo group at all time intervals after the drug had been administered. The mean PPD of initially deep (initially 6 mm or more) pockets in the azithromycin and placebo groups also demonstrated improvements between baseline and 22 weeks (adjusted means for both groups initially being 6.76 mm and then at 22 weeks 3.67 mm for the azithromycin group and 4.54 mm for the control group). Once again the mean PPDs were statistically significantly ( $P < 0.05$ ) shallower in the azithromycin group than in the placebo group at all time intervals after the drug had been administered [193].

Similar results were reported by Mascarenhas et al. [115] who reported after SRP and AZ administration (two 250 mg tablets the first day and one 250 mg tablet for each of the next 4 days), reducing PPD and improving ALs in smokers with moderate to advanced attachment loss. In shallow sites (<4 mm), the data demonstrate that both control and test groups showed a reduction in PPD compared to baseline (0.02 and 0.43 mm, respectively), a difference that was statistically significant for the AZM group at both the 3- and 6-month time points compared to baseline ( $P<0.05$ ). The test group showed a statistically significant CAL gain (0.55 vs 0.11 mm). A statistically significant difference ( $P < 0.05$ ) between groups was only identified at baseline. In moderate sites (PD=4–6 mm), there was a statistically significant difference between groups for the reduction of PPD at 6 months (1.0 and 1.7 mm for control and test groups, respectively,  $P<0.05$ ). CAL levels were statistically different between groups at baseline (4.95 and 5.47 mm,  $P < 0.05$ ) but were identical at 6 months. Drug therapy resulted in a greater reduction of PPD in deep sites (>6 mm) compared to controls (3.52 vs 1.98 mm,  $P<0.05$ ), and this difference was sustained for the duration of the study. CAL changes were larger for the test group compared to the control and the differences were significantly different at 6 months (2.56 vs 1.32 mm,  $P < 0.05$ ). In addition, patients receiving the drug showed an increasing trend in CAL gain at 6 months while the control group showed almost no further gain after 3 months.

In patients with recurrent or refractory disease, several studies have revealed the beneficial effect of systemic antibiotics (doxycycline, amoxicillin/clavulanate, clindamycin, amoxicillin/metronidazole, metronidazole) on periodontal status [73, 92, 108, 115, 118, 119, 217].

In patients with aggressive periodontitis, nine out of ten studies provided evidence for significant CAL change in test groups receiving systemic antibiotics (amoxicillin/metronidazole, metronidazole, azithromycin, minocycline, tetracycline, SDD, doxycycline) compared with controls [65, 68, 86, 132, 147, 154, 177–179, 182, 226] in a study period between 6 and 24 months.

### 3.8.4 Alveolar Bone Loss

Systemic antibiotic therapy may improve radiographic alveolar bone level (Slots and Ting 2002). **Metronidazole** therapy (500 mg, 2×, 7 days; [212]) or for 10 days (200 mg, 3×, 10 days; [179]) revealed a reduction in the proportion of alveolar bone loss >50% at 6 months compared with baseline (from  $15.9 \pm 9.8\%$  to  $9.6 \pm 4.9\%$ ) [212]. In nonsmokers but not in smokers with advanced periodontitis, Söder et al. [194] found that systemic metronidazole therapy (400 mg, 3×, 7 days) led to increased alveolar bone height at 5 years re-evaluation compared with baseline ( $82.7 \pm 4.1$  vs  $80.5 \pm 5.4$ ,  $P < 0.05$ ).

Noval et al. (1991) demonstrated that in early identified lesions of localized juvenile periodontitis the therapy with systemically administered **tetracycline** alone (1 gm/day for 6 weeks) resulted in the arrest of disease progression, decreased PPDs (from 7.1 to 3.6 mm), gains in clinical attachment (from 3.8 to 0.9 mm), and significant repair of alveolar defects (angular bone defects had filled by an average of 72%), up to 4 years after the completion of tetracycline therapy. Similar results were reported by Saxén et al. [177, 178] also in patients with localized juvenile periodontitis.

Payne et al. [149] have evaluated the efficacy of 2-year continuous SDD (20 mg, 2×) on alveolar bone in 123 postmenopausal osteopenic, estrogen-deficient women undergoing periodontal maintenance. There was significant evidence based on the categorical computer-assisted densitometric image analysis (CADIA) measure and the continuous CADIA measure revealed that the treatment effect over time differed by smoking status (time by treatment by subgroup interaction,  $P < 0.01$  for each end point). Among smokers, SDD was associated with reduced alveolar bone density loss at 1-year relative to placebo while no significant

association was seen at 2 years (smokers at 1 year: SDD/placebo OR=0.56, 95% CI: 0.37–0.85,  $P = 0.07$ ; difference in mean change=1.85, 95% CI: 0.053–3.66,  $P = 0.04$ ). Based on the continuous CADIA measure, SDD was associated with reduced alveolar bone density loss relative to placebo among nonsmoking subjects (difference in mean change=1.13, 95% CI: 0.003–2.25,  $P = 0.05$ ). Based on the continuous measure of CADIA change, there was a significant interaction among study drug, time, and baseline PPD ( $P = 0.03$ ). Among sites with a baseline PPD  $\geq 5$  mm, SDD was associated with reduced alveolar bone density loss relative to placebo (difference in mean change=2.45, 95% CI: 0.85–4.04,  $P = 0.003$ ). There was significant evidence that the effect of the study drug differed by time after menopause for alveolar bone height change (drug by menopause interaction,  $P = 0.04$ ). Among subjects who were beyond 5 years of menopause, SDD was associated with a 29% reduction in the odds of more progressive disease (bone height loss) (SDD/placebo OR=0.71, 95% CI: 0.50–0.99,  $P = 0.05$ ).

### 3.8.5 Gingival Crevicular Fluid Changes

Very few studies have assessed the impact of adjunctive systemic antibiotic therapy on GCF. Compared with controls who received only full-mouth SRP, in patients with both CP and coronary artery disease, the systemic SDD administration in conjunction with SRP (20 mg, 2×, 6 weeks) did not statistically significantly reduce the two groups for MMP-1, -8, and -13 and total collagenase in GCF, after 6 weeks of treatment [202].

In patients with CP, from baseline to 12 months, GCF scores and GCF MMP-8 levels were significantly reduced in two study groups: SRP+OHI (oral hygiene instructions)+SDD (20 mg, 2×, 3 months) versus SRP+OHI+placebo (SDD group: from  $0.58 \pm 0.04$  mL at baseline to  $0.15 \pm 0.04$  mL at 12 months; placebo group: from  $0.50 \pm 0.05$  mL at baseline to  $0.29 \pm 0.06$  mL at 12 months) ( $P < 0.0125$ ). Hence, decreased enzyme levels in both LDD and placebo groups during active periodontal therapy may suggest the effectiveness of nonsurgical periodontal therapy in reducing the bacterial load in the periodontal environment. The GCF MMP-8 total amount in the placebo group tended to increase at 6 months compared with levels detected at

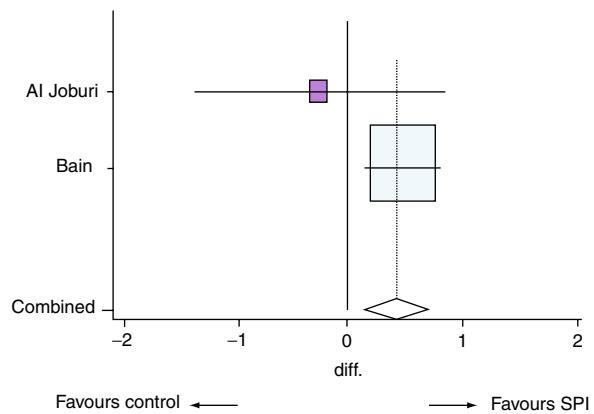
3 months, while on the contrary, GCF MMP-8 levels in the SDD group continued to decrease over a 6-month period. Thereafter, no additional decrease was observed in either group, although the levels were still significantly lower than the baseline levels. The total amount of GCF MMP-8 in the LDD group was found to be about 50% lower than that of the placebo group at 3 months and about 70% at 6 months. After 6 months, adjunctive LDD therapy did not seem to provide any additional effect on GCF levels. Based on these finding it can be suggested that decreased GCF MMP-8 levels in the LDD group during 6 months could be depended on the host-modulation properties of the LDD [51].

In postmenopausal osteopenic women, the SDD-treated postmenopausal women showed ~50% reduction in GCF collagenase activity over the 2 years compared with their own baseline values. In contrast, the placebo values appeared to decrease only slightly. Moreover, based on linear regression analysis, the SDD-treated group showed a statistically significant 22% reduction in median GCF collagenase activity compared to placebo-treated subjects over the study period, based on intent-to-treat analysis (95% CI: 37% lower to 5% lower;  $P=0.01$ ), and a 29% reduction in median GCF collagenase activity compared to placebo subjects based on the per-protocol analysis (95% CI: 48% lower to 4% lower;  $P=0.02$ ) after adjusting for baseline values. For subgroup analyses, the effect of SDD seemed to depend on smoking status ( $P=0.05$ ), and there was a significant interaction between time and treatment for nonsmokers ( $P=0.02$ ). At 1 year, median levels of collagenase activity per pool of GCF were 40% lower for SDD subjects compared to placebo subjects in the nonsmoking group, which was statistically significant (95% CI: 53% lower to 22% lower;  $P<0.0001$ ) [58].

ICTP (pyridinoline cross-linked carboxyterminal telopeptide of type I collagen) is a breakdown product of type I collagen, and this collagen makes up >90% of the organic matrix of bone. In contrast, SDD therapy over the study period seemed to reduce the median ICTP levels per pool of GCF by ~30% compared to this group's own baseline values. Using linear regression analysis, the SDD-treated group showed a 16% reduction in median GCF ICTP levels compared to placebo-treated subjects, after adjusting for baseline values ( $P=0.08$ ). Focusing on changes in the dominant type of collagenase, MMP-8, in the GCF of these postmenopausal women, and based on

intent-to-treat analysis, SDD therapy reduced the odds of elevated MMP-8 values (across the ordered categories of 0–1.00, 1.001–2.5, and >2.5 units) by 60% compared to placebo during the 2-year study period. This treatment effect was highly statistically significant ( $OR=0.40$ ; 95% CI: 0.21–0.77;  $P=0.006$ ). Consistent with this pattern, SDD therapy increased the odds of lower values (among the ordered categories of 0–1.00, 1.001–2.5, and >2.5 units) for this type of collagenase, compared to placebo therapy, over the study period. Based on per-protocol analysis, this effect was even more dramatic because the odds of higher values for MMP-8 in SDD-treated subjects were 78% lower than in those receiving placebo tablets ( $OR=0.22$ ; 95% CI: 0.07–0.66;  $P=0.007$ ) [58].

Laminin-5 is a well-known epithelial cell-derived adhesive protein localized to the anchoring filaments within the lamina lucida space of the basal membrane zone of the junctional and gingival epithelium. While intact laminin-5 serves to anchor epithelial cells on the BM, it stimulates epithelial cell migration after having been cleaved by proteolytic enzymes. The cleaved Ln-5  $\gamma 2$  chain can direct inflammatory reactions by regulating cell adhesion, migration, and proliferation of fibroblasts and epithelial cells, and also act as chemoattractant for leukocytes. After LDD plus SRP, the total amount of GCF Ln-5  $\gamma 2$  chain fragments significantly decreased at 3 months compared to baseline (Fig. 3.4) ( $P=0.0125$ ). The 6-month levels were similar



**Fig. 3.4** 23Meta-analyses (Forrest plot) comparing periodontal probing depth (PPD) change in deep pockets for spiramycin. *diff.* Difference between mean changes for scaling and root planning (SRP) and SRP + spiramycin, in millimeters [80] (Reprinted with permission John Wiley & Sons)

to those at 3 months. The Ln-5  $\gamma$ 2 chain fragment levels slightly increased at 9 and 12 months, with no significant differences compared to baseline ( $P > 0.0125$ ). The mean changes in the total amount of the GCF Ln-5  $\gamma$ 2 fragments at 3, 9, and 12 months in the SRP plus LDD group were greater than those obtained by the SRP plus placebo therapy ( $P = 0.0211$ ,  $P = 0.0102$ ,  $P = 0.0233$ , respectively), although at 6 months the mean changes were similar between the two groups ( $P > 0.05$ ). When the GCF Ln-5  $\gamma$ 2 chain fragments data were expressed as concentrations, no significant decrease was found in the GCF Ln-5  $\gamma$ 2 chain fragment concentration in either group at any time point compared to baseline ( $P > 0.0125$ ) [52].

The plasminogen-activating system is activated by plasminogen activators (PAs), which belong to the trypsin-like serine protease family. The destructive potential of the PAs plays an important role in the spread of inflammatory reactions and thereby could contribute to the initiation and progression of periodontal disease. After LDD plus SRP therapy, GCF t-PA total amount showed marked improvements over the course of the study. In other words, the enzyme levels significantly decreased compared to baseline at all time points ( $P < 0.0125$ ). GCF t-PA total amount was still significantly low at 9 months compared to the baseline values ( $P < 0.0125$ ), but this amount tended to increase at 12 months compared to 9 months, which were still significantly lower than the baseline levels ( $P < 0.0125$ ) [54].

SDD therapy in combination with SRP reduced GCF EMMPRIN levels, also called CD147, a highly glycosylated plasma membrane-bound glycoprotein of 45–55 kDa. [53].

GCF bone marker assessment (ICTP: pyridinoline cross-linked carboxyterminal telopeptide of type I collagen) levels in response to SRP alone or SRP+aztreomycin (two 250 mg tablets the first day and one 250 mg tablet for each of the next 4 days) were reduced in both groups after baseline evaluation. The observed reduction was statistically different from baseline at the 14-day and 6-month time points, and was slightly greater for test group but the difference did not reach a statistically significant level. At the 6-month evaluation, the reduction in ICTP was 65.9 pg/site in the control group compared to 87.5 pg/site in the test group. For both groups, the ICTP reduction at 3 months was not statistically significant compared to baseline ( $P > 0.05$ ) [115].

In CP patients, azithromycin administration in conjunction with full-mouth SRP (SRP+AZ, 500, 1 $\times$ ,

3 days) significantly reduced the GCF volume. The volume of GCF at baseline was assumed to be 100%. In the test group, GCF at 5 weeks from baseline was  $50.7 \pm 16.1\%$ , and GCF remained at the same level after 13 and 25 weeks at  $47.9 \pm 20.2\%$  and  $53.9 \pm 24.0\%$ , respectively. In the control group (SRP only), decrease of GCF was minimal; the values at 13 and 25 weeks from baseline were  $88.6 \pm 31.0\%$  and  $102.6 \pm 39.7\%$ , respectively. A statistically significant difference ( $P < 0.01$ ) in GCF was recognized between the test and control groups at 13 and 25 weeks [61].

## 3.9 Limitations of Available Data

As Herrera et al. ([80]; Herrera et al. [78]) summarized, the heterogeneity of the experimental designs used precludes any attempt at a more systematic approach in reviewing the literature, including meta-analysis.

This variability includes the following.

### 3.9.1 The Type of the Periodontal Disease

Most of the groups for comparison had an initial diagnosis of adult periodontitis, or what can be considered nowadays as CP. Additionally, cases with progressive periodontitis, recurrent periodontitis, refractory periodontitis, localized juvenile periodontitis, and generalized aggressive periodontitis were also studied (Table 3.11). Most of the studies included patients untreated, or those after a minimum period of 3 months, 6 months, or 6 years since the last treatment [80].

### 3.9.2 The Number of Subjects

Loesche et al. [102] revealed that the use of probing measurements as an outcome has an effect on the sizes of the treatment groups. The measurement of ALs has a standard deviation of +1 mm and this measurement error, i.e., 33% in a 6-mm pocket, decreases the ability to show a treatment effect between/among groups. As a result, it would increase the need for a larger sample size for statistical significance to be shown. Power calculations indicate that about 25–30 patients would be

**Table 3.11** Summary of particular aspects of studies assessing systemic antibiotic therapy as adjunct to periodontal therapy

| Reference | Study design     | Aggressive periodontitis/<br>chronic periodontitis | Nr. of baseline | % Smokers | Age range | Gender male/<br>female   | Type of AB | Placebo | Nr. of study groups | Assessment of the compliance of treatment | % Compliance of treatment   | Study length |          |
|-----------|------------------|--|-----------------|-----------|-----------|--|------------|---------|---------------------|---|---|--------------|----------|
| [4]       | RCT              | CP   | 45              | NA        | 0         | 33–61  | 21/24      | DOXY    | No                  | 3   | Not given   | Not given    |          |
| [5]       | RCT              | CP   | 96              | 79        | NA        | NA   | >34        | SPIR    | Yes                 | 3   | Not given   | Not given    |          |
| [13]      | RCT              | CP   | 16              | 16        | NA        | 35–58 years  | 6/10       | A/M     | Yes                 | 4   | Not given   | Not given    |          |
| [24]      | RCT              | CP   | 60              | 44        | 27.27%    | >35 years  | 14/30      | MET     | Yes                 | 4   | Counting the remaining capsules at day 10 and calling the subject every 2 days during the antibiotic administration phase | Not given    |          |
| [23]      | RCT              | CP   | 60              | 44        | 27.27%    | >35 years  | 14/30      | MET     | Yes                 | 4   | Counting the remaining capsules at day 10 and calling the subject every 2 days during the antibiotic administration phase | Not given    |          |
| [26]      | RCT multi-center | CP   | 190             | 183       | 40.43%    | 30–75 years  | 94/89      | SDD     | Yes                 | 2   | Not given   | Not given    |          |
| [30]      | RCT              | CP   | 32              | 32        | Not given | 25–64 years  | 17/15      | SDD     | Yes                 | 2   | Not given   | Not given    |          |
| [34]      | RCT              | CP   | 51              | 47        | 34.04%    | Mean age (control:<br>$50.5 \pm 13.6$ years/<br>Test:<br>$50.6 \pm 8.6$ years) | 17/30      | A/M     | Yes                 | 2   | Returned pills  | Not given    | 6 months |

|      |     |    |    |    |   | A/M  | Yes   | 2   | Returned pills | Not given | 6 months  |
|------|-----|----|----|----|---|--|-------|-----|----------------|-----------|---|
| [35] | RCT | CP | 51 | 47 | 34.04%  | Mean age:<br>(Control:<br>$50.5 \pm 13.6$ years/<br>test:<br>$50.6 \pm 8.6$ years) |       |     |                |           |   |
| [41] | RCT | CP | 30 | 30 | 100%<br>Smokers<br>(≥one pack<br>of cigarettes/<br>day) | 35–65<br>49.40–7.81  | 17/13 | AZ  | Yes            | 2         | Verbally asking<br>the patients if<br>they consumed<br>all tablets as<br>directed,<br>requesting that<br>they return to<br>the 2-week<br>appointment<br>with the vial that<br>contained the<br>medication/<br>placebo, and<br>counting how<br>many tablets<br>remained. |
| [46] | RCT | CP | 48 | 35 | 8.57%   | Mean age:<br>$51.1 \pm 10.7$ years   | 16/19 | A/M | No             | 2         | Counting pills  |
| [51] | RCT | CP | 30 | 20 | 40%   | 37–61  | 10/10 | SDD | Yes            | 2         | Giving the<br>capsules in<br>labeled bottles<br>biweekly  |
| [51] | RCT | CP | 30 | 20 | 40%   | 37–61  | 10/10 | SDD | Yes            | 2         | Giving the<br>capsules in<br>labeled bottles<br>biweekly  |
| [54] | RCT | CP | 65 | 46 | 45.65%  | 34–61  | 30/16 | SDD | Yes            | 2         | Giving the<br>capsules in<br>labeled bottles<br>biweekly  |

(continued)

Table 3.11 (continued)

| Reference | Study design      | Aggressive periodontitis/<br>chronic periodontitis | Nr. of baseline | Nr. of final | % Smokers  | Age range                        | Gender male/<br>female | Type of AB   | Placebo | Nr. of study groups            | Assessment of the compliance of treatment   | % Compliance of treatment | Study length |
|-----------|-------------------|--|-----------------|--------------|------------|----------------------------------|------------------------|--------------|---------|--------------------------------|---|---------------------------|--------------|
| [53]      | RCT               | CP   | 30              | 24           | 41.67%     | 37–61                            | 12/12                  | SDD          | No      | 2                              | Giving the capsules in labeled bottles biweekly and was assessed by counting the number of pills returned | Not given                 | 6 months     |
| [57]      | RCT               | CP   | 48              | 38           | Not given  | >30 years, mean age 46 years     | 17/21                  | A/M          | No      | Counting the number of tablets | 99.8% for the prescribed MET and 93.9% for MOX  | 1 year                    |              |
| [61]      | RCT               | CP   | 34              | 34           | 0          | >25 years                        | 16/18                  | AZ           | No      | 2                              | Not given   | Not given                 | 25 weeks     |
| [64]      | RCT               | CP   | 66              | 66           | Not stated | 20–56 years (mean age: 43 years) | 15/18                  | SDD          | No      | 2                              | Not given   | Not given                 | 12 months    |
| [65]      | RCT               | AgP  | 41              | 41           | 21.95%     | Mean age 31.3 (28.8, 33.7)       | 13/28                  | A/M          | No      | 2                              | Pill counts   | 90.24%                    | 6 months     |
| [66]      | RCT               | CP   | 35              | 26           | 34.61%     | 34–56                            | 19/7                   | SDD          | Yes     | 2                              | Pill counts   | Not given                 | 1 year       |
| [68]      | RCT               | AgP  | 25              | 24           | 20.83%     | 13–26                            | 13/11                  | AZ           | Yes     | 2                              | Pill counts   | 100                       | 1 year       |
| [72]      | RCT               | AgP and ChP  | 98              | 92           | 10%        | 22–77                            | 59/33                  | AZ, MET, SDD | No      | 4                              | Pill counts   | >95%                      | 1 year       |
| [86]      | RCT               | AgP  | 36              | 36           | 50%        | 21–39                            | 15/21                  | A/M          | No      | 2                              | Pill counts   | 94.44%                    | 6 months     |
| [87, 89]  | CCT               | CP   | 35              | 35           | NA         | Mean age 32.5±8 years            | 13/19                  | OFL/O        | No      | 2                              | Not given   | Not given                 | 12 months    |
| [103]     | RCT (split mouth) | AgP and CP   | 25              | 25           | 52%        | 16–58                            | 12/13                  | A/M          | No      | 2                              | Not given   | Not given                 | 12 months    |
| [104]     | RCT               | CP   | 46              | 39           | 41.025     | 38–68                            | 6/39                   | A/M          | Yes     | 2                              | Pills count   | 92.03%                    | 12 months    |

|            |     |            |     |     |                     |                           |           |             |     |   |  |   |           |
|------------|-----|------------|-----|-----|---------------------|---------------------------|-----------|-------------|-----|---|--|---|-----------|
| [105]      | RCT | CP         | 22  | 22  | 40.90%<br>(9 OUT22) | 38–68                     | 7/15      | A/M         | Yes | 2 | A decline or disappearance of spirochetes from subgingival plaque has been suggested as a means for measuring patient compliance in taking metronidazole | 100%  | 12 months |
| [115]      | RCT | CP         | 31  | 30  | 100%                | 30 years old or older     | 19/11     | AZ          | No  | 2 | Answered questionnaires and pill count   | 100%  | 6 months  |
| [116]      | RCT | CP         | 45  | 43  | 100%                | >30 years (mean 40.5±8.2) | 19/24     | A/M,<br>MET | Yes | 3 | Pills counts<br>Phone calls for monitoring compliance  | Not given   | 3 months  |
| [122]      | RCT | CP         | 55  | 50  | 0                   | 21/29                     | 17–51     | A/M         | Yes | 2 | Pills counts   | Not given   | 8 weeks   |
| [125]      | RCT | AgP and CP | 16  | 16  | 42.85%              | 25–65                     | Not given | A/M         | Yes | 2 | Not given  | Not given   | 12 months |
| [135]      | CCT | CP         | 32  | 32  | 47%                 | 32–72<br>12–70 years      | 18/14     | DOXY        | Yes | 4 | Not given  | Not given   | 24 weeks  |
| [136]      | RCT | CP         | 180 | 171 | 42.69%              | 24–71                     | 77/94     | SDD         | Yes | 2 | Pills counts   | >83.33%   | 6 months  |
| [146]      | RCT | CP         | 90  | 85  | 32.94%              | 35–65                     | 43/47     | MET         | No  | 3 | Not given  | Not given   | 6 months  |
| [149]      | RCT | CP         | 128 | 113 | 20.31%              | 45–70                     | 0/128     | SDD         | Yes | 2 | Pills count  | 11–15% of the placebo subjects took ≥80% of the prescribed study drug compared with 4–14% of the SDD subjects | 2 years   |
| [152, 153] | RCT | CP         | 209 | 157 | 32.05%              | 30–75                     | 125/84    | SDD         | Yes | 2 | Pill counts  | >87%  | 9 months  |

*(continued)*

Table 3.11 (continued)

| Reference | Study design | Aggressive periodontitis/ chronic periodontitis | Nr. of baseline | Nr. of final                        | % Smokers       | Age range | Gender male/ female   | Type of AB      | Placebo | Nr. of study groups | % Compliance of treatment  | Study length        |
|-----------|--------------|---|-----------------|-------------------------------------|-----------------|-----------|-----------------------|-----------------|---------|---------------------|--|---------------------|
| [154]     | RCT          | AgP and ChP                                     | 266             | 227                                 | 28.94% (77/266) | 23–82     | 120/146               | SDD-40          | Yes     | 2                   | Compliance with study drug therapy was recorded by counting the number of tablets dispensed and returned | 92–95% 9 months     |
| [160]     | CCT          | CP  | 115             | 89                                  | 57.39%          | 24–60     | 54/61                 | TET             | No      | 2                   | Not given  | 13 years            |
| [164]     | RCT          | CP  | 128             | 113                                 | 20.31%          | 45–70     | 0/128                 | SDD             | Yes     | 2                   | Pills count  | 78.76% 2 years      |
| [58]      | RCT          | CP  | 128             | 113                                 | 20.31%          | 45–70     | 0/128                 | SDD             | Yes     | 2                   | Pills count  | 78.76% 2 years      |
| [165]     | RCT          | CP  | 28              | 25                                  | 0               | 30–66     | 8/17                  | A/M             | Yes     | 2                   | Pill counts  | 76.92% 6 months     |
| [171]     | RCT          | AgP and CP                                      | 66              | 62                                  | Not given       | Not given | 20–45                 | A/M, MET, AMOX  | No      | 4                   | Not given  | Not given 6 months  |
| [180]     | RCT          | Deep intrabony defects                          | 34              | 34                                  | 20.58%          | 12/22     | Not given             | A/M             | No      | 2                   | Not given  | Not given 1 year    |
| [182]     | RCT          | AgP   | 48              | 48                                  | 0               | 20/28     | Mean age 32.4 years   | DOXY, MET, CLIN | No      | 4                   | Not given  | Not given 24 months |
| [193]     | RCT          | AgP and CP                                      | 46              | 44                                  | 22.72%          | 21/23     | Mean age 42.68 (8.66) | AZ              | Yes     | 2                   | Not given  | Not given 22 weeks  |
| [195]     | RCT          | CP  | 98              | 92                                  | NA              | 52/46     | 35–45 years           | MET             | Yes     | 2                   | Not given  | Not given 6 months  |
| [194]     | RCT          | CP  | 98              | 64 (37 smokers and 27 non-smokers), | 42.18%          | 32/32     | Mean age 36.5 years   | MET             | Yes     | 2                   | Not given  | Not given 5 years   |

|       |     |                    |     |     |        |             |                                      |               |     |   |  |           |           |
|-------|-----|--------------------|-----|-----|--------|-------------|--------------------------------------|---------------|-----|---|--|-----------|-----------|
| [202] | RCT | CP                 | 36  | 36  | 0      | 35/3        | Less than 70 years (52.2±6.91 years) | SDD           | Yes | 2 | Subjects were questioned                                 | Not given | 6 weeks   |
| [211] | RCT | Class II furcation | 24  | NA  | 54.16% | NA          | NA                                   | MET, CIPR     | No  | 2 | Not given  | Not given | 9 months  |
| [216] | RCT | CP                 | 128 | 110 | 20%    | 45–75 years | 0/128                                | SDD           | Yes | 2 | Pill counts  | Not given | 2 years   |
| [224] | RCT | CP                 | 21  | 21  | 23.8%  | 28–66       | 6/15                                 | AMOXI/ CLAV   | Yes | 2 | Booklet of medication, pill counts                       | Not given | 12 months |
| [225] | RCT | CP                 | 54  | 49  | 65.30% | 28–63       | 21/28                                | A/M           | Yes | 2 | Return any medication tablets that remained after 7 days | 100%      | 3 months  |
| [226] | RCT | AgP                | 47  | 43  | 34.88% | 22–49       | 22/21                                | AM, DOXY, MET | No  | 4 | No   | Not given | 6 months  |

*AZI* azithromycin, *SP* spiramycin, *TET* tetracycline, *DOX* doxycycline, *MET* metronidazole, *CIN* clindamycin, *AMO*/CLAV amoxicillin plus clavulamate, *RCT* randomized clinical trial, *CCT* controlled clinical trial, *NA* not available, *w* weeks; *m* months; *y* years, *Ag* P aggressive periodontitis, *Ch* P chronic periodontitis, *AgP* aggressive periodontitis

needed in each treatment group to have a difference of 1 mm in AL to be significant between/among the groups. In the majority of the clinical studies, there were 15 or fewer subjects in each treatment group, so that only a very obvious difference between/among the groups would be significant. Thus, the possibility of a Type 2 error or false-negative result is great, i.e., reporting no treatment effect when indeed there may be one [102].

However, several recent studies have addressed this problem by calculating the necessary sample size for investigation [53, 65, 67, 68, 72, 86, 116, 123, 149, 154, 165, 180, 202, 211, 226].

Power analysis to determine superiority of antibiotic treatment showed that a 12 per group sample size would yield 93% power to detect a 1.5 mm difference and 64% power to detect a 1 mm difference in the study performed by Vest et al. [211].

Guerrero et al. [65] stated that the sample size calculation determined by the 17 subjects per treatment arm would provide 80% power to detect a true difference of 1.0 mm between test and placebo using PPD reduction in pockets  $\geq 7$  mm as the primary outcome variable, assuming that the common standard deviation is 1.0 mm. Accordingly, a sample of 21 subjects per arm (42 in total) were to be recruited to compensate for possible drop-out during the study period.

The sample size of 12 subjects in each of the two treatment groups was calculated by Mohammad et al. [123] to yield >80% power of detecting a statistically significant result assuming a within-group standard deviation of 1.5 mm and a mean difference of 1.8 mm ( $\alpha=0.05$ ).

According to calculations of Xajigeorgiou et al. [226] in order to detect differences of  $2 \pm 1$  mm PPD at 95% power analysis their groups should include 25 subjects each (Statmate2 Graphpad Inc., San Diego, CA, USA).

Kaner et al. [86] considered the review done by Herrera et al. [80] which revealed that adjunctive systemic antibiotics may result in an additional reduction of approximately 0.5 mm (range of 0.06–0.6 mm) for mean full-mouth PPD when compared with SRP alone. For this reason, a difference of 0.5 mm between groups for mean full-mouth PPD reduction after 6 months was considered to be clinically relevant. Assuming 0.5 mm as the common standard deviation of full-mouth PPD change within both groups, 16 patients per treatment group would provide 80% power to detect a true difference. To

compensate for eventual drop-outs, 18 patients were recruited per treatment group.

As the major difference that occurs as the result of adjunctive systemic antibiotics takes place at sites with deeper PPDs, the power calculations in the study performed by Haffajee et al. [72] were based on sites with  $PPD > 6$  mm. It was felt that a difference of 1 mm between groups for CAL change at these sites would be clinically significant. Further, it was determined that the standard deviation of CAL change at sites with  $PPD > 6$  mm was 1.1 mm based on their earlier studies of subjects receiving SRP alone or combined with different adjunctive antibiotics. Based on these values, the study would require 20 subjects per group with an  $\alpha$  of 0.05 and 80% power, for a total of 80 subjects. Based on anticipated attrition of about 15%, 98 subjects were recruited.

Payne et al. [149] showed that a total sample size of 102 subjects (51 per treatment group), with an average of 18 tooth- or site-level measures made at two follow-up time points, results in an 80% power to detect true differences between the placebo probability of alveolar bone density loss of 14% versus 7% in the SDD arm assuming a two-sided significance level of 0.05 and an exchangeable correlation parameter of 0.14. The estimated probabilities of bone density loss were based on unpublished pilot data using the CADIA method, as measurements using the RA method were not available in this pilot study. To adjust for an expected 20% drop-out rate before the 2-year visit, the total number of randomized subjects was 128, or 64 per treatment group.

Tütür et al. [202] could not prospectively calculate the power needed in their study from the literature because the literature did not contain any studies that examined the effects of SRP and LDD on subjects who were diagnosed with both periodontitis and coronary artery disease. Using the data that were generated with their study, they retrospectively calculated the power [(1- $\beta$ ) calculation] to observe a statistically significant difference in PPD between the two groups of 82.96%. A  $P$  value  $<0.05$  was considered to be statistically significant for all statistical tests.

Emingil et al. [53] selected the sample size to detect a 0.5-mm mean difference in PPD and CAL values between treatment groups with a significance level of 0.05% and 80% power.

The study performed by Gürkan et al. [67] was planned to detect a 1.0-mm difference between treatment groups in PPD reduction from baseline for deep sites and

0.6-mm intergroup difference for moderate sites with a significance level of 0.05 and 80% power.

Preshaw et al. [154] designed their study to detect a difference  $\geq 0.2$  mm in mean change from baseline CAL between treatment arms (two-tailed tests) with 80% power, assuming that 72 subjects would complete the study in each group. An intent-to-treat analysis strategy was used for all subjects who were randomized to study medication and who took at least one dose.

The ideal sample size to assure adequate power to the clinical trial performed by Matarazzo et al. [116] was calculated considering differences of at least 1 m for CAL and a standard deviation of 0.94 mm between groups in initially deep periodontal pockets ( $>6$  mm). Based on these calculations, it was defined that 14 subjects per group would be necessary to provide an 80% power with an  $\alpha$  of 0.05.

According to Haas et al. [68], based on a preliminary analysis, a sample size of 24 subjects was estimated to be necessary to achieve 80% power to detect a difference of 1 mm (SD 0.85) between the two groups' mean PPD reduction. This mean difference was chosen for being regarded as clinically relevant for an adjunct treatment and greater than the measurement error of the examiner. A two-sided two-sample *t*-test with a significance level of 5% was used for the sample size calculation. An attrition rate of 15% was assumed, yielding a total number of 28 recruited subjects.

In the study conducted by Ribeiro et al. [165], power analysis indicated that with 12 subjects in each group, the study would have  $>80\%$  power to detect a 1-mm difference in CAL between groups.

The problem of a small sample size can be partially addressed by the statistical technique known as meta-analysis. Meta-analysis represents a re-analysis of published data in which results from several studies are combined if they meet predetermined inclusion criteria. The added statistical power obtained with a larger sample size then improves the chances that statistical significance can be shown where previously none was found [102].

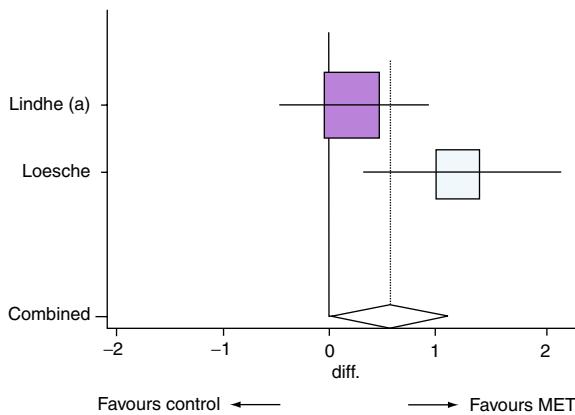
Several meta-analyses were performed for analyzing the efficacy of systemic antibiotics in periodontal therapy. In a meta-analysis assessing the use of systemic tetracycline in the treatment of periodontal disease Hayes et al. [75] concluded that data from the published literature does not demonstrate that the use of systemic tetracycline is more beneficial than conventional treatment in

the management of adult periodontal disease. Elter et al. [50] suggested that metronidazole in conjunction with SRP may offer a benefit over SRP alone in the treatment of adult periodontitis patients in managing pockets of 4 mm or greater, but the additional benefit would not be evident if initial PPD is less than 4 mm or follow-up is beyond 13 weeks.

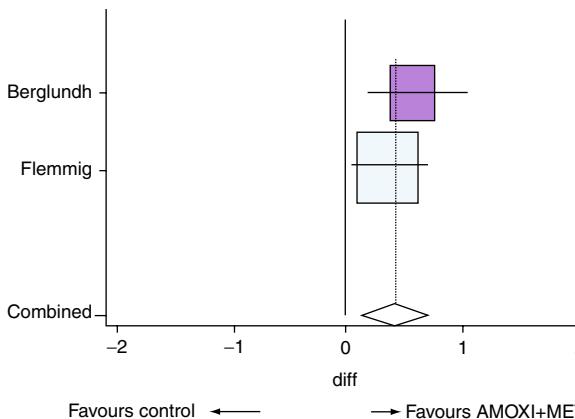
Preshaw et al. [152] performed a meta-analysis of the data gathered in two studies [26, 151] investigating the efficacy of adjunctive SDD in the treatment of CP. At each time point, adjunctive SDD resulted in statistically significantly greater mean PPD reductions and AL gains compared with SRP alone ( $P < 0.05$  in all cases). The benefits of adjunctive SDD were apparent as early as 3 months after commencing treatment, and were maintained for the 9 months of the study.

Another meta-analysis was performed by Reddy et al. [163] based on the evaluation of seven papers [7, 26, 39, 59, 137, 153, 155]. For sites with pretreatment PPD of 4–6 mm and  $\geq 7$  mm, a statistically significant adjunctive benefit of CAL was found when SDD was used in combination with SRP. For PPD changes, a significant adjunctive benefit was noted after a combination of SRP and SDD for pretreatment PPDs of 4–6 mm and  $\geq 7$  mm [163].

Herrera et al. [80] assessed the effectiveness of the adjunctive use of systemic antimicrobials with SRP versus SRP alone in the treatment of chronic or aggressive periodontitis. After an initial selection, 158 papers were identified by the manual and electronic searches; 25 papers were eligible for inclusion. In general, selected studies showed high variability and lack of relevant information for an adequate assessment, and as a consequence meta-analyses had to be restricted to a limited number of drugs (spiramycin, amoxicillin plus metronidazole, and metronidazole), and only two studies for each drug were included. The meta-analyses showed a statistically significant additional effect of spiramycin with regard to PPD change (pooled estimate 0.407; 95% CI: 0.081, 0.733;  $P = 0.014$ ; Fig. 3.5), and of amoxicillin plus metronidazole with regard to CAL change (pooled estimate 0.450; 95% CI: 0.192, 0.709;  $P = 0.001$ ; Fig. 3.6), both for initial PPD  $> 6$  mm. For CAL change in initially deep pockets, the effect of metronidazole was close to the level of statistical significance (pooled estimate 0.551; 95% CI: -0.017, 1.119;  $P = 0.057$ ; Fig. 3.5), while spiramycin for deep pockets (PD  $> 6$  mm) (pooled estimate 0.262; 95% CI: -0.044, 0.569;  $P = 0.093$ ) and amoxicillin plus metronidazole



**Fig. 3.5** Meta-analyses (Forrest plot) comparing clinical attachment level (CAL) change in deep pockets for metronidazole. *diff.*: Difference between mean changes for scaling and root planning (SRP) and SRP+metronidazole, in millimeters [80] (Reprinted with permission John Wiley & Sons)



**Fig. 3.6** Meta-analyses (Forrest plot) comparing clinical attachment level (CAL) change in deep pockets for amoxicillin plus metronidazole. *diff.*: Difference between mean changes for scaling and root planning (SRP) and SRP+spiramycin, in millimeters [80] (Reprinted with permission John Wiley & Sons)

for moderate pockets (PD 4–6 mm) (pooled estimate 0.154; 95% CI: −0.172, 0.480;  $P = 0.354$ ) did not reach significant results with regard to CAL change. In summary, systemic antimicrobials in conjunction with SRP can offer an additional benefit over SRP alone in the treatment of periodontitis, in terms of CAL and PPD change, and reduced risk of additional CAL loss [80].

Another meta-analysis performed by Haffajee et al. [71] evaluated whether systemically administered antibiotics improve a primary clinical outcome measure, periodontal AL change. Studies involving the use of LDD, combinations of local plus systemic antibiotics, or

where the control group included a systemically administered antibiotic were excluded. Twenty-two studies (27 comparisons) were used in the meta-analysis, evaluating if the antibiotics provided a consistent benefit in mean CAL change for different patient populations, for different therapies, and for different antibiotics. For the majority of the comparisons, systemically administered antibiotics exhibited a more positive AL change than the control group in the study. The combined results were statistically significant ( $P < 0.001$ ). The systemic antibiotics were uniformly beneficial in providing an improvement in AL when used as adjuncts to SRP and were consistently beneficial, although of borderline significance, when used as adjuncts to SRP plus surgery or as a stand-alone therapy. When examining the effects of individual or combination antibiotics, it was found that there were statistically significant improvements in CAL for tetracycline, metronidazole, and an effect of borderline statistical significance for the combination of amoxicillin plus metronidazole. Improvements in mean CAL were consistent for both chronic and aggressive periodontitis subjects, although the aggressive periodontitis patients benefited more from the antibiotics. It was concluded that the use of systemically administered adjunctive antibiotics with and without SRP and/or surgery appeared to provide a greater clinical improvement in AL than therapies not employing these agents.

A recent cumulative meta-analysis by Moles et al. [124] reviewed 25 studies and included five trials [12, 57, 95, 108, 145, 224] investigating the effectiveness of a variety of antibiotic systems: long-term tetracycline, Augmentin, metronidazole, and a combination of metronidazole and amoxicillin. They reported that the overall effect size for adjunctive antimicrobials in all/moderate sites from the meta-analyses is a mean difference in CAL change of 0.18 mm (95% CI: 0.00, 0.37;  $P = 0.05$ ) in favor of the adjunctive antimicrobial. For deep sites, the conventional meta-analysis indicates a mean difference in CAL change of 0.37 mm (95% CI 0.12, 0.61;  $P = 0.004$ ).

### 3.9.3 Characteristics of the Study Population

In regard to smoking, very few studies assessed this habit. In six studies, smokers were excluded [4, 60, 61, 122, 165, 182, 202], while in two studies the study

group consisted of only smokers [41, 116]. Gender distribution and the range of ages were usually described (Table 3.11).

### **3.9.4 The Nature of the Clinical Measurements Performed in the Studies**

The best clinical outcome for a periodontal patient following treatment would be the retention of formerly diseased teeth for a lifetime. The measurement of tooth loss following treatment would be a clinical outcome that would be appreciated by the patient and would meet the definition of a true clinical end point [81, 102].

Only one study has related the antibiotic use for medical or dental reasons to subsequent tooth loss in a cohort of 12,631 persons with destructive periodontal disease [40]. The number of days of antibiotics dispensed during the first 3 years of the study for medical or dental reasons was categorized as (a) no use, (b) 1–13 days, (c) 14–20 days, or (d) 21 or more days. The categories were chosen to reflect the possible prescription patterns of physicians and dentists, i.e., antibiotics filled for 1–13 days, representing one course of the prescribed antibiotics; 14–20 days, representing two courses; and 21 or more days, representing three or more courses. The analysis was performed for any antibiotic use and for the following antibiotic classes: penicillins, clindamycin, tetracyclines, metronidazole, and macrolides.

It was found that the antibiotic use for 1–13 days and for 14–20 days during the first 3-year period was not associated with subsequent tooth loss (rate ratio (RR)=1.0; 95% CI=0.8, 1.1 and RR=1.2; 95% CI=0.9–1.4, respectively), compared with no antibiotic use. Antibiotic use for 21 days or more was associated with a 20% (95% CI=1.0, 1.3) increased tooth loss rate, compared with no antibiotic use. A linear trend of increased tooth loss rate was observed ( $P<0.05$ ). The adjusted rate for 21+days on antibiotics was 1.5 teeth per 100 persons (95% CI=0.3–2.7), which was higher than the adjusted rate for no antibiotic use. The RRs and differences in tooth loss rates associated with diabetes, current smoking, caries treatment, periodontal status, and tooth loss during baseline were higher than those associated with antibiotic use.

High use of preventive dental treatment was associated with decreased tooth loss.

Before adjustment, tetracyclines were associated with a 20% reduced tooth loss rate (95% CI=0.6–1.0). After adjustment for potential confounding factors, penicillin, metronidazole, tetracyclines, and macrolides used during the first 3 years were not associated with subsequent tooth loss. Relative to no clindamycin use, 1–13 days on clindamycin was associated with a 70% increased risk of tooth loss (95% CI=1.3, 2.2), and a linear trend for increased tooth loss rate was observed ( $P<0.05$ ).

The utility of tooth loss as an outcome in many studies is undermined by the fact that the “hopeless” teeth, whose response to treatment could be a sensitive indicator of an effective treatment, have often been extracted prior to any treatment. This means that secondary, or surrogate, clinical outcomes are used to evaluate the success of treatment in these studies. The most widely used surrogate outcomes are reduction in PPD and gain in attachment, although as many as 153 surrogate end points have been reported in the periodontal literature [81, 102].

### **3.9.5 The Prescribed Antibiotics and Their Dosage and Duration of Administration**

The creativity of the dental community in devising dosages is not to be underestimated, judging from the responses to the American Academy of Periodontology questionnaire, where over 300 different antibiotic regimens were used following periodontal surgery [102]. Even among the research investigations, it is difficult to compare between studies because of the different dosages used [102]. Antibiotic dosages varied among the different studies, especially for metronidazole (Table 3.10). The drug dosage was evaluated by calculating the total dosage, also taking into account the duration of the prescription and the number of cycles [78].

Addressing this topic, van Winkelhoff et al. [210] discussed the results of the study performed by Palmer et al. [146] on the clinical effects of initial periodontal treatment with or without application of local or systemic antibiotics, where the authors concluded that adjunctive systemic metronidazole did not result in an

extra clinical effect in comparison to scaling alone. This conclusion was based on comparison of clinical data in a smoking as well as in a nonsmoking patient group.

van Winkelhoff et al. [210] considered that the authors have used a systemic metronidazole dose which is much too low for adult patients with a severe, chronic bacterial infection (200 mg, 3x, 7 days). For adult patients, the amount of metronidazole needed for an effective concentration in body fluids amounts to 20–25 mg/kg body weight. There is also considerable difference in serum concentration between a 200- and a 500-mg dose of metronidazole; the peak concentrations of metronidazole in serum after a single given dose are for 200 mg: 4.8 µg/mL, for 250 mg: 6.2 µg/mL, and for 500 mg: 13.1 µg/mL.

However, in response, Palmer et al. [146] proposed that an alternative explanation for the lack of efficacy of metronidazole in their study is that the nonsurgical treatment was sufficiently effective such that any response to the antimicrobial was clinically insignificant. It was suggested that in the majority of cases of adult CP seen in practice, the use of metronidazole is not indicated, and that conventional nonsurgical treatment is effective.

If antimicrobial agents are to be used in the treatment of periodontal infections, their efficacy will be dependent upon patient compliance. This is an important issue when the results of treatment are evaluated, because an unsatisfactory result could indicate either an ineffective agent or a noncompliant patient. An effective agent that is not used by the patient is of little value [102]. As reviewed by Herrera et al. [80], none of the papers carried out a supervised antibiotic intake, although a few pointed out some method of assessment of the compliance, mainly through capsule use evaluation or a drug diary. Data from four studies reported a compliance ranging between 92% and 100% [80].

### **3.9.6 Characteristics of the Interventions**

As summarized by Herrera et al. [78], the relation between the debridement and the antibiotic usage can be evaluated as follows:

1. With debridement (coincidence of both antibiotic and debridement, both starting the same day and lasting equally).
2. With debridement plus additional time of drug intake (similar to the previous, but the antibiotic lasted longer than debridement).
3. Immediately after debridement (antibiotic intake starts after the last session of debridement).
4. With new debridement (antibiotic was given when a subsequent debridement was performed, not after initial debridement).

To optimize the effect of an antimicrobial agent in the periodontal pocket, one could increase the dosage, as occurs with the local-release delivery vehicles, or decrease the numbers of bacteria on the tooth surface. Both approaches would decrease the ratio of bacteria to antimicrobial agent. If an agent is taken that delivers 5 µg/day to the pocket, then the microbe-to-agent ratio is 20,000,000:1 (100,000,000 divided by 5). If we assume that scaling of the teeth will reduce bacterial levels in a pocket by 90%, i.e., from 100,000,000 to 10,000,000, and the same 5 µg of agent is delivered to the pocket, then the microbe-to-antimicrobial agent ratio is reduced to 2,000,000:1. If the debridement is thorough, such as would occur with root planning, then the bacterial load may be reduced by 99%, giving a microbe-to-agent ratio of 200,000:1. Clearly, after the teeth are debrided, the antimicrobial agent should encounter fewer bacteria in the pocket, and this phenomenon might explain the success of those double-blind studies in which the antimicrobial agent was given after debridement [102].

In the vast majority of studies, antibiotics were prescribed more often during and immediately after SRP, although in two protocols a delay of 4–6 weeks occurred. SRP was usually full-mouth (excepting split-mouth studies), although different treatment protocols were described in different studies: as more than one full-mouth SRP, as a previous session of scaling before root planning, or a renewed localized debridement immediately before antibiotic intake. In most articles the experience and skill of the operator was not reported. A dental hygienist was the most common operator, although a few studies used a periodontist or dental students. The time used in the mechanical therapy was reported in some papers, ranging between 30 min and 8 h, in 1–6 sessions, with an interval of 2 days to 2 weeks. The method used for scaling was seldom mentioned; some studies used only ultrasonic devices, while others combined curettes and ultrasonics (as reviewed by Herrera et al. [80]; Herrera et al. [78]).

### 3.9.7 The Study Design

As can been seen in Table 3.11, in the studies that evaluated the effects of systemic antibiotics in periodontal therapy there were both randomized clinical trials (RCTs) and two controlled clinical trials (CCTs), using/or not placebo, and in most of the cases there were more than one test group. As Herrera et al. [80] revealed, several studies presented results in different articles using the same population: Joyston-Bechal et al. [83] and Joyston-Bechal et al. [84]; Listgarten et al. [97] and Hellden et al. [76]; Palmer et al. [145]; McCulloch et al. [119] and Kulkarni et al. [92]; Reinhardt et al. [164], Golub et al. [58] and Walker et al. [216].

The length of the follow-up in these studies ranged between 22 weeks and 13 years, although most of the studies presented “short-term” results (from 22 weeks to 9 months) (Table 3.11).

Different antimicrobials have been evaluated: metronidazole (MET alone, or in combination with spiramycin (SPIR/MET) or amoxicillin (A/M); spiramycin (SPI); amoxicillin with clavulanic acid (AMOXI/CLAV); tetracycline chlorhidrate (TETt); clindamycin (CLIN); and doxycycline, both as an antimicrobial (Dox) and as low-dose, long-term, anticollagenase drug (SDD) [78, 80].

### 3.10 What Dosage to Use?

As was reviewed in the Position Papers by the American Academy of Periodontology [191], relatively few studies have been performed regarding which antibiotics should be selected for aggressive periodontitis patients in whom the subgingival microbiota have been characterized through microbiological testing. In addition, the optimal dose of antibiotics remains unclear since most current antibiotic regimens are empirically developed rather than through systematic research [49, 144].

Microbial analysis can be used to determine the specific antimicrobial susceptibility pattern of the suspected pathogens, can help to choose the appropriate antibiotics, and may be followed up with additional testing to verify the elimination or suppression of the putative pathogens. For some clinicians, microbial

analysis may be reserved for cases that are refractory to an initial course of antimicrobial therapy. Common antibiotic therapies for the treatment of periodontitis include metronidazole, 500 mg, three times a day for 8 days; clindamycin, 300 mg, three times a day for 8 days; doxycycline or minocycline, 100–200 mg, every day for 21 days; ciprofloxacin, 500 mg, twice a day for 8 days; azithromycin, 500 mg, every day for 4–7 days; metronidazole and amoxicillin, 250 mg of each drug, three times a day for 8 days; and metronidazole and ciprofloxacin, 500 mg of each drug, twice a day for 8 days [173, 190].

There is not enough scientific evidence in the literature to provide a unique treatment regime for periodontal abscesses. Three therapeutic approaches have been discussed, including drainage and debridement; systemic antibiotics with or without other treatments; and periodontal surgery procedures [79]. For adult patients with acute periodontal abscesses, an antibiotic regimen as an adjunct to incision and drainage is amoxicillin (1 g loading dose followed by 500 mg, three times a day for 3 days), with patient follow-up re-evaluation. For patients with allergies to β-lactam drugs, antibiotic regimens include azithromycin (1 g loading dose followed by 500 mg, every day for 2 days) or clindamycin (600 mg loading dose followed by 300 mg, four times a day for 3 days) [173, 190].

### 3.11 Considering Systemic Antibiotics as Monotherapy in the Treatment of Periodontal Disease?

One question that arises regarding periodontal therapy is: “Can systemic antimicrobials be efficacious if the biofilm is not disrupted?” Analyzing four papers describing metronidazole therapy [36, 95] and the combination therapy of amoxicillin and metronidazole [104, 225] either alone or adjunctive to conventional therapy (supra- and subgingival debridement), three meta-analyses performed (one for adjunctive therapy to SRP, another to surgery, and a third as monotherapy), and the use as monotherapy was found to be the only one not reaching significant results (mean effect of 0.849 mm,  $P=0.083$ ); therefore, the conclusion in the consensus report was that “there was insufficient evidence to support the use of systemic antibiotics as a monotherapy in

periodontitis patients” [78]. Furthermore, Topoll et al. [201] reported the development of multiple periodontal abscesses in patients with advanced periodontal disease who had been prescribed systemic antibiotic therapy without subgingival debridement. The patients had received broad-spectrum oral antibiotics (penicillin and tetracycline) 1–3 weeks prior to the development of abscesses. It was concluded that in patients with advanced periodontal disease, systemic antibiotic therapy without subgingival debridement might change the composition of the subgingival microbiota, resulting in multiple periodontal abscesses [77].

The scientific periodontal reviews performed on the topic [191, 71] clearly suggest that the use of systemic antimicrobials as monotherapy in the treatment of periodontitis is not recommended [78].

Lopez et al. [105] challenged this approach, based on two factors: (1) the recognition that systemically administered antibiotics can have a beneficial effect on clinical parameters of periodontal diseases and may have similar beneficial effects on the subgingival microbiota; and (2) there is a need to find cost-effective measures to control periodontal infections in populations where access to periodontal care is limited. They performed a pilot clinical trial to evaluate the changes in the levels and proportions of 40 bacterial species in the subgingival microbiota of previously untreated chronic periodontitis patients after the administration of metronidazole plus amoxicillin as the only therapy. A group of patients with chronic periodontitis treated with scaling and root planning alone were used as control.

Before the onset of the study, each subject received a supragingival scaling (SGS) to remove gross calculus to allow periodontal probing. Subjects in both groups received instructions from an experienced periodontist in self-performed oral hygiene measures (IOH): toothbrushing three times a day using the modified Charter’s technique with a soft toothbrush and regular toothpaste with fluoride. SGS and SRP were also administered by the same experienced periodontist using an ultrasonic scaler (Cavitron SPS, Denstply Detrey, GmbH, Germany) with insert FSI-10. Manual Gracey curettes (Gracey Hu Friedy Instruments, Chicago, IL, USA) were also used to perform root planning, as needed. The test group received only SGS for all teeth in two sessions of 45 min each, 3 days apart. Each patient of the test group received 21 tablets of metronidazole 250 mg and 21 tablets of amoxicillin 500 mg, and was asked to take one tablet of each medication every 8 h

for 7 days. The control subjects received SGS and SRP under local anesthesia. Clinical measurements including sites with plaque, BOP, PPD, and AL were made at baseline and at 3, 6, 9, and 12 months. Subgingival plaque samples were taken from the mesio-buccal aspect of all teeth, except third molars, in each subject at baseline and at the 3-, 6-, 9-, and 12-month monitoring visits. Counts of 40 subgingival species were determined in each plaque sample using a modification of the checkerboard DNA–DNA hybridization technique.

There was a statistically significant reduction over time in the mean PPD, the relative AL, the percentage of sites with BOP, with PPD 4–6 mm and ≥6 mm, and the percentage of sites with plaque for both treatment groups. The mean PPD values ( $\pm$ SEM) at baseline and at 3, 6, 9, and 12 months for the M/A group were  $2.80 \pm 0.45$ ,  $2.14 \pm 0.06$ ,  $2.20 \pm 0.06$ ,  $2.06 \pm 0.04$ , and  $1.95 \pm 0.05$  ( $P < 0.001$ ), and for the SRP group were  $2.38 \pm 0.41$ ,  $1.90 \pm 0.07$ ,  $1.93 \pm 0.10$ ,  $2.01 \pm 0.05$ , and  $1.95 \pm 0.10$  ( $P < 0.001$ ). The percentage of sites with PPD 4–6 mm and with PPD  $\geq 6$  mm were significantly reduced at 3 months after treatment in the SRP and M/A groups ( $P < 0.001$ ) and continued at lowered levels between 3 and 12 months.

Corresponding mean relative AL values for the M/A group were  $10.07 \pm 0.39$ ,  $10.06 \pm 0.38$ ,  $9.96 \pm 0.37$ ,  $10.7 \pm 0.36$ , and  $9.77 \pm 0.34$  ( $P < 0.001$ ), and for the SRP group  $9.94 \pm 0.28$ ,  $9.88 \pm 0.25$ ,  $9.80 \pm 0.25$ ,  $9.89 \pm 0.22$ , and  $9.77 \pm 0.26$  ( $P < 0.001$ ). Twelve months after therapy, subjects in the M/A group showed an overall mean gain of attachment of 0.3 mm, and subjects in the SRP group of 0.17 mm. This difference compared with baseline was statistically significant ( $P < 0.001$ ) for both groups.

There was also a significant decrease in the percentage of sites that bled on probing in both treatment groups. The mean values at time points from baseline to 12 months were  $40.54 \pm 5.52$ ,  $16.60 \pm 3.15$ ,  $15.00 \pm 1.44$ ,  $14.75 \pm 1.62$ , and  $14.0 \pm 1.35$  ( $P < 0.001$ ) for M/A-treated subjects, and  $38.54 \pm 5.1$ ,  $17.90 \pm 3.41$ ,  $15.41 \pm 1.51$ ,  $17.96 \pm 2.57$ , and  $18.99 \pm 2.84$  ( $P < 0.001$ ) for SRP-treated subjects. There were no significant differences between the two treatment groups at the different time points in any of the clinical parameters, except for mean PPD at baseline and at 3 and 6 months and percentage of sites with PPD 4–6 mm at baseline and 3 months.

Mean total DNA probe counts and counts of the majority of the 40 test species were significantly reduced over time in both groups, with no significant

differences detected at any time point between groups. At 12 months many of the species were still present at significantly lowered levels compared with their baseline counts in both groups.

In summary, the changes in clinical and microbiological parameters were similar after receiving systemically administered M/A as the sole therapy or after receiving SRP alone only [105].

The study raised controversy on the use of antimicrobials for the treatment of periodontitis and, in particular, used as monotherapy [56, 127, 221]. All comments stated that the risk of using antimicrobials (systemic side effects, increase in antimicrobial resistance) should lead to restriction in their use in periodontitis in certain patients and certain conditions. In addition, their use should be combined with debridement, based on the knowledge of the biofilm characteristics and the evidence available from clinical studies [78].

1. The subgingival biofilm needs to be mechanically removed or disturbed in order for systemic antibiotics to be effective. As this structure protects the bacteria from the immune system of the host as well as from antimicrobial agents, a biofilm is a difficult therapeutic target. In the field of periodontal microbiology, it has been demonstrated recently that several antibiotics need to be much higher concentrated to reach the MIC in a biofilm compared with microorganisms grown in a planktonic culture [48]. To date, the only predictable way to disturb the dental biofilm is by using mechanical means [221].
2. Antibiotic treatment as a sole or adjunctive therapy has been suggested as an effective treatment from a strictly economic point of view. However, the following aspects must be recognized: (a) the problem of patient compliance, (b) the need for mechanical biofilm disruption as initial as well as repeated maintenance treatment, and (c) the increasing antibiotic resistance with a further need for expensive development of more effective antimicrobial drugs. Therefore, antibiotic treatment as a sole therapy does not seem to be a low-cost approach to reach long-term periodontal health [221].
3. In the observed subjects, the mean PPD and the percentage of sites with PPD 4–6 mm and PPD $\geq$ 6 mm was quite low and, therefore, the periodontitis may be classified as slight rather than moderate to advanced disease severity. SGS has been performed by an experienced periodontist using an ultrasonic scaler on all

teeth in two sessions of 45 min each. It can be assumed that especially in sites with slight and moderate PPDs, a subgingival biofilm will also be disturbed by SGS. Therefore, the treatment performed might be an adjunctive antibiotic therapy instead of a sole antibiotic therapy at least in these sites. Moreover, there are significant differences between the groups concerning percentages of sites with PPD 4–6 mm and mean PPD. Both are higher and respectively deeper in the test group than in the control. The fact that PPD reduction is generally more pronounced in deeper pockets following mechanical periodontal treatment favors the outcome in the test group [221].

The legitimacy of placing SRP in the center of periodontal clinical activity is based on extensive clinical research. Periodontal diseases can indeed be treated successfully with mechanical means, and results can be maintained by regular mechanical cleansing of teeth [127]. This treatment can be classified as the “gold standard” of periodontal care and has been demonstrated to be successful in order to maintain periodontal health for more than 20 years [9]. Therefore, any treatment alternative used in daily dental practise should be superior to the gold standard and well proven in long-term clinical trials. Above all, this is true for those treatment modalities which present certain side effects and possibly affect the efficacy when used. Even though mechanical treatment does not predictably eliminate all bacteria from diseased sites completely, a precautionary, restrictive attitude towards using antibiotics has been recommended, basically to limit the development of microbial antibiotic resistance in general, and to avoid the risk of unwanted systemic effects of antibiotics [127].

### 3.12 Implications of Systemic Antibiotics as an Adjunct to Nonsurgical and Surgical Therapy

Because subgingival bacteria are organized in biofilms, in principle, they are less susceptible to antimicrobials, unless there is a previous disruption by mechanical debridement and in this manner the antimicrobial results should be improved. As summarized by Herrera et al. [78], different explanations have been suggested to

explain the resistance of biofilms against antimicrobial agents: (1) the biofilm extracellular matrix [109]; (2) different physiological phases of the microorganisms within a biofilm [6, 44, 222]; (3) horizontal gene transfer [167, 168]; and (4) molecular mechanism of communication among bacterial cells, quorum sensing [166].

The current consensus that mechanical instrumentation must always precede antimicrobial therapy is founded on two arguments. First, we should quantitatively reduce the large mass of bacteria, which otherwise may inhibit or degrade the antimicrobial agent. Insufficient concentrations of the active agent may again favor the emergence of resistant strains. Second, we should mechanically disrupt the structured bacterial aggregates that can protect the bacteria from the agent [127].

As a consequence it is recommended that if systemic antimicrobials are indicated as part of periodontal therapy, they should be adjunctive to **mechanical debridement**. There is no direct evidence to indicate a specific protocol for the use of adjunctive systemic antimicrobials with nonsurgical mechanical debridement. However, indirect evidence suggests that antibiotic intake should start on the day of debridement completion; debridement should be completed within a short time (preferably 1 week) and with an adequate quality, because these may help to improve the results [70].

**Periodontal surgery**, as any other surgery in the oral cavity, may be associated with the risk of developing postoperative complications, such as infection (suppuration, pain, swelling, redness, bacteraemia). However, whether the administration of systemic antimicrobials diminishes this risk is still a matter of controversy and this adjunctive use of systemic antibiotics with different surgical procedures is based more on empiricism than on scientific data [70].

Several RCTs have evaluated antimicrobials as adjuncts to the surgical treatment of periodontitis, aiming to enhance both clinical and microbiological outcomes [41, 69, 70, 87, 93, 110, 87, 147, 194]. It is suggested that there might be a beneficial effect with antimicrobial use, although often these benefits were not clearly better than those of the control groups, either because the differences did not reach the level of statistical significance or the magnitudes were of little clinical relevance. Overall, there is limited evidence to support the use of systemic antibiotics as an adjunct to periodontal surgery [70] (Table 3.12).

Haffajee et al. [70] concluded that unless there is a medical indication, there is no justification for using prophylactic antibiotic in periodontal surgery. An

indiscriminate and prolonged use of antibiotics may result in a higher rate of infection. In addition, the risks involved with the use of systemic antibiotics (adverse events etc.) must always be considered against the limited benefits [70].

A meta-analysis by Haffajee et al. [71] reviewed three studies and included four comparisons [69, 93, 147]. They reported that systemically administered antimicrobial agents provide a significant clinical benefit in terms of mean CAL gain (weighted mean 0.609,  $P=0.007$ ) [70].

There are very few CCTs assessing the need and the long-term efficacy of the adjunctive use of systemic antibiotics in **periodontal regenerative surgical procedures** [42, 43, 103, 126, 139, 181, 211] and the results are controversial [70] (Table 3.13).

### 3.13 Recommendations for Treating Periodontitis with Antibiotics

Antibiotic therapy is usually reserved for patients having continued periodontal breakdown after conventional mechanical treatment. However, some patient categories with recognized increased risk for periodontal breakdown, such as progressive adolescent periodontitis and other types of early onset periodontitis, may be treated with systemic antibiotics as an adjunct to initial mechanical therapy. It is especially important to consider antibiotic therapy in the treatment of aggressive periodontitis, which often involves several specific pathogens with the potential to invade pocket epithelium and connective tissue. In patients with chronic periodontitis, the utility of systemic antibiotics is not as clear. Since most clinical studies of antibiotic efficacy have been conducted in patients with chronic periodontitis, who respond well to scaling and root planning, they may have underestimated the value of adjunctive systemic antibiotics in aggressive types of periodontitis [189].

In general, adult periodontitis can and should be treated without systemic antibiotics. In patients with chronic periodontitis, the utility of systemic antibiotics is not as clear. Since most clinical studies of antibiotic efficacy have been conducted in patients with chronic periodontitis, who respond well to scaling and root planning, they may have underestimated the value of adjunctive systemic antibiotics in aggressive types of periodontitis [189].

**Table 3.12** RCT assessing systemic antibiotic therapy as adjunct to periodontal surgery

| Study                | Type of study | No. Patients | Periodontal condition                               | Study period | Periodontal treatment   | Surgical procedure   | Outcome  |
|----------------------|---------------|--------------|---|--------------|---|----------------------|--|
| Palmer et al. [147]  | RCT           | 38           | Localized and generalized early onset periodontitis | 12 months    | 1. OHI+SRP+TET (250, 4x, 14d) followed by SURG+CHX+TET (250, 4x, 14d)<br>2. OHI+SRP+placebo followed by SURG+CHX+placebo  | Modified Widman flap | In the test group, 58% of the originally affected teeth required surgery compared to 75% in the control group. Surgery produced further reductions in mean PPD but no further gains in CAL. There were no further statistically significant differences between test and control groups for any of the clinical measures, although the tetracycline group appeared to maintain an advantage. In summary, systemically administered tetracycline is a useful adjunct in the management of early onset periodontitis, particularly in nonsurgical treatment.   |
| Kunihira et al. [93] | RCT           | 16           | Localized juvenile periodontitis                    | 62 weeks     | 1. OHI+SRP followed by SURG+PEN (250, 4x, 10d) then SPT<br>2. OHI+SRP followed by SURG+placebo then SPT   | Open curettage       | In both groups there was a significant decrease in plaque scores, gingival inflammation, gingival bleeding, and probable depths for all sites and for affected sites. Similarly there was a significant increase in AL and radiographic bone height, and a total elimination of suppuration. The favorable changes were apparent at the first postsurgical recall (week 26 of the study) and remained essentially the same through week 62. The magnitude of change in these parameters was similar to that reported by others for treatment regimes including tetracycline therapy. In summary, however, there were no differences in any parameters between the placebo and penicillin groups.   |
| Haffajee et al. [69] | RCT           | 98           | Severe periodontal disease                          | 10 months    | 1. SRP+SURG+CHX+TET (250, 4x, 30d) then SPT<br>2. 1. SRP+SURG+CHX+AMOX/CLAV (250, 3x, 30d) then SPT<br>3. 1. SRP+SURG+CHX+IBU then SPT<br>4. 1. SRP+SURG+CHX+placebo then SPT | Modified Widman flap | Subjects receiving antibiotics exhibited significantly more AL “gain” ( $0.57 \pm 0.15$ mm) than subjects receiving either ibuprofen or a placebo ( $0.02 \pm 0.10$ mm). The differences between AMO/CLAV and TET groups were not significant, nor were the differences between ibuprofen and placebo. Subjects receiving systemically administered antibiotics had a greater decrease in the number of sites colonized by <i>P. gingivalis</i> , <i>B. forsythus</i> , <i>P. intermedia</i> , and <i>P. micros</i> post therapy than subjects not receiving antibiotics. In summary, the results of this investigation indicate that adjunctive systemic antibiotics increase periodontal attachment “gain” and decrease the levels of some suspected periodontal pathogens in subjects with evidence of current disease progression. |

(continued)

**Table 3.12** (continued)

| Study                    | Type of study | No. Patients | Periodontal condition                                 | Study period | Periodontal treatment   | Surgical procedure                                   | Outcome   |
|--------------------------|---------------|--------------|---|--------------|---|--|---|
| Kleinfielder et al. [87] | CCT           | 35           | Periodontitis with <i>A. actinomycetemcomitans</i>    | 12 months    | 1. SURG+OFLO (200, 2x, 5d) then SPT (N=25)<br>2. SURG then SPT (N=10)                         | Open flap surgery                                    | At 3 and 12 months following therapy mean PPD at monitored sites in the test group changed from 6.8±1.3 mm to 3.6±1.0 mm, 3.8±1.1 mm and CAL from 7.5±1.4 mm to 5.4±1.4 mm, 5.5±1.3 mm. In the control group PPD changed from 6.5±0.7 mm to 4.0±1.7 mm, 4.1±1.6 mm and CAL from 7.5±1.0 mm to 6.3±1.7 mm, 6.4±1.8 mm ( $P<0.05$ ). 3 and 12 months following adjunctive systemic ofloxacin therapy, <i>Aa</i> was suppressed below detectable levels in 22 of 22, test patients, whereas <i>Aa</i> could not be recovered in only two of the ten controls ( $P<0.0001$ ). In summary, systemic ofloxacin as adjunct to open flap surgery is able to suppress <i>A.a</i> below detectable level in patients harboring this organism at baseline.   |
| Dastoor et al. [41]      | RCT           | 30           | Moderate to advanced chronic periodontitis in smokers | 6 months     | 1. SURG+CHX + Ibuprofen+AZ (500, 1x, 3d) (N=15)<br>2. SURG+CHX + Ibuprofen 1 + placebo (N=15) | Apically positioned flap with osseous reconstructing | Surgical treatment of moderate (PPD =4–6 mm) and deep (PPD > 6 mm) pockets significantly improved clinical parameters of treated and untreated teeth (CAL gain, PPD reduction, and reduction of BOP). The additional use of AZM did not enhance this improvement nor did it promote reduction of cross-linked telopeptide of type I collagen levels in GCF. Compared to the control group, the test group had significantly better WHI scores at 1 month, significantly less GI at 2 weeks, and sustained reductions of red-complex bacteria with trypsin-like enzyme activity at 3 months. For nonsurgery teeth, only the test group showed significant gains in overall CAL compared to baseline. In summary, the findings of this pilot study demonstrated that in heavy smokers, adjunctive systemic AZM in combination with pocket reduction surgery did not significantly enhance PPD reduction or CAL gain. However, the clinical value of adjunctive AZM may be appreciated by more rapid wound healing, less short-term gingival inflammation, and sustained reductions of periopathogenic bacteria. |

|                       |     |                                    |                                     |          |  |                      |  |
|-----------------------|-----|------------------------------------|-------------------------------------|----------|--|----------------------|--|
| Mahmood & Dolby [110] | RCT | 15                                 | Moderate and advanced periodontitis | 6 months | 1. SURG+MET (200, 3 x, 7d)<br>2. SURG+placebo  | Modified Widman flap | PPDs and SBIs were reduced significantly at all stages, in both groups. Probing attachment levels increased at 7 days, to significant levels only in the MET group, subsequently PALs decreased in both groups with no significant differences between the groups. Although the differential bacterial count altered markedly in both groups at all times, only the straight rod count at 1 month was significantly ( $P$ less than 0.05) lower in the MET group. In summary, MET with surgery did not exert a significantly greater beneficial effect than placebo with surgery.  |
| Söder et al. [194]    | RCT | 64 (37 smokers and 27 non-smokers) | Severe periodontal disease          | 5 years  | 1. SRP+MET (400, 3x, 7d) ( $N=32$ )<br>2. SRP+placebo<br><br>Regular follow-up examinations at 6-month intervals for oral hygiene and SRP. SURG when PPD increased $\geq 2$ mm between 2 visits ( $N=32$ ) | Modified Widman flap | Nonsmoking patients who required only nonsurgical therapy in the intervention group showed statistically significant improvement in the clinical parameters after 5 years (PPD at baseline $2.8 \pm 0.2$ , at 7 years $2.1 \pm 0.4$ ; CAL at baseline $3.8 \pm 0.4$ , at 5 years $3.1 \pm 0.4$ ; bone height percentage at baseline $79.7 \pm 3.2$ , at 5 years $82.3 \pm 2.7$ ). Patients with complete healing, defined as the absence of inflamed sites $\geq 5$ mm, after 5 years were found only in the intervention group (% of teeth with pockets $\geq 5$ mm in nonsmokers with intervention: at baseline $25.9 \pm 12.9$ vs at 5 years $4.7 \pm 8.4$ ; no sites with pockets $\geq 5$ mm in nonsmokers with intervention at baseline: $9.6 \pm 5.6$ vs at 5 years $1.6 \pm 2.8$ , $P < 0.01$ ). Smokers responded less favorably to periodontal therapy than nonsmokers. The number of patients infected with <i>Aa</i> , <i>Pg</i> , <i>Pi</i> , and spirochetes decreased during the study. Most patients who harbored spirochetes at the end of the study had these microorganisms at the beginning. The patients considered healthy after 5 years were the same patients found to be healthy after 6 months. In summary, decisive factors in the sustained long-term improvement of patients who respond satisfactorily to treatment are probably initial scaling and root planning; a brief course of metronidazole; and regular follow-up examinations at 6-month intervals for oral hygiene and SRP. |

**Table 3.13** RCT assessing systemic antibiotics as adjunct to periodontal regenerative therapy

| Study                 | Type of study     | No. patients/defects | Periodontal condition | Study period | Periodontal treatment  | Outcome  |
|-----------------------|-------------------|----------------------|-----------------------|--------------|--|--|
| Mombelli et al. [126] | RCT (split-mouth) | 10 patients          | Class II furcation    | 50 weeks     | 1. GTR (ePTFE) + Ornidazole (1000, 1x, 10d)<br>2. Ornidazole (1000, 1x, 10d)<br>3. GTR (ePTFE) + placebo<br>4. Untreated | More horizontal attachment gain and increase in bone density was obtained in patients receiving the active drug than in patients receiving the placebo. Treatment with membrane plus ornidazole resulted in 0.7 mm mean recession and -1.2 mm mean decrease in horizontal periodontal probing depth. Sites treated with membranes generally tended to be positive for 15 target microorganisms more often than sites treated without a membrane. This was particularly evident for <i>Fusobacterium</i> , <i>Prevotella intermedia</i> , and <i>Actinomyces odontolyticus</i> . While GTR-treated sites were often already positive upon removal of the membrane, re-emergence of target organisms seemed to be more delayed in the conventionally treated sites.  |
| Demolon et al. [43]   | CCT               | 15                   | Class II furcation    | 4 weeks      | 1. GTR + CHX (N=7)<br>2. GTR + CHX + Augmentin (AMO 250, CLAV 125, 3x, 10d, 1 h before surgery) (N=8)                    | At baseline no parameter showed statistical differences between groups or sites. At week 1 significantly greater levels of <i>P. intermedia</i> type I ( $P < 0.05$ ) and <i>Fusobacterium nucleatum</i> ( $P < 0.01$ ) were found in group 1. At week 4, paper-point samples from test sites ( $P < 0.05$ ) and e-PTFE materials ( $P < 0.001$ ) showed significantly higher presence of <i>Bacteroides forsythus</i> in group 1. No significant microbial changes were found for control sites over time or between groups. The total bacterial load at test sites over time increased similarly for patients administered or not administered the antibiotic. Clinical signs of inflammation were significantly greater in group 1 and associated with the presence of <i>B. forsythus</i> ( $P < 0.01$ ).  |
| Demolon et al. [42]   | CCT               | 15                   | Class II furcation    | 1 year       | 1. SRP + GTR + CHX (N=7)<br>2. SRP + GTR + CHX + Augmentin (AMO 250, CLAV 125, 3x, 10d, 1 h before surgery) (N=8)        | After 1 year, the reduction in mean periodontal probing depth of the furcation invasions was $2.0 \pm 1.2$ mm for group 1 and $1.8 \pm 1.1$ mm for group 2. An overall gain of 0.8 mm of clinical attachment was found. Twenty-two of the 24 sites were re-entered. Wide individual variations were found but the changes between pretreatment and 1-year data for any of six linear measurements of hard tissue landmarks did not differ between groups or between pretreatment and re-entry. A combination of an overall ABL of 0.4 mm at the crest and 0.3 mm gain of bone at the bottom of the furcation defects was found. Volumetric analysis indicated an average 32% bone fill for both groups, ranging from a decrease in defect volume by 84% (gain) to an increase of the size of the furcation invasion by 66% (loss). A decrease in defect volume $> 30\%$ was found at seven sites from each group. In summary, the antibiotic may have controlled initial inflammation, but 12 months later it had no direct effect on bone regeneration or soft tissue attachment. |

|                      |     |             |  |          |  |  |
|----------------------|-----|-------------|--|----------|--|--|
| Nowzari et al. [139] | RCT | 18 patients | 2- to 3-wall periodontal bony defects        | 6 months | 1. OHI+SRP followed by GTR (ePTFE)+AUG (AMOX/CLAV 500/125, 3x, 8d, 1 h prior membrane) followed by SPT<br>2. OHI+SRP followed by GTR (ePTFE) followed by SPT   | At baseline, no microbial or clinical parameter showed statistical differences between groups. At 6 months, the Augmentin group demonstrated a significantly higher ( $P=0.032$ ) mean CAL gain (36.5%) than the 9 control patients (22.4% of potential gain). At the time of removal, membranes in the Augmentin group showed significantly fewer organisms than membranes in the control group ( $52.2 \times 10^6$ versus $488.6 \times 10^6$ ). Sites free of pathogens on the membrane surface toward the tooth gained the most CAL, even in the presence of various pathogens on the gingiva-facing membrane surface.  |
| Loos et al. [103]    | RCT | 25          | Intraosseous periodontal defects $\geq 6$ mm | 1 year   | 1. M+A+ : CHX+GTR (polylactic acid)+A/M (375/350 mg, 3x, 8d, 4 days before the surgical procedure)<br>2. M+A- : CHX+GTR (polylactic acid)<br>3. M-A+ : CHX+A/M (375/350 mg, 3x, 8d, 4 days before the surgical procedure)<br>4. M-A- : CHX | Reduction in PPD at 12 months postoperatively varied between 2.54 and 3.06 mm in the four treatment modalities, but overall no main effect of MEM or AB was found. Gains in CAL at 12 months postoperatively varied between 0.56 and 1.96 mm for the four treatments. In the overall analysis for PAL, no main effect of MEM or AB was found. Gains in PBL and percentage defect fill at 12 months in the treatment groups were: Group 1: $2.09 \pm 0.35$ , $43 \pm 9$ ; Group 2: $1.90 \pm 0.38$ , $33 \pm 9$ ; Group 3: $1.39 \pm 0.35$ , $29 \pm 9$ ; Group 4: $1.53 \pm 0.38$ , $44 \pm 9$ . Again, overall, no main effects of MEM or AB were found for PBL. Exploratory statistical analyses indicated that smoking and not MEM or AB is a determining factor for gain in PBL ( $P < 0.0009$ ). On the day of surgery in the AB+ group, the number of defects culture positive for <i>A. actinomycetemcomitans</i> , <i>P. gingivalis</i> , <i>P. intermedia</i> , and <i>B. forsythus</i> was substantially reduced. Among these AB+ subjects, the number of culture positive sites for <i>P. micros</i> and <i>F. nucleatum</i> was somewhat reduced (25–55% and 50–60%, respectively). In the AB- patients, no substantial reduction of the number of defects culture positive for <i>P. gingivalis</i> , <i>P. intermedia</i> , and <i>B. forsythus</i> was observed. The prevalence of several periodontal pathogens, on the day of surgery or postoperatively, and specific defect characteristics, were not determining factors for gain in CAL and PBL. In summary, neither the application of barrier membranes nor the use of systemic antibiotics showed an additional effect over control on tissue measurements in the treatment of intraosseous defects. |

(continued)

**Table 3.13** (continued)

| Study                | Type of study | No. patients/defects | Periodontal condition  | Study period | Periodontal treatment   | Outcome   |
|----------------------|---------------|----------------------|------------------------|--------------|---|---|
| Sculean et al. [180] | RCT           | 34                   | Deep intrabony defects | 1 year       | 1. EMD+CHX+A/M (AMO 375, 3x, 7d and MET 250, 3x, 7d, on the first day of surgery) followed by SPT ( $N=17$ )<br>2. EMD alone+CHX ( $N=17$ )   | The results have shown that in the EMD+A/M group the PPD decreased from $9.1 \pm 1.5$ mm to $4.5 \pm 1.1$ mm ( $P < 0.0001$ ) and the CAL changed from $11.0 \pm 1.6$ mm to $7.5 \pm 1.4$ mm ( $P < 0.0001$ ). In the EMD group the PPD decreased from $9.0 \pm 1.7$ mm to $4.3 \pm 1.7$ mm ( $P < 0.0001$ ) and the CAL changed from $10.6 \pm 1.6$ mm to $7.3 \pm 1.5$ mm ( $P < 0.0001$ ). There were no significant differences in any of the investigated parameters between the two groups. In summary, it can be concluded that the systemic administration of amoxicillin and metronidazole adjacent to the use of EMD for the surgical treatment of intrabony periodontal defects does not produce statistically superior PPD reduction and CAL gain when compared to treatment with EMD alone. Hence, the present results do not support the routine administration of amoxicillin and metronidazole following regenerative treatment with EMD. |
| Vest et al. [211]    | RCT           | 24                   | Class II furcation     | 9 months     | 1. Test group: GTR (polylactide bioabsorbable membrane)+DFDBA+antibiotics (from the first day of surgery ciprofloxacin 250, 2x, 7d +MET, 250, 3x, 7d followed by DOXY 50, 7d) ( $N=12$ )<br>2. Control group: GTR (polylactide membrane)+DFDBA ( $N=12$ ) | Mean open horizontal PPD reductions at 9 months were greater for the test than for the control group ( $2.92 \pm 1.78$ versus $2.50 \pm 1.62$ mm, $P > 0.05$ ). Fifty-eight percent of furcations in the test group demonstrated >50% vertical defect fill at 9 months compared to 67% in the GB group. There were no significant differences in mean open horizontal PPD reduction between smokers and nonsmokers in either the GBA or GB groups. Membrane exposure did not appear to affect regenerative healing in either the test or control groups. In summary, the administration of postsurgical antibiotics did not produce statistically superior osseous healing of Class II furcation defects.   |

DFDBA demineralized freeze-dried bone allograft

Periodontal abscesses and acute necrotizing ulcerative gingivitis (ANUG) lesions presenting systemic manifestations should be also considered for antibiotic therapy [74, 185].

Antibiotic therapy is indicated for periodontal abscesses with systemic manifestations (fever, malaise, lymphadenopathy). Antibiotics for the treatment of abscesses should be prescribed in conjunction with surgical incision and drainage [189]. Two adult regimens may be used with acute periodontal abscesses: amoxicillin: loading dose of 1.0 g followed by a maintenance dose of 500 mg, three times per day, for 3 days, followed by a patient evaluation to determine whether further antibiotic therapy or dosage adjustment is required; and in patients with allergy to  $\beta$ -lactam drugs, azithromycin: loading dose of 1.0 g on day 1, followed by 500 mg, four times a day on days 2 and 3, or clindamycin: loading dose of 600 mg on day 1, followed by 300 mg, four times a day for 3 days [144].

A practical approach to antibiotic therapy for patients with progressive adult periodontitis and early-onset periodontitis was proposed van Winkelhoff et al. [207] and Slots [189]:

1. Initial periodontal therapy should include thorough mechanical root debridement combined with surgical access if needed. Supplemental subgingivally applied broad-spectrum antiseptic agents may be used. Numerous periodontal abscesses were observed in untreated advanced periodontal diseases after systemic administration of antibiotics for non-oral infections [201].
2. One to 3 months after completion of the mechanical therapy, the clinical response is evaluated. A microbiological examination of the subgingival microbiota is required to determine the presence and the level of the remaining putative periodontal pathogens.
3. Antibiotics should be prescribed on the basis of the clinical need for further treatment, the microbiological findings, and the medical status and current medications of the patients. Short-term high-dose antibiotic regimens should be favored.
4. At 1–3 months after systemic antimicrobial therapy another microbiological test may be warranted to verify the subgingival elimination of target pathogens as a screen for possible superinfecting organisms. High levels of viridans streptococci and *Actinomyces* sp. are suggestive of periodontal health or minimal disease.

5. After resolution of the periodontal infection, the patient should be placed on an individually tailored maintenance care program. Good patient home plaque control after systemic antimicrobial therapy is essential for long-term treatment success. Recurrence of progressive disease may prompt additional microbiological testing and further therapy targeting the specific periodontal pathogens involved.
6. Screening for and eradication of exogenous pathogens (*A. actinomycetemcomitans* and *P. gingivalis*) in family members might be considered to prevent reinfection and possible recurrence of disease.

### 3.14 Final Considerations

The evidence available suggests that disadvantages and safety aspects of systemic antimicrobial use in the management of periodontal diseases significantly outweigh the benefits. Antimicrobial prescribing should be the exception rather than the rule and, in the majority of cases, only considered after conventional therapies have been unsuccessful [2]:

1. Systemic antimicrobials should always be used as adjuncts to mechanical therapies [2].
2. Systemic antimicrobials should only be used in acute periodontal conditions where drainage or debridement is impossible, there is local spread of the infection, or systemic upset has occurred [2].
3. Systemic antimicrobials are contraindicated as adjuncts to the treatment of chronic periodontal diseases where a satisfactory level of oral hygiene has not been achieved [2].
4. Systemic antimicrobials offer little if any adjunctive benefit to the treatment of periodontal disease in patients who smoke tobacco products [2].
5. The use of systemic antimicrobials in early-onset and rapidly progressive periodontal diseases should be considered either from the outset or when nonsurgical methods are preferred over surgical interventions. Even in these rare disease states a nonsurgical approach alone should be considered first [2].
6. The use of systemic antimicrobials may be considered for “refractory” and aggressive diseases [2].

7. Evidence remains insufficient to provide guidelines for antimicrobial use in patients who have concomitant disease predisposing them to periodontal disease, notably diabetes, and the decision remains empirical [2].
8. To date, evidence only supports the benefit of a very limited number of antimicrobials in the management of chronic periodontal diseases and, in particular, the combination of metronidazole and amoxicillin [2].
9. It is essential that the patient be fully informed of the pros and cons of adjunctive systemic antimicrobial therapy and, in particular, possible side effects of particular regimens and how these may be minimized or prevented, e.g., alcohol and metronidazole [2].
10. The practitioner must identify those individuals most at risk from the side effects of prescribed systemic antimicrobials, notably in respect of the medical history and concomitant medication relating to the patient [2].

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## The Topical Use of Antibiotics in Periodontal Pockets

The concept that local delivery of an antibiotic into the periodontal pocket achieves a greater, more potent concentration of drug than available with systemic delivery is very appealing. The amount of drug delivered often creates sulcular medication concentrations exceeding the equivalent of 1 mg/mL (1,000 µg/mL). This level is considered bactericidal for the majority of bacteria that exhibit resistance to systemically delivered concentrations. Equally important, local delivery of an antibiotic exhibits a negligible impact on the microflora residing in other regions of the body [110].

The periodontal pocket is a unique infection site. It is readily accessible to the clinician and can be individually monitored to determine treatment response. Microbiologically, the concept of treating only those sites that are deemed to be in need of treatment by mechanically removing subgingival plaque, then subsequently applying a locally delivered antimicrobial, appears ideal. In theory, mechanical debridement serves to disrupt and displace the biofilm. Locally administered antibiotics, at concentrations much greater than can be achieved systemically, aid in site-specific elimination of residual bacteria. Tetracycline, doxycycline, minocycline, and metronidazole have been individually incorporated into local continuous delivery devices and made commercially available to the practitioner [110].

### 4.1 Advantages and Disadvantages of Local Antimicrobial Agent Pocket Delivery

The topical use of antibiotics in the treatment of periodontal disease presents several advantages, but also

disadvantages. The local route of antibiotic administered can accomplish 100-fold greater therapeutic doses in subgingival sites than those possible by systemic therapy. Furthermore, local therapy enables utilization of antimicrobial agents which do not lend themselves to systemic administration. Also, professionally administrated topical therapy reduces problems with patient compliance. Local antibiotic therapy may be especially useful for women with a propensity for vaginal superinfections or for individuals displaying gastrointestinal or other side effects after systemic antibiotic therapy [90].

Disadvantages of local antimicrobial treatment of periodontitis include difficulty in placing therapeutic concentrations of the antimicrobial agents into deeper parts of periodontal pockets and furcation lesions. Personal application of antimicrobial agents by patients as a part of their home self-care procedures is frequently compromised by the patient's lack of adequate manual dexterity, limited understanding of periodontal anatomy, and poor compliance and performance with the recommended procedure. The task of professionally applying local antimicrobial agents in periodontitis patients with numerous advanced lesions distributed throughout the mouth is time-consuming and labor-intensive. Nonsustained subgingival drug delivery is limited by only brief exposure of the target microorganisms to the applied antimicrobial agent. Antimicrobial agents locally applied into periodontal pockets do not markedly affect periodontal pathogens residing within adjacent gingival connective tissues and on extra-pocket oral surfaces (tongue, tonsils, and buccal mucosa), which increases the risk of later reinfection and disease recurrence in treated areas [77].

## 4.2 Antibiotics for Topical Use in Periodontal Therapy

### 4.2.1 Tetracycline-HCl

The tetracyclines – tetracycline-HCl, doxycycline hydiate, and minocycline-HCl – are broad-spectrum antibiotics active against both gram-positive and gram-negative bacteria. Structurally, tetracyclines consist of four fused rings, hence the name tetracyclines (Fig. 4.1). Tetracycline derivatives, primarily doxycycline and minocycline, differ from the parent compound by minor alterations of chemical constituents attached to the basic ring structure. These minor alterations in the molecular structure make both doxycycline and minocycline more lipophilic than the parent compound, resulting in better adsorption following systemic delivery and better penetration into the bacterial cell. Thus, lower and less frequent doses of doxycycline and minocycline can be given. For this reason and due to the widespread resistance to tetracycline-HCl, doxycycline and minocycline tend to be the tetracyclines most commonly used [110].

The subgingival topical application of tetracycline has been promoted in several systems (powder, gel, irrigation solution, incorporated in nonresorbable fibers (dialysis tubing or ethylene–vinyl acetate monolithic fibers) [75].

**Aqueous tetracycline-HCl solutions** irrigated subgingivally has determined conflicting results when it was used as an adjunct to subgingival debridement. Shiloah and Patters [88] showed that a single episode of pocket irrigation with 2 cc of aqueous tetracycline-HCl solutions (50 mg/mL (5%)) following thorough scaling did not augment the effects of mechanical root debridement on the clinical and microbial parameters

of periodontitis. MacAlpine et al. [49] also reported that biweekly chlorhexidine, tetracycline, or saline irrigation of deep pockets (probing depth greater than or equal to 6 mm) did not appear to augment the effects of nonsurgical periodontal therapy.

In contrast, Christersson et al. [12] using a continuous subgingival irrigation with approximately 10–15 mL of an aqueous tetracycline-HCl solution prepared freshly at a concentration of 100 mg/mL for 5 min have obtained significant clinical improvements: a decrease in pocket depth of 2.2 mm and 2.1 mm, after 3 and 6 months and an average periodontal attachment level gain of  $2.1 \pm 1.1$  mm at 3 months and  $1.8 \pm 1.1$  mm at 6 months in periodontal pockets initially  $>5$  mm. Silvestein et al. [89] reported that tetracycline irrigation alone and scaling and root planning (SRP) alone had a similar effect in changing the subgingival microflora from one associated with disease to one associated with health, in patients with moderate adult periodontitis (average probing depth (PD) of  $5.6 \pm 0.9$  mm).

Topical application of a **tetracycline paste** containing 40% tetracycline in white petrolatum, even if determined significant improvements in clinical parameters, did not appear to augment the effect of mechanical debridement in patients with adult periodontitis [104], moderate/severe periodontitis [18], or localized juvenile periodontitis over a 12-week period [105].

The **tetracycline fibers** are 0.5 mm in diameter, 23 cm in length, and consist of a nonresorbable but biologically inert plastic copolymer (ethylene–vinyl acetate) loaded with 12.7 mg tetracycline hydrochloride powder and function as a controlled-delivery device that is able to maintain subgingival concentrations above 1,590 µg/mL crevicular fluid for 10 days (Actisite, Alza Laboratories Inc., Paolo Alto, CA) [22, 100].

Placement of the fiber is relatively time-consuming, and takes between 5 and 15 min per tooth. Although the clinician becomes faster with experience, many clinicians get frustrated with the technique. Was it necessary to place the fiber completely around the tooth, or could a single-point source of tetracycline be just as effective? As a possible alternative to the tetracycline fiber, a **tetracycline strip** was developed (Periodontal Therapeutic System, Alza Laboratories Inc.). These strips are ethylene/vinyl/acetate strips approximately 0.65 mm thick, 1 mm wide, and 5 cm in length, with an estimated tetracycline content of 13.5 mg. A cyanoacrylate adhesive was applied to retain the strips into the pocket. The time

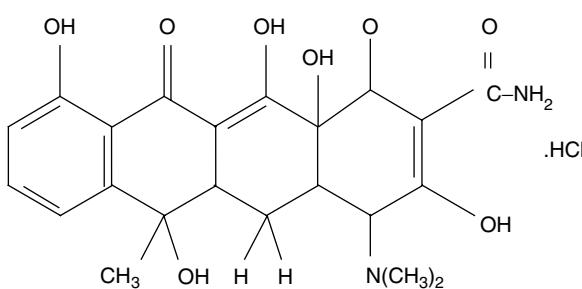


Fig. 4.1 The structural formula for tetracycline

required for placement was, on average, 1.9 min for single strips and 3.25 min for multiple strips. It was suggested that multiple strips are superior to a single strip in reducing bleeding on probing, and that local delivery of tetracycline is superior to root planning alone in reducing probing depth (PD) [22].

Several investigations have reported significant additional PD reduction and/or attachment gain when tetracycline fibers were applied after thorough scaling and root planning (Table 4.1).

A recent meta-analysis was performed by Hung & Douglass [38] on ten papers that compared the effect of local tetracycline fiber application with scaling and root planning [10, 16, 26, 32, 41, 46, 61, 76, 113, 114]. Two papers were from the same study population [61, 113]. Eight out of ten studies showed that combined treatment has a better prognosis in the reduction of periodontal PD than either treatment alone; six out of nine studies showed a larger gain of attachment level for the combined treatments. Scaling and root planning resulted in a mean periodontal PD reduction of 0.60–1.88 mm and a mean gain of attachment level of 0.20–1.61 mm; the respective results for combined treatments were 1.0–2.80 mm and 0.66–2.34 mm.

Pavia et al. [71] included 29 studies published in English prior to December 2001 with the aim of performing a meta-analysis on the effects of local tetracycline in various forms used in the treatment of periodontal disease. A significant mean reduction in PD was observed when tetracycline was used as an adjunct to SRP (mean difference at 12 weeks, 0.69 mm; 95% confidence interval, 0.57–0.81;  $P<0.001$ ). Tetracycline alone did not result in a statistically significant clinical improvement compared with SRP but did perform better than placebo for people whose PD was  $>6$  mm after 4 ( $P=0.001$ ) and 8 ( $P=0.005$ ) weeks. Weighing up the potential advantages and disadvantages of the local delivery systems studied, the author's conclusion that "local delivery systems of tetracycline should be considered cautiously and this antibiotic should rather be used as a complement to conventional therapy or when it has not proven successful" is pragmatic and useful. Gilbert [23] added to this the fact that local delivery agents are generally quite expensive and that reinfection of pockets from bacterial reservoirs in the mouth is likely and suggested that local delivery agents should be reserved for use in specific situations and not as a general panacea.

Another meta-analysis performed by Bonito et al. [8] on 16 studies of locally applied tetracycline

preparations revealed that the overall estimated PD reduction of 0.47 and 0.24 mm clinical attachment level (CAL) gain were statistically significant favoring the locally delivered adjuncts to SRP.

The difficulties of performing adequate debridement in furcations by mechanical means has prompted experimentation with chemotherapeutic agents in these areas [11]. Three studies have evaluated the therapeutic effects of various vehicles of tetracycline as a supplement to mechanical debridement: subgingival irrigation with tetracycline in furcations with class I, II, and III involvements [64], tetracycline in a cross-linked collagen film used in furcation class II involvements [57], and tetracycline-containing fibers mandibular class II furcations with persistent bleeding [101]. Aside from a significant short-term anti-inflammatory effect mirrored by the increased reduction in bleeding on probing (BOP), tetracycline in slow-delivery devices does not seem to greatly enhance or prolong the effectiveness of commonly used subgingival debridement in class II furcations. Overall, the results from the mentioned studies could not lend clear acceptance to the implementation of adjunctive local drug therapy in furcation involvements, regardless of the degree of severity [11].

## 4.2.2 Minocycline

Minocycline is an antimicrobial tetracycline derivative which is active against a broad spectrum of gram-negative and gram-positive anaerobes including pathogens associated with adult periodontitis [17, 33] (Figs. 4.2 and 4.3). It has been shown that topical ointment of minocycline significantly reduced tooth mobility, gingival index, and alveolar bone loss in a periodontitis rat [115] and mouse model [37].

Minocycline has been incorporated in an ointment, utilized for subgingival application, which consists of a bioresorbable delivery system loaded with 2% minocycline HCl (Periocline, Sun Star, Osaka, Japan; Dentomycin, Lederle Laboratories, England). The matrix is a mixture of hydroxyethyl-cellulose, aminoalkyl-methacrylate triacetate, and glycerinum [75]. Magnesium chloride was used to modify the release properties. The elimination half-life based on the assumption of a single-compartment open model was estimated to be 3.9 h, and the total time of effective

**Table 4.1** Summary of clinical studies evaluating the local administration of tetracycline fiber in periodontitis

| Authors              | Type of study            | No. patients | Type of disease  | Treatment   | Study period | Results  |
|----------------------|--------------------------|--------------|--|---|--------------|--|
| Aimetti et al. [4]   | Intrasubject split-mouth | 19           | Adult periodontitis  | - SRP<br>- SRP+ tetracycline fiber  | 12 months    | SRP plus TC fibers gave the greatest advantage in the treatment of periodontal persistent lesions at least 1.2 months following treatment  |
| Drisko et al. [16]   | Intrasubject split-mouth | 122          | Adult periodontitis with teeth with PD≥5 mm and BOP                                  | 1. SRP<br>2. SRP+ tetracycline fiber for 10 days<br>3. Fiber therapy alone for 10 days<br>4. Two 10-day serial fiber applications | 12 months    | All treatments resulted in similar improvements in clinical parameters compared to baseline and were equally effective in the treatment of periodontitis as measured by PD reduction, CAL gain, and reduction of BOP   |
| Flemming et al. [21] | Intrasubject split-mouth | 35           | Localized persistent or recurrent periodontitis under supportive periodontal therapy | - SRP<br>- SRP+ tetracycline fiber fiber therapy alone for 10 days  | 6 months     | Compared to control teeth, in test teeth at 6 months significantly ( $P < 0.01$ ) lower scores were found for gingival index, pocket PDs, and PMN elastase-alpha1-proteinase inhibitor concentrations in GCF   |
| Goodson et al. [26]  | Intrasubject split-mouth | 113          | Adult periodontitis  | - Tetracycline fiber<br>- Control fiber<br>- SRP<br>- Untreated   | 60 days      | Both TC fiber therapy and scaling decreased the number of sites infected with all the monitored species. TC fiber therapy significantly decreased PD, increased attachment level, and decreased BOP with controlled force to a greater extent than observed in all other test groups including scaling |

|                       |                          |    |  |  |           |   |
|-----------------------|--------------------------|----|--|--|-----------|---|
| Goodson et al. [27]   | Intrasubject split-mouth | 10 | Adult periodontitis  | - SRP<br>- SRP + tetracycline fiber<br>- Tetracycline fiber  | 10 days   | In no case were clinical results by scaling superior to results by local drug delivery, and by several measures local drug delivery was found to provide a better clinical response   |
| Heijl et al. [32]     | Intrasubject split-mouth | 10 | Adult periodontitis (95 teeth with PD ≥ 6 mm which initially bled on probing | - SRP<br>- SRP + tetracycline fiber  | 62 days   | The combined therapy eliminated BOP, and black-pigmented <i>Bacteroides</i> , and produced the greatest mean reduction in PD  |
| Lowenguth et al. [47] | Intrasubject split-mouth | 31 | Adult periodontitis  | 1. SRP<br>2. SRP + tetracycline fiber for 10 days<br>3. Fiber therapy alone for 10 days<br>4. Two 10-day serial fiber applications | 12 months | Numbers and proportions of detectable pathogens ( <i>Pt</i> , <i>Fn</i> , <i>Ec</i> , <i>Ct</i> , <i>Aa</i> ) exhibited a triphasic temporal response: a precipitous initial decrease immediately following therapy; a rise in proportions in the 1- to 3-month post-therapy period; and a spontaneous decline in the absence of therapy over the 3- to 12-month period. No significant differences between therapies |
| Maze et al. [54]      | Intrasubject split-mouth | 10 | Adult periodontitis  | 1. TTC-PLGA,<br>2. control strips without TTC (PLGA),<br>3. SRP, and<br>4. untreated control                                       | 12 weeks  | Applications of intrareviewicular fibers, when compared to SRP, appears to have an enhanced antibacterial effect and a similar clinical effect in supportive periodontal therapy patients   |

(continued)

**Table 4.1** (continued)

| Authors                | Type of study            | No. patients | Type of disease   | Treatment  | Study period | Results   |
|------------------------|--------------------------|--------------|---|--|--------------|---|
| Newman et al. [61]     | Intrasubject split-mouth | 113          | Localized recurrent periodontitis sites in maintenance patients | - SRP<br>- SRP + tetracycline fiber  | 6 months     | At 1, 3, and 6 months, adjunctive fiber therapy was significantly better in reducing PD ( $P < 0.05$ ) and reducing BOP ( $P < 0.05$ ) than S and RP alone. At 6 months, fiber therapy was significantly better in promoting clinical attachment gain ( $P < 0.05$ ) than SRP alone |
| Romano et al. [83]     | Intrasubject split-mouth | 11           | Persistent periodontitis  | - SRP<br>- SRP + tetracycline fiber  | 12 months    | Tetracycline local delivery gave the greatest advantage in the long-term treatment of periodontal persistent lesions  |
| Trombelli et al. [102] | Intrasubject split-mouth | 12           | Adult periodontitis with moderate to deep periodontal pockets   | - Supra- and subgingival scaling was performed with an ultrasonic scaler + supplemental irrigation with 15 mL of a 100-mg/mL tetracycline solution<br>- Supra- and subgingival scaling was performed with an ultrasonic scaler + tetracycline-loaded fiber left in place for 10 days | 60 days      | No adjunctive effect on the healing response was obtained by augmenting mechanical debridement with tetracycline  |

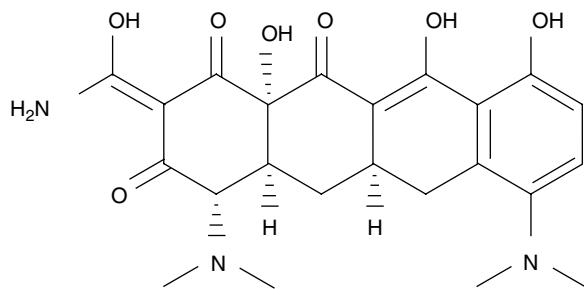
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|------------------------|--------------------------|----|---|--|----------|--|
| Wong et al. [114]      | Intrasubject split-mouth | 30 | Recurrent periodontitis (PD between 4 and 8 mm with BOP) on maintenance therapy | - SRP<br>- SRP+ tetracycline fiber   | 6 months | There was no statistically significant difference between the two treatment groups with respect to bacterial proportions or the number of positive sites   |
| Cattabriga et al. [10] | Intrasubject split-mouth | 25 | Recurrent periodontitis with PD ≥ 4 mm and BOP under maintenance therapy        | - Tetracycline fiber<br>- SRP+ tetracycline fiber  | 6 months | Use of tetracycline fibers and fibers with scaling produced 1.8 and 1.7 mm reductions in PD, respectively, 1 month after treatment; reductions declined to 1.3 and 0.8 mm at 3 months, but rebounded to 1.5 and 1.3 mm at 6 months. The percentage of teeth exhibiting BOP decreased from 100% at baseline to 68% and 50% in the fiber and fiber plus scaling groups, respectively, at 6 months. None of the differences was statistically significant |
| Radvar et al. [76]     | Parallel study           | 54 | Persistent periodontal pockets ≥ 5 mm and BOP and/or suppuration                | - SRP<br>- SRP+ 25% tetracycline fibers<br>- SRP+ 2% minocycline gel<br>- SRP+ 25% metronidazole gel | 6 weeks  | The improvements in clinical parameters were greater in all three adjunctive treatment groups than SRP alone. The mean PD reduction for SRP+ minocycline was 0.87 mm   |

(continued)

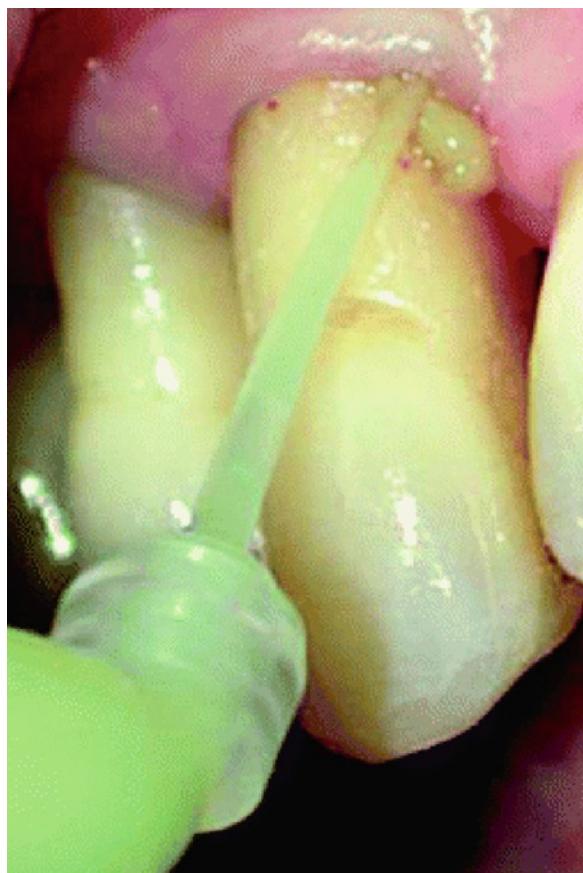
**Table 4.1** (continued)

| Authors         | Type of study            | No. patients | Type of disease   | Treatment  | Study period | Results  |
|-----------------|--------------------------|--------------|---|--|--------------|--|
| Lie et al. [46] | Intrasubject split-mouth | 18           | Moderate to severe chronic periodontitis with teeth with PD≥ 5 mm and BOP | - SRP<br>- SRP + local metronidazole gel<br>- SRP + 3% tetracycline ointment | 6 months     | The average PD reduction for the three groups at 6 months was 1.5 mm and the average gain of CAL was 0.8 mm. There were no significant differences between the effects following topical application of the metronidazole gel or the tetracycline ointment. SRP alone appeared as effective as the drug-augmented regimens, although there was a weak but nonsignificant tendency for better results in sites treated with the antibiotic drugs. <i>Aa</i> was generally not detected; <i>Pi</i> was not significantly reduced, while <ipg< i=""> was significantly reduced in all treatment groups</ipg<> |

SRP scaling and root planning, GCF gingival crevicular fluid, CAL clinical attachment level, PD probing depth, BOP bleeding on probing, *Pi* *P. intermedia*, *Fn* *F. nucleatum*, *Ec* *E. corrodens*, *Cr* *C. rectus*, *Aa* *A. actinomycetemcomitans*



**Fig. 4.2** The structural formula for minocycline



**Fig. 4.3** Application of 2% minocycline gel [55] (Reprinted with permission John Wiley & Sons)

antimicrobial activity was expected to be approximately 1 day. As judging from a study of minimal inhibition concentration of 2% minocycline against periodontopathogenic bacteria, the curve obtained from values measured up to 72 h after administration in 97 patients with periodontitis showed that the effective concentration was maintained for approximately 7 days [48].

The clinical and microbial benefits of minocycline ointment, after repeated subgingival application and in combination with thorough subgingival root planning, have been tested in several studies (Table 4.2).

A recent meta-analysis was performed by Hung and Douglass [38] who analyzed the effect from 2% *minocycline gel* from six studies [27, 41, 59, 76, 98, 107]. All of them compared the combined effect of SRP and minocycline gel with SRP alone. The adjunctive treatment of minocycline gel with SRP showed additional improvement in reduction of periodontal PD and gain of attachment in these studies. The SRP alone resulted in a mean periodontal PD reduction of 0.60–2.30 mm and a mean gain of attachment level of 0.26–1.56 mm; the respective ranges for the combined treatments were 0.87–3.64 and 0.45–1.95 mm [38].

Recently, an agent using microspheres containing minocycline hydrochloride (1 mg) was developed (Arestin, Ora Pharma Inc., Warminster, PA, USA). Once Arestin is inserted, it immediately adheres to the periodontal pocket. Gingival crevicular fluid hydrolyzes the polymer, causing water-filled channels to form inside the microspheres, allowing a controlled, sustained release of minocycline. A single application as an adjunct to root planning results in an additional reduction in the proportion of spirochetes and motile organisms as well as in the number of black-pigmented species, the red-complex bacteria, and the sum of *Porphyromonas gingivalis*, *Tannerella forsythia* (formerly *Tannerella forsythensis*), and *Treponema denticola* [24, 30, 39, 59, 75].

Studies on this agent indicate clinically beneficial effects when used as an adjunct to supra- and subgingival mechanical debridement [14, 24, 27, 30, 34, 41, 48, 53, 55, 59, 65, 69, 70, 76, 98, 106, 107, 112]; Widman modified flap [33], or conventional maintenance therapy [56] (Table 4.2).

A meta-analysis performed by Bonito et al. [8] on eight studies of locally applied minocycline, all appearing between 1993 and 2002, is highly supportive of its use as an adjunct to SRP than studies of other local chemotherapeutic agent. The mean effect size was a statistically significant 0.49 mm reduction in PD and 0.46 mm gain in CAL favoring use of local minocycline as an adjunct to SRP.

It has been shown also that local minocycline is able to induce clinical improvements of incipient peri-implantitis lesions, as an adjunct to mechanical treatment [79, 81]. For the deepest sites of the

**Table 4.2** Summary of clinical studies evaluating the local administration of minocycline in periodontal disease

| Authors              | Type of study   | No. patients | Type of disease   | Treatment                          | Device       | Study period | Results   |
|----------------------|---|--------------|---|------------------------------------|--------------|--------------|---|
| Cortelli et al. [13] | Randomized, single-blind, controlled, parallel-group design | 30           | Advanced chronic periodontitis with teeth with PDs $\geq$ 6 mm  | – SRP+placebo<br>– SRP+minocycline | Microspheres | 24 months    | When the PD values were compared, there was a bigger reduction in the test group in the 12th month (3.80mm from 7.47mm, $P<0.05$ vs. 5.17mm from 7.73mm in the controls). No significant differences were observed at 24th months.  |
| de Lima et al. [53]  | Split-mouth trial   | 11           | Periodontal defects with PD $>5.0$ mm in type I diabetes mellitus patients  | – SRP+placebo<br>– SRP+minocycline | Gel          | 12 months    | Subgingivally delivered doxycycline hyclate produces additional favorable clinical results to periodontal therapy in type 1 diabetes mellitus patients  |
| Goodson et al. [24]  | Randomized single-blind, parallel study                     | 127          | Moderate to advanced chronic periodontitis who had at least five teeth with PDs $\geq$ 5 mm                                     | – SRP<br>– SRP+minocycline         | Microspheres | 1 month      | The antimicrobial effect of locally delivered minocycline exhibited surprising specificity of action in being directed almost entirely toward inhibition of bacteria generally considered periodontal pathogens with little inhibitory effect on other species. In addition, significantly reduced red-complex bacteria (6.49% vs 5.03%), PD (1.38 vs 1.01 mm, $P=0.00004$ ), CAL (1.16 vs 0.80, $P=0.0004$ ), and BOP (25.2% vs 13.8%, $P=0.009$ ) to a greater extent than treatment by SRP alone |
| Graça et al. [27]    | Randomized, double-blind, vehicle-controlled study          | 30           | Moderate to advanced periodontitis with a minimum of two pockets with PD=5–10 mm on separate teeth with attachment loss $>4$ mm | – SRP+placebo<br>– SRP+minocycline | Gel          | 3 months     | Differences between groups in mean PD did not reach statistical significance at any visit, but mean probing attachment levels were different ( $P<0.05$ ) at both reassessments (baseline: test=6.86 mm, control=6.83 mm; 6 weeks: test=4.93 mm, control=5.30 mm; 12 weeks: test=4.91 mm, control=5.27 mm)  |

|                          |   |     |   |  |              |          |  |
|--------------------------|---|-----|---|--|--------------|----------|--|
| Grossi et al.<br>[30]    | Randomized,<br>single-blind,<br>controlled,<br>parallel-group<br>design | 127 | Moderate to<br>advanced<br>periodontitis  | – SRP<br>– SRP+<br>minocycline   | Microspheres | 1 month  | MM + SRP reduced red-complex bacteria numbers and proportions to a greater extent than SRP alone, irrespective of smoking status. Numbers and proportions of orange-complex bacteria were reduced in all groups treated with MM + SRP. Proportions of orange-complex bacteria increased in current smokers treated with SRP alone. Mean PDs for SRP + minocycline versus SRP alone were 1.40 and 1.06 mm in never-smokers, 1.45 and 1.07 in former smokers, and 1.28 and 0.86 in current smokers. Mean CAL gain for SRP + minocycline versus SRP alone were 1.15 and 0.87 mm in never-smokers, 1.17 and 0.78 in former smokers, and 1.15 and 0.74 in current smokers. Mean percentage of BOP reduction for SRP + minocycline versus SRP alone was significant in current smokers (36.5% vs 11.2%, $P < 0.05$ ) |
| Hellström et al.<br>[33] | Randomized,<br>single-blind,<br>controlled,<br>parallel-group<br>design | 60  | Moderate to<br>advanced chronic<br>periodontitis<br>with teeth with<br>PDs ≥ 5 mm | – MWF alone<br>– MWF +<br>minocycline  | Microspheres | 25 weeks | Applications of local minocycline as an adjunct to surgery were associated with statistically significantly greater reductions in PD (2.51 vs 2.18 mm) than surgery alone  |
| Kinane &<br>Radvar [41]  | Parallel study  | 79  | Persistent<br>periodontal<br>pockets ≥ 5 mm<br>and BOP and/or<br>suppuration      | – SRP alone<br>– SRP + 25%<br>tetracycline<br>fibers<br>– SRP + 2%<br>minocycline gel<br>– SRP + 25%<br>metronidazole<br>gel | Gel          | 6 months | All four therapies resulted in significant improvements from baseline in PD, CAL, BOP, and the Modified Gingival Index scores. The improvements in clinical parameters were greater in all three adjunctive treatment groups than SRP alone. The mean PD reductions at 6 months were for scaling + minocycline = 1.10 mm   |
| Lu & Choi [48]           | Split-mouth trial   | 15  | Moderate to<br>severe periodon-<br>titis with residual<br>pockets<br>(PD > 6 mm)  | – SRP<br>– SRP +<br>minocycline  | Gel          | 18 weeks | SRP with adjunctive subgingival administra-<br>tion of minocycline ointment has a signifi-<br>cantly better and prolonged effect compared<br>to SRP alone on the reduction of PD, CAL,<br>gingival index, and interleukin-1beta content<br>in GCF  |

(continued)

**Table 4.2** (continued)

| Authors              | Type of study   | No. patients | Type of disease   | Treatment   | Device       | Study period | Results   |
|----------------------|---|--------------|---|---|--------------|--------------|---|
| McColl et al. [55]   | Randomized, double-blind, controlled study                  | 40           | Moderate to advanced chronic periodontitis  | – Minocycline<br>– SRP only                                       | Gel          | 12 months    | No significant differences between the groups   |
| Meinberg et al. [56] | Randomized, single-blind, controlled, parallel-group design | 48           | Moderate to advanced chronic periodontitis with teeth with PDs $\geq 5$ mm  | – PM therapy<br>– SRP+ minocycline                                | Microspheres | 12 months    | PDs showed greater mean improvement in SRP + minocycline ( $0.9 \pm 0.1$ versus $0.4 \pm 0.1$ mm, $P = 0.02$ ), with 25% of subjects in SRP + minocycline gaining $\geq 2$ mm compared to 4.2% in PM ( $P < 0.05$ ). The mean loss in bone height and percent of subjects losing bone height were less in SRP + minocycline ( $0.05 \pm 0.05$ mm; 12.5%) than PM ( $0.09 \pm 0.08$ mm; 16.7%), but bone height differences were not statistically significant |
| Nakagawa et al. [59] | Split-mouth   | 11           | Recurrent periodontal PDs greater than 5 mm and loss of attachment greater than 2 mm within the previous 3 months | – SRP+ irrigation<br>with biological saline<br>– SRP+ minocycline | Ointment     | 3 months     | Clinical conditions improved in both groups following treatment; significantly better improvements were obtained in the test group. Microbiological study revealed that Periocline effectively eliminated periodontopathic gram-negative bacteria   |
| Oringer et al. [65]  | Randomized, single-blind, controlled, parallel-group design | 48           | Moderate to advanced chronic periodontitis with teeth with PDs $\geq 5$ mm  | – SRP+ vehicle<br>– SRP+ minocycline                              | Microspheres | 6 months     | Local administration of minocycline microspheres led to a potent short-term (1 month) reduction in GCF IL-1 levels. The pyridinoline cross-linked carboxy-terminal telopeptide of type I collagen (ICTP), a bone-specific degradation product, were reduced by 33.2% at 1 month in the test group compared to a 17.2% reduction in the controls   |

|                         |   |     |  |   |              |           |  |
|-------------------------|---|-----|--|---|--------------|-----------|--|
| Paquette et al.<br>[69] | Multicenter parallel study                            | 271 | Moderate to advanced chronic periodontitis   | - SRP<br>- SRP+ vehicle<br>- SRP+ minocycline   | Microspheres | 9 months  | Significantly greater PD reductions with SRP plus adjunctive minocycline microspheres treatment were observed at 1, 6, and 9 months ( $P<0.05$ ) versus control treatments. At 9 months, smokers treated with SRP plus minocycline microspheres exhibited a PD reduction of 1.19 mm from baseline, as compared to 0.90 mm for smokers treated with SRP alone   |
| Paquette et al.<br>[70] | Multicenter parallel study                            | 499 | Moderate to advanced chronic periodontitis with at least four teeth with PD=6–9 mm | - SRP<br>- SRP+ minocycline   | microspheres | 3 months  | Significantly more sites treated with adjunctive minocycline microspheres exhibited PDs <5 mm at 1 ( $P=0.0009$ ) and 3 ( $P=0.01$ ) months compared to sites treated with SRP alone, both in the overall population and in smokers. In addition, significantly more sites decreased by 1, 2, or 3 mm in the adjunctive minocycline group than in the SRP alone group at 1 and 3 months, both overall as well as in smokers ( $P<0.05$ ) |
| Radvar et al.<br>[76]   | Parallel study  | 54  | Persistent periodontal pockets $\geq 5$ mm and BOP and/or suppuration              | - SRP<br>- SRP+25% tetracycline fibers<br>- SRP+2% minocycline gel<br>- SRP+25% metronidazole gel | Ointment     | 6 weeks   | The improvements in clinical parameters were greater in all three adjunctive treatment groups than SRP alone. The mean PD reductions for scaling + minocycline were 0.87 mm  |
| Timmerman et al. [98]   | Randomized, double-blind, parallel, comparative study | 20  | Moderate to severe chronic adult periodontitis                                     | - SRP<br>- SRP+ minocycline   | Ointment     | 18 months | The patient group responded favorably to SRP, but did not benefit from an effect of local of minocycline   |

(continued)

**Table 4.2** (continued)

| Authors                      | Type of study   | No. patients | Type of disease  | Treatment                                     | Device       | Study period | Results   |
|------------------------------|---|--------------|--|---|--------------|--------------|---|
| van Steenberghe et al. [107] | Randomized, double-blind, parallel, comparative study | 104          | Moderate to severe chronic adult periodontitis                     | – SRP+ vehicle<br>– SRP+ minocycline          | Ointment     | 15 months    | Sites treated with minocycline ointment always produced statistically significantly greater reductions than sites which received the vehicle control. For initial PDs $\geq 5$ mm, a mean reduction in PD of 1.9 mm was seen in the test sites, versus 1.2 mm in the control sites. Sites with a baseline PD $\geq 7$ mm and bleeding index $>2$ showed an average of 2.5 mm reduction with minocycline versus 1.5 mm with the vehicle. Gains in attachment (0.9 and 1.1 mm) were observed in minocycline-treated sites, with baseline PD $\geq 5$ mm and $\geq 7$ mm, respectively, compared with 0.5 and 0.7 mm gain at control sites   |
| Williams et al. [112]        | Multicenter   | 748          | Moderate to advanced periodontitis with teeth with PDs $\geq 5$ mm | – SRP<br>– SRP+ vehicle<br>– SRP+ minocycline | Microspheres | 9 months     | Minocycline microspheres plus SRP provided substantially more PD reduction than either SRP alone or SRP plus vehicle. The mean percent of sites with PD reductions $\geq 2$ mm for the three groups was 32.87 (SRP), 28.98 (SRP+ vehicle), and 40.52 (SRP+minocycline). For patients with mean PD $\geq 6$ mm, mean reduction was 1.05 mm (SE 0.10) for the SRP group, and 1.49 mm (SE 0.09) for the SRP+minocycline group. Difference between groups was 0.41 mm, $P < 0.01$ . For patients with mean PD $\geq 7$ mm, mean reduction was 0.98 mm (SE 0.29) for the SRP group, and 1.99 mm (SE 0.31) for the SRP+minocycline group. Difference between groups was 1.01 mm, $P = 0.06$ |

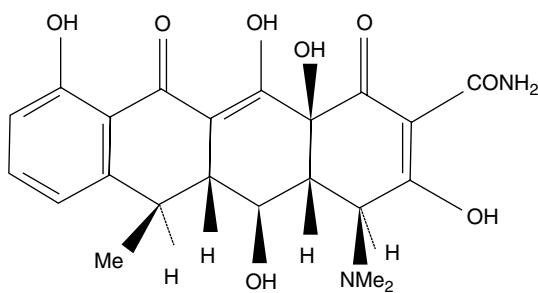
SRP scaling and root planning, GCF gingival crevicular fluid, CAL clinical attachment level, PD probing depth, BOP bleeding on probing, PM periodontal maintenance

minocycline-treated implants, mean PD was reduced from 5.0 mm to 4.1 mm at 3 months [81], while the mean PD reduction was 0.6 mm at 12 months [79, 80].

### **4.2.3 Doxycycline**

Doxycycline is a broad-spectrum semisynthetic tetracycline. Doxycycline is bacteriostatic, inhibiting bacterial protein synthesis due to disruption of transfer RNA and messenger RNA at ribosomal sites (Figs. 4.4 and 4.5). In vitro testing has shown that *P. gingivalis*, *Prevotella intermedia*, *Campylobacter rectus*, and *Fusobacterium nucleatum*, which are associated with periodontal disease, are susceptible to doxycycline at concentrations  $\leq 6.0 \mu\text{g/mL}$  [90].

A two-syringe mixing system for the controlled release of doxycycline (Atridox, Tolmar Inc., Fort Collins, CO, USA) has been evaluated in a number of investigations. The Atridox product is a subgingival controlled-release product composed of a two-syringe mixing system. Syringe A contains 450 mg of the Atrigel® Delivery System, which is a bioabsorbable, flowable polymeric formulation composed of 36.7% poly (DL-lactide) (PLA) dissolved in 63.3% *N*-ethyl-2-pyrrolidone (NMP). Syringe B contains doxycycline hydiate, which is equivalent to 42.5 mg doxycycline. The constituted product is a pale yellow to yellow viscous liquid with a concentration of 10% of doxycycline hydiate. Upon contact with the crevicular fluid, the liquid product solidifies and then allows for controlled release of the drug for a period of 7 days [92]. It was shown



**Fig. 4.4** The structural formula for doxycycline hydrate. Empirical formula:  $(C_{22}H_{24}N_2O_8 \cdot HCl)_2 \cdot C_2H_6O \cdot H_2O$  and a molecular weight of 512.9



**Fig. 4.5** Instillation of doxycycline gel with marginal overflow [19] (Reprinted with permission John Wiley & Sons)

that after subgingival application of biodegradable doxycycline gel, mean doxycycline levels in gingival crevicular fluid that exceeded 16 µg/mL could be maintained for at least 12 days. The antibiotic effect was limited mainly to the subgingival sites of application of the doxycycline gel [42].

Studies on this agent revealed that the use of locally delivered doxycycline may constitute an important adjunct for the active and supportive treatments of severe periodontal disease (Table 4.3).

It was shown that adjunctive local doxycycline resulted in a greater reduction in the frequency of *P. gingivalis* following initial and supportive therapy compared to conventional treatment (SRP alone) [50, 52, 86]. Similar results were reported by Ratka-Krüger et al. [78], who showed that the addition of subgingival instillation of a 14% doxycycline gel to SRP resulted in pronounced reduction of periodontal pathogens (*Actinobacillus actinomycetemcomitans*, *T. forsythensis*, *P. gingivalis*, *T. denticola*) after 3 months and stabilizing results up to 6 months after therapy. In contrast,

**Table 4.3** Summary of clinical studies of doxycycline hyclate (ATRIDOX) in periodontal disease

| Authors               | Type of study   | No. patients | Type of disease                      | Treatment  | Study period | Results  |
|-----------------------|---|--------------|--------------------------------------|--|--------------|--|
| Ağan et al. [3]       | Intrasubject split-mouth                                | 18           | Chronic and aggressive periodontitis | – SRP teeth of chronic periodontitis patients<br>– SRP+doxycycline teeth of chronic periodontitis patients<br>– SRP teeth of aggressive periodontitis patients<br>– SRP+doxycycline teeth of aggressive periodontitis patients | 6 months     | The reduction in PD (mm) at 1, 3, and 6 months with respect to the baseline was statistically significant in all of the groups ( $P = 0.0051$ ), but there was no statistically significant difference between the mean PD reduction of the four groups at each time point. The improvements of CAL did not have statistically significant difference either between the groups at each time point, or when compared with the baseline. No differences on GCF matrix metalloproteinase (MMP)-8 levels between the groups   |
| Akalin et al. [6]     | Three study groups with intrasubject split-mouth design | 45           | Chronic periodontitis                | – Systemic doxycycline<br>– Systemic doxycycline+SRP<br>– Doxycycline local<br>– SRP+doxycycline<br>– SRP alone  | 7 weeks      | No significant difference was found between local doxycycline and SRP treatments. Significant PD reduction for local doxycycline alone (from $0.50 \pm 0.13$ mm to $0.34 \pm 0.09$ mm) compared with systemic doxycycline (from $0.72 \pm 0.19$ mm to $0.49 \pm 0.13$ mm) ( $P < 0.05$ ). The local DOX alone treatment seemed more effective than SD alone treatment on PD reduction, but no significant difference was found between them when combined with the SRP. Local DOX may be more preferable than systemic DOX as an adjunct to mechanical treatment since local DOX seems more effective than systemic DOX on PD reduction and does not have the side effects of systemic DOX |
| Jorgensen et al. [40] | Intrasubject split-mouth                                | 8            | Moderate to advanced periodontitis   | – SRP alone<br>– SRP+doxycycline   | 4 weeks      | Sites receiving SRP + DOX and sites receiving SRP alone exhibited similar levels of periodontal pathogens at baseline and did not differ significantly in total viable counts and proportional recovery of periodontopathic bacteria post treatment  |
| Machion et al. [52]   | Intrasubject split-mouth                                | 43           | Chronic periodontitis                | – SRP+irrigation with saline solution<br>– SRP+doxycycline   | 6 months     | CAL gain was greater for SRP + DOX ( $1.63 \pm 0.93$ mm) than for SRP ( $1.04 \pm 0.71$ mm). In addition, deep pockets ( $> 7$ mm) showed a significant reduction ( $3.78 \pm 1.41$ vs $2.60 \pm 1.28$ mm) and CAL gain ( $2.54 \pm 1.27$ vs $1.29 \pm 0.95$ mm) when doxycycline was applied. The proportion of sites showing CAL gain of 1–2 mm was 36.8% versus 21.7% for SRP + DOX and SRP   |

|                        |                          |     |   |   |          |  |
|------------------------|--------------------------|-----|---|---|----------|--|
| Machion et al. [50]    | Intrasubject split-mouth | 16  | Chronic periodontitis   | – SRP+ irrigation with saline solution<br>– SRP+doxycycline   | 3 months | No statistically significant difference was found in the reduction of <i>Actinobacillus actinomycetemcomitans</i> in either the SRP + DOX or SRP group ( $P > 0.05$ ). The reduction in <i>Tannerella forsythensis</i> , <i>Porphyromonas gingivalis</i> , and <i>T. forsythensis</i> + <i>P. gingivalis</i> was statistically significant for SRP + DOX only ( $P = 0.016$ , $0.027$ , and $0.027$ , respectively). The proportion of sites free of <i>T. forsythensis</i> at 3 months was 53% for SRP + DOX and 9% for SRP ( $P = 0.02$ ). For <i>P. gingivalis</i> , this proportion was 82% and 40%, respectively ( $P = 0.05$ ). The use of locally delivered doxycycline may promote the elimination of <i>T. forsythensis</i> and <i>P. gingivalis</i> in a greater proportion of sites compared to conventional SRP in smokers |
| Machion et al. [51]    | Intrasubject split-mouth | 48  | Chronic periodontitis   | – SRP+ irrigation with saline solution<br>– SRP+doxycycline   | 2 years  | In initially deep pockets ( $>7$ mm), SRP + DOX showed greater PD reduction than SRP at 6 and 18 months (mean difference between groups of 1.18 and 1.73 mm, respectively; $P < 0.05$ ) and greater CAL gain in all periods after 3 months (mean difference between groups of 1.16, 1.99, and 1.78 mm, at 6, 18, and 24 months, respectively; $P < 0.05$ )   |
| Wennström et al. [111] | Multicenter study        | 105 | Moderately advanced chronic periodontitis with PD $\geq 5$ mm | – SRP+ full-mouth supra-/subgingival debridement + doxycycline<br>– Debridement (supra- and subgingival ultrasonic instrumentation without analgesia)+doxycycline | 6 months | At 3 months, the proportion of sites showing PD $<4$ mm was significantly higher in the debridement group than in the SRP group (58% vs 50%; $P < 0.05$ ). The CAL gain at 3 months amounted to 0.8 mm in the debridement group and 0.5 mm in the SRP group ( $P = 0.064$ ). The proportion of sites demonstrating a clinically significant CAL gain ( $>2$ mm) was higher in the debridement group than in the SRP group (38% versus 30%; $P < 0.05$ ). At the 6-month examination, no statistically significant differences in PD or CAL were found between the two treatment groups   |

(continued)

Table 4.3 (continued)

| Authors                        | Type of study               | No. patients | Type of disease                   | Treatment  | Study period | Results   |
|--------------------------------|-----------------------------|--------------|-----------------------------------|--|--------------|---|
| Martorelli de Lima et al. [53] | Intrasubject split-mouth    | 11           | Type 1 diabetes mellitus patients | – SRP+ placebo gel<br>– SRP+ doxycycline                             | 12 months    | For PD, the adjunctive doxycycline group when compared with the SRP alone group at 6 weeks, and 6, 9, and 12 months demonstrated mean PDs of 2.5 versus 3.3 mm (difference = 0.08 mm) and 2.1 versus 3.6 mm (difference = 1.5 mm), and 2.0 mm versus 3.8 mm (difference = 1.8 mm) and 2.0 versus 3.9 mm (difference = 1.9 mm), respectively. A statistically significant difference for PD between study groups was only present at 12 months favoring the adjunctive doxycycline group ( $P < 0.05$ ). For attachment level, the adjunctive doxycycline group when compared with the SRP alone group at 6 weeks and 6, 9, and 12 months, demonstrated mean attachment level values of 4.5 versus 5.1 mm (difference = 0.6 mm), 4.1 versus 5.4 mm (difference = 1.3 mm), 4.0 versus 5.6 mm (difference = 1.6 mm), and 4.0 versus 5.7 mm (difference = 1.7), respectively. A statistically significant difference for CAL between study groups was only seen at 12 months favoring the adjunctive doxycycline group ( $P < 0.05$ ) |
| Salvi et al. [85]              | Intersubject parallel study | 47           | Adult periodontitis               | – SRP+ doxycycline<br>– SRP + Elyzol Dental Gel<br>– SRP + PerioChip | 18 weeks     | Between the baseline and 18-week examinations, subjects treated with Atridox showed a significantly greater gain in mean CAL of $0.33 \pm 0.09$ mm than subjects treated with Elyzol Dental Gel ( $0.03 \pm 0.09$ mm) ( $P = 0.03$ ). However, the gain in CAL of $0.16 \pm 0.10$ mm found after PerioChip application did not differ significantly from that obtained following the application of Atridox ( $P = 0.27$ ). Of the sites treated with Atridox, 42% gained $\geq 1$ mm CAL and 9% $\geq 2$ mm CAL as opposed to the sites treated with Elyzol Dental Gel, in which 34% gained $\geq 1$ mm CAL and 8% gained $\geq 2$ mm CAL. Of the sites treated with PerioChip, 36% gained $\geq 1$ mm and 6% gained $\geq 2$ mm CAL following a completed initial periodontal therapy   |
| Shaddox et al. [86]            | Intrasubject split-mouth    | 16           | Chronic periodontitis             | – SRP+ irrigation with saline solution<br>– SRP+ doxycycline         | 15 months    | The reduction in the number of sites positive for <i>P. gingivalis</i> and <i>T. forsythia</i> was statistically significant for SRP + DOX at 3 months (68% and 41.3%, respectively). The SRP group showed a greater frequency of <i>P. gingivalis</i> than the SRP + DOX group at 3 months (58% and 25%, respectively). There also was a greater reduction in the frequency of <i>P. gingivalis</i> at 3 months following retreatment with SRP + DOX compared to SRP (47% and 8%, respectively)  |

|                       |                             |     |  |  |           |  |
|-----------------------|-----------------------------|-----|--|--|-----------|--|
| Dannevitz et al. [15] | Intersubject parallel study | 39  | Recurrent moderate to severe periodontitis       | – SRP<br>– SRP+doxycycline   | 12 months | SRP + DOX resulted in better improvement of furcation involvement than SRP alone 3 months after treatment ( $P=0.041$ ). However, SRP + DOX failed to show a significant difference between both groups in the number of reinstrumentations  |
| Gupta et al. [31]     | Intersubject parallel study | 30  | Moderate to advanced chronic periodontitis       | – SRP<br>– SRP+doxycycline<br>– SR + chlorhexidine gel   | 3 months  | All treatments showed significant reductions in PD and CAL at 1 and 3 months when compared to baseline values ( $P<0.001$ ). At 3 months, sites treated with SRP + DOX and SRP + CHX showed an additional reduction in PD of $0.86 \pm 1.0$ mm and $0.66 \pm 1.58$ mm, respectively, significantly greater than SRP alone ( $P<0.02$ ). Differences in mean PD reduction between SRP + DOX and SRP + CHX were not significant ( $P=0.46$ ). At 3 months, differences in relative CAL between both SRP + DOX ( $0.80 \pm 0.92$ ) and SRP + CHX ( $0.63 \pm 1.47$ ) and SRP alone were statistically significant ( $P<0.02$ ). Differences in relative CAL between SRP + DOX and SRP + CHX were not significant ( $P=0.54$ ) |
| Bogren et al. [7]     | Intersubject parallel study | 128 | Treated moderate/ advanced chronic periodontitis | – Supportive periodontal therapy (SPT) (mechanical debridement, polishing, and oral hygiene reinforcement) only<br>– SPT+doxycycline | 3 years   | A statistically significant difference in favor of the adjunctive doxycycline therapy was found between the two groups only at the 3-month examination for: BOP (test group: BOP decrease 51% at baseline to 37% at 3 months; control: from 56% to 50%, $P<0.01$ ), PD (test: from 5.4 to 4.5 mm; controls: from 5.6 to 4.9 mm, $P<0.001$ ), and CAL (test: 0.8 mm CAL improvement at 3 months compared to 0.5 mm in the control group, $P<0.01$ ) and for a minority of bacterial species ( <i>Fusobacterium periodonticum</i> , <i>Prevotella nigrescens</i> , and <i>P. gingivalis</i> ) at 2 years   |
| Tomasi et al. [99]    | Intersubject parallel study | 32  | Chronic periodontitis                            | – Ultrasonic instrumentation alone<br>– Ultrasonic instrumentation + doxycycline   | 9 months  | The mean PD reduction at 3 months was 0.9 mm (95% CI: 0.6–1.2) in the control group and 1.0 mm (95% CI: 0.7–1.3) in the test group ( $P>0.05$ ). At 9 months, both treatment groups showed a mean PD reduction of 1.1 mm. The mean CAL gain was 0.6 mm at 3 months and approximately 0.8 at 9 months for both groups. Locally delivered doxycycline failed to improve the healing outcome of reinstrumentation of periodontal pockets showing a poor initial response to pocket/root debridement   |

(continued)

**Table 4.3** (continued)

| Authors                  | Type of study                           | No. patients | Type of disease   | Treatment   | Study period | Results   |
|--------------------------|---|--------------|---|---|--------------|---|
| Novak et al. [62]        | Multicenter Intersubject parallel study | 171          | Moderate to severe chronic periodontitis                | – HMT + TAT (in pockets $\geq 5$ mm) + SRP<br>– SRP + placebo                   | 6 months     | The experimental protocol of SRP + TAT + HMT showed significantly greater reductions in PD at 3 and 6 months than were seen with SRP + placebo. For PDs that were 4–6 mm at baseline, there were reductions in PD of 1.5 mm for the experimental group versus 0.9 mm for controls by 3 months ( $P < 0.01$ ) and 1.7 versus 1.2 mm, respectively, by 6 months ( $P < 0.01$ ). For PDs $\geq 7$ mm at baseline, significant reductions of 2.1 mm were noted in the experimental group versus 1.4 mm for controls   |
| Eickholz et al. [19]     | Multicenter Intersubject parallel study | 111          | Untreated or recurrent moderate to severe periodontitis | – SRP<br>– SRP + subgingival vehicle control (VEH)<br>– SRP + doxycycline gel   | 6 months     | SRP + DOX provided statistically significantly more favorable PD reduction (SRP: $-2.4 \pm 1.4$ mm, VEH: $-2.7 \pm 1.6$ mm, SRP + D: $-3.1 \pm 1.2$ mm; SRP versus SRP + D, $P = 0.0066$ ) and CAL gain (SRP: $1.6 \pm 1.9$ mm, VEH: $1.6 \pm 2.2$ mm, SRP + D: $2.0 \pm 1.7$ mm; SRP versus SRP + D, $P = 0.027$ , VEH versus SRP + D, $P = 0.038$ ) than SRP and VEH after 6 months   |
| Ratka-Krüger et al. [78] | Multicenter Intersubject parallel study | 110          |   | – SRP<br>– SRP + subgingival vehicle control (VEH)<br>– SRP + doxycycline gel   | 3 months     | At the end of the 6-month study period, the investigated microorganisms ( <i>A.actinomycetemcomitans</i> , <i>T.forsythus</i> , <i>P.gingivalis</i> , <i>T.denticola</i> ) were reduced in a highly significant manner in all three study groups, inter-group differences being most pronounced between the SRP and SRP + D treatment groups ( $P < 0.001$ )  |
| Eickholz et al. [20]     | Multicenter Intersubject parallel study | 37           | Residual or recurring systematic periodontal treatment  | – Subgingival application of a 14% doxycycline gel<br>– SRP with a sonic scaler | 6 months     | The two groups exhibited statistically significant ( $P < 0.001$ ) reductions of PD (DOX: $-1.43 \pm 0.22$ mm; SRP: $-1.14 \pm 0.18$ mm) and gains of CAL (DOX: $0.79 \pm 0.22$ mm; SRP: $0.72 \pm 0.19$ mm). Multilevel regression analyses, considering the therapy of different numbers of teeth in different patients, failed to show statistically significant differences concerning PD reduction and CAL gain between both therapies. For both therapies, PD reduction was significantly better in deeper pockets (PD $\geq 7$ mm) than in shallow pockets (PD 5–6 mm) |

CAL clinical attachment level, PD probing depth, BOP bleeding on probing, SRP scaling and root planning, MfW modified Widman flap surgical periodontal therapy, GCF gingival crevicular fluid, SRP + DOX SRP + doxycycline, CI confidence interval, TAT confidence interval, HMT systemically delivered doxycycline hydiate gel (10%), CHX chlorhexidine

Jorgensen et al. [40] failed to demonstrate that controlled-release doxycycline placed in moderate to deep periodontal pockets caused a significant additional reduction in the subgingival pathogenic microbiota (*A. actinomycetemcomitans*, *P. gingivalis*, *Dialister pneumosintes*, *T. forsythia*, *P. intermedia*, *Prevotella nigrescens*, *Campylobacter* sp., *Eubacterium* sp., *Fusobacterium* sp., *Peptostreptococcus micros*, *Eikenella corrodens*, *Staphylococcus* sp., enteric gram-negative rods, beta-hemolytic streptococci, and yeasts) compared to thorough SRP alone.

Application of the biodegradable sustained-release device after initial periodontal treatment resulted also in a significant gain in mean probing attachment levels and a significant reduction in pocket PDs in patients with peri-implantitis [9].

#### 4.2.4 Metronidazole

Metronidazole was introduced in the treatment of periodontal infections because this drug is accumulated by obligate anaerobic bacteria and leads to cell death by interfering with the synthesis of nucleic acids [75] (Fig. 4.6). Metronidazole is effective against gram-positive and gram-negative anaerobes, including *P. intermedia*, *P. gingivalis*, and *Fusobacterium* sp. [73].

**Elyzol 25%** (Elyzol Dental gel, Dumex GmbH, Bad Vilbel, Germany) dental gel is a suspension of metronidazole benzoate (40%) in a mixture of glyceryl mono-oleate and triglyceride (sesame oil) (Fig. 4.5). With a melting point at about 30°C, the gel flows and fills out the pocket after application. When in contact with the aqueous part of either gingival crevicular fluid or saliva, highly viscous fluid crystals are spontaneously formed in the gel. This prevents the gel from being easily washed out of the periodontal pocket and thereby provides the subgingival area with therapeutic

levels of metronidazole over a prolonged period of time [45]. The glyceryl mono-oleate is decomposed by lipases and the sparingly soluble metronidazole benzoate is slowly released into the gingival pocket. The esterases present in GCF hydrolyse the microbiologically inactive metronidazole benzoate to free metronidazole and benzoic acid [97].

The minimum inhibitory concentration (MIC) of metronidazole needed ( $\text{MIC}_{50}$ ) to affect strains relevant to periodontal pathology is below 1 µg/mL. It has been shown that after one application of a 25% metronidazole gel, the concentration obtained was higher than 1 µg/mL in all samples after 4 and 8 h, in 92% after 12 h, in 50% after 24 h, and in 8% after 36 h [96]. Thus, after 24 h, the metronidazole concentration in the crevicular fluid still remains above the MIC for 50% killing of key periodontal pathogens [75]. It was shown that the amount of gel matrix is generally cleared after 24 h of application, disappearing completely within a few days following application [97].

Table 4.4 summarizes the series of studies in which the metronidazole gel was used as an adjunct to SRP. Except for two studies [29, 63], there was no statistically significant difference between the treatments for any of the clinical parameters or in the microbiological data, when SRP was combined with the metronidazole gel application [29, 45, 46, 63, 66, 67, 82, 85, 94].

When compared to conventional SRP, a similar clinical effect was achieved by the metronidazole gel therapy, which was revealed as an alternative therapy to mechanical cleaning [5, 43, 44, 63, 72, 84, 93, 95].

The meta-analysis performed by Hung and Douglass [38] evaluated the treatment effect of 25% metronidazole gel treatment in 11 studies [5, 41–46, 63, 67, 76, 82, 84, 94, 95] and one study that tested the effect of 15% metronidazole gel [43]. Although the metronidazole gel treatments alone did not show a better improvement than SRP, most studies observed that there was an additional benefit from combined treatment of 25% metronidazole and SRP. The SRP alone resulted in a mean periodontal PD reduction of 0.60–1.98 mm, while the combined treatments had a mean periodontal PD reduction of 0.93–2.09 mm [38].

A later meta-analysis of Bonito et al. [8] on 11 studies of SRP plus locally delivered metronidazole revealed an overall effect size of 0.32 mm PD reduction and 0.12 mm gain in CAL favoring adjunctive local metronidazole.

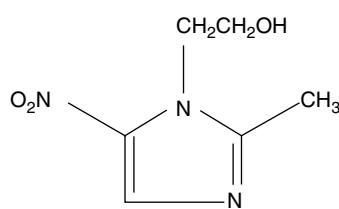


Fig. 4.6 The structural formula for metronidazole

**Table 4.4** Summary of clinical studies evaluating the local administration of metronidazole gel (Elyzole Dental Gel) in periodontal disease

| Authors               | Type of study                              | No. patients | Type of disease                             | Treatment   | Study period | Results   |
|-----------------------|--|--------------|---|---|--------------|---|
| Ainamo et al. [5]     | Multicenter Intersubject parallel study    | 206          | Adult periodontitis with teeth with PD≥5 mm | – SRP<br>– Local metronidazole gel  | 24 weeks     | The mean PD was 5.9 mm before gel application and 5.8 mm before scaling ( $P=0.31$ ). BOP was 88% in both treatment groups. 24 weeks after the treatment, PD and BOP were significantly reduced in both groups and for both parameters ( $P<0.01$ ). PD was reduced by 1.3 mm after gel application and 1.5 mm after scaling; BOP was reduced by 32% and 39%, respectively. The difference between the treatments was statistically significant, but considered as clinically unimportant             |
| Griffiths et al. [29] | Intrasubject split-mouth                   | 97           | Chronic periodontitis                       | – SRP<br>– SRP+Local metronidazole gel  | 9 months     | Both treatments effectively reduced the signs of periodontitis. At each follow-up visit, reduction in PD, CAL, and BOP after the combined treatment was greater than for SRP alone. At the end of the study, the mean reductions for PD were 1.0 mm (SRP) compared to 1.5 mm (SRP+gel), and for CAL they were 0.4 mm (SRP) compared to 0.8 mm (SRP+gel), with mean difference for CAL between treatments of $0.4\pm0.6$ mm (95% CI: 0.3–0.6 mm)   |
| Klinge et al. [43]    | Multicenter Intrasubject split-mouth study | 61           | Adult periodontitis with teeth with PD≥5 mm | 1. Local metronidazole 25% dental gel administered once a week for 2 weeks<br>2. Local metronidazole 15% dental gel applied once a week for 2 weeks<br>3. Local metronidazole 15% dental gel applied twice a week for 2 weeks<br>4. SRP performed once only | 12 weeks     | All three antibiotic treatments reduced the symptoms of periodontal pathology and yielded results comparable to those seen after subgingival scaling  |
| Knöfler et al. [44]   | Intrasubject split-mouth study             | 39           | Chronic periodontitis                       | – SRP<br>– Local metronidazole gel  | 6 months     | Both treatment procedures resulted in a gain of CAL: 0.67 mm for metronidazole and 0.50 mm for SRP ( $P<0.001$ ). The median PD was significantly reduced by 0.66 mm for metronidazole and 1.00 mm for SRP ( $P<0.001$ ) after 6 months. No change of aspartate aminotransferase in GCF was found. Alpha2-macroglobulin in GCF was significantly reduced for SRP and metronidazole after 3 and 6 months ( $P<0.001$ ). No significant difference was found between the two procedures at any variable |

|                     |                             |    |  |   |          |  |
|---------------------|-----------------------------|----|--|---|----------|--|
| Leiknes et al. [45] | Intrasubject split-mouth    | 21 | Recurring periodontitis with teeth with PD≥5 mm and BOP                  | - SRP<br>- SRP+local metronidazole gel  | 6 months | Both treatments yielded a statistically significant ( $P=0.001$ ) reduction in PD (1.9 and 1.8 mm), gain of CAL (1.6 and 1.0 mm), and reduction of BOP (38.1% and 33.3%) for test and control sites, respectively, at 6 months. There was no statistically significant difference between the treatments for any of the clinical parameters  |
| Lie et al. [46]     | Intrasubject split-mouth    | 18 | Moderate to severe chronic periodontitis with teeth with PD≥5 mm and BOP | - SRP<br>- SRP+local metronidazole gel<br>- SRP+3% tetracycline ointment  | 6 months | The average PD reduction for the three groups at 6 months was 1.5 mm and the average gain of CAL was 0.8 mm. There were no significant differences between the effects following topical application of the metronidazole gel or the tetracycline ointment. SRP alone appeared as effective as the drug-augmented regimens, although there was a weak but nonsignificant tendency for better results in sites treated with the antibiotic drugs. <i>Actinobacillus actinomycetemcomitans</i> was generally not detected; <i>Prevotella intermedia</i> was not significantly reduced, while <i>Porphyromonas gingivalis</i> was significantly reduced in all treatment groups |
| Noyan et al. [63]   | Intrasubject split-mouth    | 5  | Chronic periodontitis with teeth with PD≥5 mm                            | 1. SRP<br>2. Local metronidazole gel (E)<br>3. SRP+local metronidazole gel<br>4. Systemic metronidazole treatment<br>5. SRP+systemic metronidazole<br>6. No treatment             | 7 weeks  | All treatments resulted in clinical improvements (GI, PD, CAL) except for the untreated group. Parallel to the clinical changes, all treatments reduced the number of total bacteria and proportions of obligately anaerobic microorganisms. Local metronidazole in combination with SRP seems to be more effective in terms of producing both clinical (mean PD reduction: 2.09 mm vs SRP: 1.31 mm and E: 1.41 mm, mean CAL gain: 1.25 mm vs SRP: 0.59 mm and E: 0.63 mm) and microbial improvements  |
| Palmer et al. [66]  | Intersubject parallel study | 90 | Moderate to advanced periodontitis                                       | 1. SRP (subgingival scaling using ultrasonic scalers)<br>2. SRP+systemic metronidazole<br>3. SRP+local metronidazole gel (two applications of 25% metronidazole gel 1 week apart) | 6 months | Mean PD was reduced following treatment by greater than 1.6 mm (Group 1 = 1.68 mm, Group 2 = 1.62 mm, Group 3 = 1.74 mm at 6 months post treatment) but no significant differences were detected between treatment groups at any time point. Similarly, no significant differences were detectable between treatments for changes in mean CAL, BOP, PI, or proportions of bacterial morphotypes  |

(continued)

**Table 4.4** (continued)

| Authors                | Type of study                  | No. patients                 | Type of disease   | Treatment   | Study period | Results   |
|------------------------|--------------------------------|------------------------------|---|---|--------------|---|
| Palmer et al. [67]     | Intersubject parallel study    | 28 Smokers and 56 nonsmokers | Moderate to advanced periodontitis                        | 1. SRP (subgingival scaling using ultrasonic scalers)<br>2. SRP + systemic metronidazole<br>3. SRP + local metronidazole gel (two applications of 25% metronidazole gel 1 week apart) | 6 months     | There were no differences in any clinical measures in response to the three treatment regimens at 2 or 6 months for either smokers or nonsmokers. Multiple linear regression analysis on PD at 6 months demonstrated that smoking was a significant explanatory factor ( $P < 0.001$ ) for poor treatment outcome, whilst the presence or absence of adjunctive metronidazole was not ( $P = 0.620$ )   |
| Pedrazzoli et al. [72] | Intrasubject split-mouth study | 24                           | Chronic periodontitis                                     | - SRP<br>- Local metronidazole gel  | 6 months     | Both treatments were effective in reducing PD and BOP. Metronidazole tended to be a little better than scaling during the study period and the clinical effects of both treatments persisted during the whole 6-month observation period. Local metronidazole treatment induced a significant and longlasting shift in the subgingival flora towards a composition more compatible with health and comparable to that obtained by mechanical debridement. Proportions of black-pigmented anaerobes including <i>P. intermedia</i> and the number of spirochetes were significantly reduced after both treatments with a concomitant increase in the proportions of streptococci         |
| Riep et al. [82]       | Intrasubject split-mouth       | 30                           | Recurrent periodontitis with teeth with PD > 6 mm and BOP | - SRP<br>- SRP + local metronidazole gel  | 3 months     | At the final examination visit, PD reduction was $1.7 \pm 0.9$ mm after test and control treatment which was statistically highly significant ( $P < 0.001$ ). Furthermore, CAL gain was statistically highly significant ( $P < 0.001$ ) with $1.1 \pm 0.8$ mm after SRP and $1.3 \pm 0.8$ mm after adjunctive metronidazole application. There was no statistically significant difference between test and control site. <i>P. gingivalis</i> was statistically significantly reduced after both treatments with no statistically significant difference between test and control. The level of <i>A. actinomycetemcomitans</i> was not changed significantly after either treatment |
| Rudhart et al. [84]    | Intrasubject split-mouth       | 46 On maintenance therapy    | Recurrent periodontitis with teeth with $PD \geq 5$ mm    | - SRP<br>- Local metronidazole gel (two applications of 25% metronidazole gel 1 week apart)   | 6 months     | Both treatments resulted in PD reduction and CAL gain. PD reduction was statistically significant ( $P < 0.01$ ) for both treatment modalities after 6 months. The CAL gain was not significant for either treatment. There was no statistical significance between scaling and antibiotic therapy. <i>T. denticola</i> , <i>P. gingivalis</i> , and <i>P. intermedia</i> were significantly reduced after therapy; however, there were no statistically significant differences between treatments. If <i>A. actinomycetemcomitans</i> was present before therapy, it was also present after treatment in both groups  |

|                                 |                             |    |  |   |           |  |
|---------------------------------|-----------------------------|----|--|---|-----------|--|
| Salvi et al.<br>[85]            | Intrasubject parallel study | 47 | Adult periodontitis                                | - SRP+doxycycline<br>- SRP+local metronidazole gel<br>- SRP+PerioChip | 18 weeks  | Between the baseline and 18-week examinations, subjects treated with Atridox showed a significantly greater gain in mean CAL of $0.33 \pm 0.09$ mm than subjects treated with Elyzol Dental Gel ( $0.03 \pm 0.09$ mm) ( $P = 0.03$ ). However, the gain in CAL of $0.16 \pm 0.10$ mm found after PerioChip application did not differ significantly from that obtained following the application of Atridox ( $P = 0.27$ ). Of the sites treated with Atridox, 42% gained $\geq 1$ mm CAL and 9% $\geq 2$ mm CAL as opposed to the sites treated with Elyzol Dental Gel, in which 34% gained $\geq 1$ mm CAL and 8% gained $\geq 2$ mm CAL. Of the sites treated with PerioChip, 36% gained $\geq 1$ mm and 6% gained $\geq 2$ mm CAL following a completed initial periodontal therapy. |
| Stelzel & Flores-de-Jacoby [95] | Intrasubject split-mouth    | 30 | On maintenance therapy                             | Recurrent periodontitis with teeth with PD $\geq 5$ mm                | 24 weeks  | At the end of the 6-month follow-up period, the mean reduction in PD was 1.3 mm after gel treatment and 1.5 mm after subgingival scaling. BOP was reduced by 35% and 42%, respectively. No significant differences between the two treatments were detected  |
| Stelzel & Flores-de-Jacoby [93] | Intrasubject split-mouth    | 24 | On maintenance therapy                             | Recurrent periodontitis with teeth with PD $\geq 5$ mm                | 24 months | The average reduction in PD was 1.3 mm in the gel group and 1.5 mm in the scaling group, with the tendency to bleeding being reduced by ~50% in both groups. After 24 months, improvements of 0.6 and 0.5 mm, respectively, were observed in PD. No statistically significant differences were observed between the two methods  |
| Stelzel & Flores-de-Jacoby [94] | Intrasubject split-mouth    | 59 | Adult periodontitis with teeth with PD $\geq 5$ mm | - SRP<br>- SRP+local metronidazole gel                                | 259 days  | Comparison of the two treatments revealed a statistically significant improvement in the clinical parameters for both treatment methods over the study period. Between baseline and day 259, significant differences in PD (SRP+Metro: from 6.00 to 4.63 mm, SRP: from 6.02 to 4.83 mm) and BOP (SRP + Metro: from 67% to 31%, SRP: from 64% to 36%) were observed between the two treatment groups. Evaluation according to different patient groups demonstrated significant advantages of the combined therapy in previously untreated patients   |

CAL clinical attachment level, PD probing depth, BOP bleeding on probing, SRP scaling and root planning, M/FW modified Widman flap surgical periodontal therapy, GCF gingival crevicular fluid, SRP + DOX SRP+doxycycline, CI confidence interval, TAT systematically delivered doxycycline hyclate (HMT; 20 mg, twice a day), HMT locally delivered doxycycline hyclate gel (10%), CHX chlorhexidine

The difficulty of performing adequate debridement in furcations by mechanical means has prompted experimentation with chemotherapeutic agents in these areas [11]. Needleman and Watts [60] tested the adjunctive effect of 1% metronidazole gel irrigation into furcation areas with class II and III involvements during periodontal maintenance with subgingival scaling. Clinically, no further improvement was seen for the furcations treated with metronidazole. Likewise, lack of adjunctive effect exerted by the metronidazole gel was reported for proportions of spirochetes, motile rods, and cocci observed with dark-field microscopy [11].

Two other slow-release systems (an acrylic strip with a 50% weight/weight incorporation of the antimicrobial agent and a 95% collagen/5% metronidazole device) were described [1, 2, 35, 108]. The water-soluble discs with 80% metronidazole appeared to be a safe adjunct to SRP in slowing periodontal disease progression [68]. The choice of collagen as a delivery system presented several advantages, especially in tolerance, with no injury to inflammatory reaction. Hitzig et al. [35] showed that a single subgingival application of 5% metronidazole in a collagen carrier can be effective, when associated with debridement, in the treatment of adult periodontitis.

Recently, metronidazole-loaded 50/50 poly(DL-lactide-co-glycolide) (PDLGA), 75/25 PDLGA, and poly(DL-lactic acid) (PDLLA) films have been developed. These films were designed to be inserted into the periodontal pocket and treat infections with controlled-release metronidazole for  $\geq 1$  month. The drug released from films loaded with 10% w/w metronidazole resulted in a significant decrease in bacterial viability within several days. When exposed to human gingival fibroblasts in cell culture conditions, these films maintained their normal fibroblastic features [87].

### 4.3 Comparison of Treatment Methods

Most studies have tested a single form of local drug delivery or systemic administration instead of comparing various forms of therapy. Understandably, developers and distributors have the primary interest to register and promote their own product for the broadest possible usage, and not to differentiate specific benefits or shortcomings of various applications [58].

The efficacy of three commercially delivered systems as adjuncts to SRP was assessed in four studies [31, 41, 76, 85] (Table 4.5).

Unfortunately, data were combined from studies exploring various modes of local treatment, including irrigation, impregnated strips, and pastes. One cannot exclude, however, that the differences noted between the drugs primarily reflect differences in modes of application and study populations, not the potency of the agent [58].

Few studies have addressed the problem of incorporating local or systemic antimicrobial therapy into an overall treatment strategy [58] (Table 4.6). A key issue requiring clarification refers to the selection of a local or a systemic delivery approach whenever the use of an antibiotic is indicated [58]. Akalin et al. [6] compared the clinical efficacies of systemic and local doxycycline in the treatment of chronic periodontitis. Forty-five patients were studied in three main groups with five treatments: (1) systemic doxycycline alone, (2) systemic doxycycline + SRP, (3) local doxycycline alone, (4) local doxycycline + SRP, and (5) SRP alone. Antibiotic-treated patients were given doxycycline treatment alone in one quadrant of their upper jaws, and doxycycline + SRP was given in the contralateral quadrant. PD, CAL, gingival index, sulcular bleeding index, and plaque index values were recorded at baseline and at week 7. The systemic and local doxycycline treatments alone provided significant clinical healing. The local doxycycline treatment provided significantly higher PD reduction than the systemic doxycycline treatment ( $P < 0.05$ ). No significant difference was found between the systemic doxycycline + SRP and the local doxycycline + SRP. There was no significant difference between systemic doxycycline + SRP and SRP alone treatment ( $P > 0.05$ ), and between local doxycycline and SRP treatment. The local doxycycline alone treatment seemed more effective than systemic doxycycline alone treatment on PD reduction, but no significant difference was found between them when combined with SRP. It was suggested that local doxycycline may be more preferable than systemic doxycycline as an adjunct to mechanical treatment since local doxycycline seems more effective than systemic doxycycline on PD reduction and does not have the side effects of systemic doxycycline. Three studies [63, 66, 67] revealed no differences in any clinical measure in response to the local or systemic metronidazole treatment regimens.

**Table 4.5** Comparison of different treatment methods of local antibiotics in periodontal therapy

| Authors              | Type of study               | No. patients | Type of disease  | Treatment   | Study period | Results   |
|----------------------|-----------------------------|--------------|--|---|--------------|---|
| Radvar et al. [76]   | Parallel study              | 54           | Persistent periodontal pockets ≥ 5 mm and BOP and/or suppuration | - SRP<br>- SRP + 25% tetracycline fibers<br>- SRP + 2% minocycline gel<br>- SRP + 25% metronidazole gel       | 6 weeks      | The improvements in clinical parameters were greater in all three adjunctive treatment groups than SRP alone. The mean PD reductions for SRP + minocycline were 0.87 mm   |
| Kinane & Radvar [41] | Parallel study              | 79           | Persistent periodontal pockets ≥ 5 mm and BOP and/or suppuration | - SRP alone<br>- SRP + 25% tetracycline fibers<br>- SRP + 2% minocycline gel<br>- SRP + 25% metronidazole gel | 6 months     | All 4 therapies resulted in significant improvements from baseline in PD, CAL, BOP, and the Modified Gingival Index scores. The improvements in clinical parameters were greater in all three adjunctive treatment groups than SRP alone. The mean PD reductions at 6 months were for SRP + minocycline = 1.10 mm   |
| Salvi et al. [85]    | Intersubject parallel study | 47           | Adult periodontitis  | - SRP + doxycycline<br>- SRP + local metronidazole gel<br>- SRP + PerioChip                                   | 18 weeks     | Between the baseline and 18-week examinations, subjects treated with Atridox showed a significantly greater gain in mean CAL of $0.33 \pm 0.09$ mm than subjects treated with Elyzol Dental Gel ( $0.03 \pm 0.03$ ) ( $P = 0.03$ ). However, the gain in CAL of $0.16 \pm 0.10$ mm found after PerioChip application did not differ significantly from that obtained following the application of Atridox ( $P = 0.27$ ). Of the sites treated with Atridox, 42% gained ≥ 1 mm CAL and 9% ≥ 2 mm CAL as opposed to the sites treated with Elyzol Dental Gel, in which 34% gained ≥ 1 mm CAL and 8% gained ≥ 2 mm CAL. Of the sites treated with PerioChip, 36% gained ≥ 1 mm and 6% gained ≥ 2 mm CAL following a completed initial periodontal therapy |
| Gupta et al. [31]    | Intersubject parallel study | 30           | Moderate to advanced chronic periodontitis                       | - SRP<br>- SRP + doxycycline<br>- SR + chlorhexidine gel  | 3 months     | All treatments showed significant reductions in PD and CAL at 1 and 3 months when compared to baseline values ( $P < 0.001$ ). At 3 months, sites treated with SRP + DOX and SRP + CHX showed an additional reduction in PD of $0.86 \pm 1.0$ mm and $0.66 \pm 1.58$ mm, respectively, significantly greater than SRP alone ( $P < 0.02$ ). Differences in mean PD reduction between SRP + DOX and SRP + CHX were not significant ( $P = 0.46$ ). At 3 months, differences in relative CAL between both SRP + DOX ( $0.80 \pm 0.92$ ) and SRP + CHX ( $0.63 \pm 1.47$ ) and SRP alone were statistically significant ( $P < 0.02$ ). Differences in relative CAL between SRP + DOX and SRP + CHX were not significant ( $P = 0.54$ )                    |

CAL clinical attachment level, PD probing depth, BOP bleeding on probing, SRP scaling and root planning, MFW modified Widman flap surgical periodontal therapy, GCF gingival crevicular fluid, SRP + DOX SRP + doxycycline, CI confidence interval, TAT systemic locally delivered doxycycline hydrcate (HMT; 20 mg, twice a day), HMT locally delivered doxycycline hydrcate (10%), CHX chlorhexidine

**Table 4.6** Comparison of treatment methods of local and systemic antibiotics in periodontal therapy

| Authors              | Type of study                                       | No. patients                 | Type of disease                                 | Treatment   | Study period | Results   |
|----------------------|---|------------------------------|---|---|--------------|---|
| Noyan et al. [63]    | Intrasubject split-mouth                            | 5                            | Chronic periodontitis with teeth with PD ≥ 5 mm | 1. SRP<br>2. Local metronidazole gel (E)<br>3. SRP+local metronidazole gel<br>4. Systemic metronidazole treatment<br>5. SRP+systemic metronidazole<br>6. No treatment             | 7 weeks      | All treatments resulted in clinical improvements (GI, PD, CAL) except for the untreated group. Parallel to the clinical changes, all treatments reduced the number of total bacteria and proportions of obligately anaerobic microorganisms. Local metronidazole in combination with SRP seems to be more effective in terms of producing both clinical (mean PD reduction: 2.09 mm vs SRP: 1.31 mm and E: 1.41/mm, mean CAL gain: 1.25 mm vs SRP: 0.59 mm and E: 0.63 mm) and microbial improvements   |
| Palmer et al. [66]   | Intersubject parallel study                         | 90                           | Refractory periodontitis                        | 1. SRP (subgingival scaling using ultrasonic scalers)<br>2. SRP+systemic metronidazole<br>3. SRP+local metronidazole gel (two applications of 25% metronidazole gel 1 week apart) | 6 months     | Mean PDs were reduced following treatment by greater than 1.6 mm (Group 1=1.68 mm, Group 2=1.62 mm, and Group 3=1.74 mm at 6 months post treatment) but no significant differences were detected between treatment groups at any time point. Similarly, no significant differences were detectable between treatments for changes in mean CAL, BOP, PI, or proportions of bacterial morphotypes   |
| Palmer et al. [67]   | Intersubject parallel study                         | 28 Smokers and 56 nonsmokers | Moderate to advanced periodontitis              | 1. SRP (subgingival scaling using ultrasonic scalers)<br>2. SRP+systemic metronidazole<br>3. SRP+local metronidazole gel (two applications of 25% metronidazole gel 1 week apart) | 6 months     | There were no differences in any clinical measure in response to the three treatment regimens at 2 or 6 months for either smokers or nonsmokers. Multiple linear regression analysis on PD at 6 months demonstrated that smoking was a significant explanatory factor ( $P<0.0001$ ) for poor treatment outcome, whilst the presence or absence of adjunctive metronidazole was not ( $P=0.620$ )   |
| Punucker et al. [74] | Intersubject parallel study                         | 30                           | Generalized aggressive periodontitis            | – SRP+systemic amoxicillin/clavulanic acid<br>– SRP+local tetracycline fiber in pockets with PD ≥ 5 mm  | 9 months     | In both treatment groups, PD decreased significantly in the 9-month period (6.2±1.5 mm to 4.7±1.4 mm for SRP + T and 6.5±1.4 mm to 4.2±0.6 mm for SRP + A). Similarly, in both treatment groups, there were small but significant gains in CAL in the 9-month period (12.0±1.8 mm to 11.3±1.8 mm for SRP + T and 12.3±1.5 to 11.2±1.2 mm for SRP + A). There was no statistically significant difference between the two groups in pocket reduction and CAL gain. At the final examination, both groups showed significant PD reduction and CAL gain ( $P<0.0001$ ) compared to baseline. The frequency and percentage of bleeding sites decreased significantly in both groups. At week 54, this decrease was significantly greater in the SRP + A group (31.67% for SRP + T versus 3.85% for SRP + A) |
| Akalin et al. [6]    | 3 Study groups with intrasubject split-mouth design | 45                           | Chronic periodontitis                           | – Systemic doxycycline<br>– Systemic doxycycline+SRP<br>– Doxycycline local<br>– SRP+doxycycline<br>– SRP alone   | 7 weeks      | No significant difference was found between local doxycycline and SRP treatments. Significant PD reduction between local doxycycline alone (from 0.50±0.13 to 0.34±0.09 mm) and systemic doxycycline (from 0.72±0.19 to 0.49±0.13 mm) ( $P<0.05$ )  |

CAL clinical attachment level, PD probing depth, BOP bleeding on probing, SRP scaling and root planning, MFW modified Widman flap surgical periodontal therapy, GCF gingival crevicular fluid, SRP + DOX SRP+doxycycline, CI confidence interval, TAT systemically delivered doxycycline hydiate (HMT; 20 mg, twice a day), HMT locally delivered doxycycline hydiate gel (10%), CHX chlorhexidine

#### 4.4 Antimicrobial Effects of Local Delivery Agents

The benefit of local antimicrobial therapy is the higher concentration of an antimicrobial agent which can be attained in subgingival sites compared with a systemic drug regimen [103]. Several studies have used local antimicrobial agents as an adjunct to SRP, and evaluated the microbiological effects of subgingivally administered antimicrobial agents in chronic periodontitis patients [36, 39, 46, 47, 72, 82, 84, 91, 93, 98, 107, 109, 114, 116].

Although most studies showed that antibiotic therapy combined with mechanical treatment was equally effective to conventional SRP, there was little or no difference between the two treatment modalities in the numbers/incidence of periodontal pathogens. However, most of these studies showed only a transient reduction of periodontal pathogens, and may not provide a compelling reason for routine use of antimicrobial agents in conjunction with root debridement [103].

#### 4.5 Appropriate Time to Employ Local Drug Delivery: Active Versus Maintenance Therapy

The question arises as to the most opportune time to use local drug delivery: during active (initial) therapy or maintenance procedures? When employed as an adjunct to active therapy, numerous studies indicated that there was a statistically significant improvement with respect to PD reductions and/or gains of clinical attachment. However, differences between test and control groups were often noted to be limited to tenths of millimeters. This was also observed in large clinical trials (>100 participants) and in meta-analyses. In addition, the necessary number to treat (NNT=the number of persons or sites that must be treated in the test group during a given time period to achieve one additional treatment result or to prevent one adverse incident compared to the control group) determinations indicated that a substantial effort may be needed with combined therapy to attain an additional site with a ≥2-mm PD reduction compared to SRP alone. Furthermore, it appears that many patients respond to

conventional treatment; therefore, the routine application of adjunctive chemotherapy during active therapy may not be advantageous. In contrast, numerous investigators have beneficially employed local delivery at locations in maintenance patients who were not responding to SRP alone [28].

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## The Use of Chemical Supragingival Plaque Control in Periodontal Therapy

According to the classic version of the nonspecific theory in microbial etiology of inflammatory periodontal disease, the indigenous oral bacteria, in the absence of oral hygiene, colonize the gingival crevice to form plaque. Inflammatory periodontal disease develops in the case of bacterial proliferation above the threshold of host resistance, caused by the combined biologic effects of the total plaque flora. All plaque bacteria are thought to have some of the various virulence factors causing gingival inflammation and periodontal destruction, and it is implied that plaque will cause disease regardless of its composition. Therefore, total plaque control is considered necessary in treatment and prevention, and this approach has certainly proved effective, where it can be carried out [252].

The principle of prevention of periodontal disease by methods of supragingival plaque control is well established. Hence, from the epidemiological and behavioral data it could be concluded that compliance to regular mechanical oral hygiene is relatively poor. Consequently, antiplaque agents should be used to augment mechanical plaque control [261].

Some terms have previously been defined by the 2nd European Workshop on Periodontology in 1996 [6].

- Antimicrobial agents: chemicals that have a bacteriostatic or bactericidal effect *in vitro* that alone cannot be extrapolated to a proven efficacy *in vivo* against plaque
- Plaque reducing/inhibitory agents: chemicals that have only been shown to reduce the quantity and/or affect the quality of plaque, which may or may not be sufficient to influence gingivitis and/or caries
- Antiplaque agents: chemicals that have an effect on plaque sufficient to benefit gingivitis and/or caries

- Antigingivitis agents: chemicals which reduce gingival inflammation without necessarily influencing bacterial plaque (includes antiinflammatory agents)

As a result of the confusion regarding this terminology for both clinicians and the public alike, it has been suggested that chemicals/products could be grouped into one of the three categories based on their individual properties [66, 167].

- Group A agents were described as antiplaque (by definition, chemicals that inhibit plaque formation to such an extent that they prevent the development of gingivitis). These chemicals include chlorhexidine, acidified sodium chlorate, salifluor, and delmopinol. The efficacy of these chemicals is reflected in that in mouthwash form they can be used instead of conventional mechanical plaque removal such as when the individual cannot effectively clean his or her teeth.
- Group B agents include cetylpyridinium chloride, essential oil and triclosan rinses. These rinses should be used as adjuncts to mechanical cleaning, such as toothbrushing, and are termed plaque inhibitory.
- Group C (low to moderate activity) are chemicals/rinses with little or no effect on plaque accumulation and would be expected to have a largely cosmetic role, such as breath freshening. Rinses in this group include products containing sanguinarine, oxygenating agents, and rinses containing the saturated pyrimidine, hexetidine. The use of such a classification may help the clinician in recommending appropriate mouth rinses for specific clinical situations. However, as time goes by it has become apparent that the results of some newer clinical studies have repositioned specific

chemicals into a different category, such as positioning essential oils in and hexetidine in group A. Although this attempt to classify chemicals in terms of efficacy and recommended usage would be useful, it can be seen that there is still confusion as to where to position individual products in terms of efficacy [167].

## 5.1 Delivery Methods Vehicles for Periodontal Health Benefits

The main delivery routes for administration of agents for the control of supra- and subgingival plaque formation and/or for control of inflammation and tissue breakdown includes: mouthrinse; gel; toothpaste; floss, toothpicks or interdental devices; lozenges or tablets; irrigators; slow-release devices [55].

### 5.1.1 Mouth rinses

Mouth rinses are frequently used by the public for social reasons, such as improving the breath and freshening of the mouth. Other indications are more focused on preventing oral problems, such as dental caries, and inhibiting plaque formation and chronic gingivitis. It is perceived that because mechanical control of plaque by most individuals is often inadequate, antiplaque or plaque-inhibitory home use mouth rinses would be of value in reducing plaque missed by toothbrushing [167]. The in-office use of mouth rinses prior to and after mechanical debridement may reduce the levels of the periodontal pathogens in the oral cavity and reduce the risk of bacteremia [267].

Regarding the control of plaque and gingivitis, mouthwashes effectively reach all inaccessible areas of the mouth may be very much dependent on the ability of individuals to rinse effectively. For mentally and physically handicapped patients, the ability to rinse effectively may be severely compromised and direct application of chemical antiplaque agents would be more appropriate. It is also acknowledged that mouthwashes have little penetration into the subgingival environment, but they reach very well the interproximal areas, as a common problem with all interdental

cleaning aids is patient dexterity and motivation [167] and references therein; [269].

Generally, it has been recommended that mouth rinsing twice a day (morning and evening) would be the ideal regimen in controlling plaque; this is partly based on studies with chlorhexidine and essential oil rinses, which has been reported to be present in the mouth for up to 12 h following a single rinse. For acetyl pyridinium chloride, increasing the rinsing frequency to four times a day has been suggested to produce efficacy equivalent to that of chlorhexidine [28, 29, 141, 167].

Presently, it is not known whether sporadic use of mouth rinses, such as once a day, once a week, or three times a week, would produce any benefit. The recommendations of mouthwash manufacturers indicate that chlorhexidine may be used over short, sharp periods (such as 5 to 7 days) for mild gingivitis, continuing for up to 31 days for chronic periodontitis, the number of days of use thus being dependent on the severity of disease present. The more recent introduction of commercially available lower-concentration chlorhexidine mouth rinses (0.06%) containing fluoride could be more acceptable for daily use over longer periods and not just over the short to medium term, which has been the suggested norm for chlorhexidine products [6, 167].

Generally speaking, mouth rinses and their constituents are safe to use but over the years some issues raising concern have arisen. These include increased risk of developing precancerous leukoplakia associated with sanguinarine [151, 152], potential enamel erosions produced by the essential oil and hexetidine rinses [194], the potential of fluoride rinses to cause both systemic toxicity and dental fluorosis [10], and, of course, other issues involving the inclusion of alcohol in mouth rinses. This was added (i) to solubilize antimicrobial compounds in order to make them bioavailable; (ii) to solubilize flavor-masking agents; (iii) to improve the shelf life of the mouth rinse, and to some extent improve the pleasurable characteristics of mouth rinsing [167].

Recently there has been an increasing demand for alcohol-free mouth rinses, as significant quantities of alcohol in their content (10–20%) may have a number of possible disadvantages:

1. Alcohol toxicity in case it is accidentally swallowed particularly by young children [66]
2. Increased risk of developing oral and pharyngeal cancer presents weak, inconsistent, and even

contradictory evidence in the literature [32, 37, 128, 132, 156, 284]. The following statement was approved by the Board of Directors of The International Academy of Periodontology at their meeting in Miami Beach, Florida, on April 3, 2009 (<http://www.perioiap.org/alcohol.htm>) : The International Academy of Periodontology is in agreement with the following positions of the British Dental Health Foundation Statement: “A recent, and more thorough review of all available evidence carried out by leading experts on behalf of the foundation concluded there were no proven links between alcohol-containing mouthwashes and increased incidence of mouth cancer. The public should not worry.” and the *British Dental Association Statement*: “Excessive consumption of alcohol and tobacco are well recognized in the UK as risk factors for developing oral cancers. This paper raises interesting issues but the evidence showing any link between the prolonged use of mouthwashes containing alcohol and oral cancer is not conclusive. Further research is required to establish if there is a genuine connection. “Where patients are in any doubt about using mouthwash, they should consult their dentist” (<http://www.perioiap.org/alcohol01.htm>, Accessed on 16.05.2010).

3. Oral mucosa discomfort increases linearly with increasing concentrations of alcohol as certain individuals are sensitive to alcohol [26].
4. The use of alcohol containing mouthwashes may increase transiently the alcohol content of exhaled breath and could thus change the readings of the police breath test [167].
5. Softening and color altering effect of composite and hybrid resin restorations [98, 157, 158, 190, 276].
6. Religious objections were also rinsed [167].

The presence of alcohol in mouth rinses is contraindicated for patients with mucositis, patients with sensitive tissues associated with head and neck radiation therapy, immunocompromised patients, patients sensitive to alcohol, and patients with composite restorations. Another significant group of patients for whom alcohol-containing mouth rinses are contraindicated are the patients who have undergone radiation therapy for head and neck cancer. Head and neck radiation is known to cause xerostomia, ulcerating gingivitis, and tissue damage, which result in extreme sensitivity in these areas. Alcohol-containing mouth rinses may further exacerbate these conditions [65].

Several studies have revealed the efficacy of the alcohol-free mouth rinses [65, 135].

### 5.1.2 Gels

The most common gel is a simple thickened aqueous system containing humectant but neither abrasive nor foaming agents [54]. Among other vehicles, gels have become available with CHX concentrations of about five to 15 times those of chlorhexidine solutions. Moreover, due to the high viscosity of a gel, one would expect a lower clearance of the active agent from the periodontal pocket, thereby further promoting pharmacotherapeutic effects [45]. Frequent applications are necessary as after 3 h, the percentage of live bacteria achieved with the gel and sprays was similar to the basal figures (80–91%). At 7 h, bacterial vitality recovered basal percentages [84]. Nevertheless, there seem to be no data available indicating that treatment outcome of scaling and root planning (SRP) will benefit from the adjunctive subgingival administration of a chlorhexidine gel. It appears that gel vehicles are not suitable as compensatory aids when SRP become less effective due to limited access [45]. Staining and mucosal erosion can occur with chlorhexidine gel use [10].

### 5.1.3 Dentifrices

Tooth pastes are complex delivery vehicles because they are designed to provide multiple functions: minimizing build-up of plaque, strengthening teeth against caries, cleaning the teeth by removing stain, food debris, and freshening up of the mouth [55, 76].

A toothpaste contains a number of ingredients that serve a definite purpose in providing the attributes required by the consumer in a modern product:

1. A *polishing or abrasive agent* has two purposes. Firstly, its mild abrasive action helps to eliminate plaque from the teeth, hence reducing plaque buildup. Secondly, the abrasive agent removes stained pellicle from the teeth, polishes the surfaces, restores the natural luster, and enhances enamel whiteness. The abrasive system should be insoluble, inert, nontoxic, and preferably white. Commonly

used abrasive materials include calcium carbonate, dicalcium phosphate dihydrate, alumina, and silicas. In gel toothpaste, the abrasive system is usually a special porous silica that becomes transparent when blended into the gel system. Transparency is achieved when the refractive index of the soluble portion of the formula matches that of the silica. In this way it is possible to produce a clear gel containing an insoluble polishing agent [76].

2. *The binder or thickener controls* the stability and consistency of a toothpaste, and also affects the ease of dispersion of the paste in the mouth. Choice of the correct binder and concentration is critical to ensure that the product can be readily squeezed from the tube and yet have a good appearance when it is on the toothbrush. Commonly used thickeners can be divided into two classes – water soluble, including carrageenans, alginates, and sodium carboxymethylcellulose, and water insoluble, including magnesium aluminum silicate, sodium magnesium silicate, and colloidal silica [76].
3. *The surfactant agent* provides the foam that eases the removal of food debris and aids dispersion of the product in the mouth. The detergent used most widely by all major manufacturers is sodium lauryl sulfate, which has a history of decades of safe and effective use all over the world. Sodium lauryl sulfate also has antimicrobial properties in its own right and thus helps to preserve the toothpaste during manufacture and use. Another positive role that sodium lauryl sulfate has is to help solubilize key ingredients such as flavors and certain antimicrobial agents [76].
4. *A humectant* is a material that helps to reduce the loss of moisture from a preparation. In toothpaste, the humectant minimizes plug formation in tube nozzles and improves the texture and feel of the product in the mouth. It can also act as a sweetening agent. Examples are glycerin, sorbitol, and polyethylene glycol. Glycerin and sorbitol are the humectants used most often [76].
5. The choice of *flavor* is very important to the consumer. It renders the product pleasant to use and should leave a fresh taste in the mouth after use. Soluble saccharin is usually added as an additional sweetening agent. The types of flavor used include peppermint oil, spearmint oil, and wintergreen (methyl salicylate). All around the world, pepper-

mint and spearmint are the most important flavor types. Other flavor materials such as wintergreen, aniseed, lemon oil, and eucalyptus are also usually added to improve the acceptability of the flavor and to add individual notes to the flavors, which can be important in certain types of toothpaste, such as medicinal formulations [76].

6. A toothpaste is an excellent vehicle for delivering other *oral health benefits*, and for this reason many different therapeutic agents are added. These include anticaries agents, antiplaque agents, antitartrar agents, antisensitivity agents, and whitening agents [76].
7. Other ingredients can include titanium dioxide to whiten the appearance of the product and possibly a preservative (such as benzoates) to ensure that microorganisms do not grow in the paste [76].
8. Using a toothbrush as a delivery device, it was found that toothpastes can penetrate only up to 0.9 mm into the periodontal pockets [268].

It appears that toothpastes have the potential to harm the dental hard tissues by virtue of their degree of abrasivity and could play a role in localizing sites of dentine hypersensitivity, either by cooperating with the toothbrush in gingival recession or with erosion in removing enamel at the cervical areas. Further side effects of toothpastes that have been reported in clinical trials are allergic reactions and extrinsic tooth staining and increased supragingival calculus associated with the use of chlorhexidine-containing toothpaste (for review see [10]).

Toothpastes containing triclosan/copolymer and triclosan/zinc citrate improve plaque control and gingival health, both safely and effectively, in studies of 6 months duration. However, the data supporting the effectiveness of triclosan/pyrophosphate are weak. Stannous fluoride toothpastes have been inconsistent in their effect on dental plaque but have consistently improved gingival health. Their use, however, is accompanied by staining of the teeth. The data on toothpastes containing zinc citrate and amine fluoride/stannous fluoride are insufficient to make firm recommendations regarding their efficacy [60]. Recently, Paraskevas et al. [187, 188] revealed that the use of dentifrice does not contribute to the instant mechanical plaque removal during manual toothbrushing. A higher dentifrice

abrasivity does not seem to contribute to increased plaque removal with a manual toothbrush. It appears that the mechanical action provided by the use of a toothbrush is the main factor in the plaque-removing process [187, 188].

#### **5.1.4 Chewing-Gums**

Chewing gums have been studied and used as delivery vehicles for a host of dental substances such as calcium, bicarbonate, carbamide, chlorhexidine, fluoride and polyol sweeteners, as well as medicinal substances such as nicotine, methadone, aspirin, motion sickness antihistamine agents, antifungal agents, caffeine, and vitamins. Chewing polyol-sweetened gum particularly xylitol-containing gum, alone or in combination with other dental-protective substances in oral health and caries-prevention programs for high-risk populations have the potential to significantly improve oral health status (for review see [140]).

#### **5.1.5 Varnishes**

A varnish is a polymer-based matrix that slowly releases an agent onto the (tooth) surface to which it is applied and also to saliva [55]. Since 1964, varnishes have been used for local delivery of fluor and are reported to be an effective and easy to use vehicle. During the past decade, varnishes for local delivery of antimicrobial agents such as chlorhexidine have been developed and investigated *in vitro* and *in vivo* (for review see [153]).

#### **5.1.6 Dental Floss, Toothpicks, and Other Interdental Aids**

Dental floss requires hydration to release and deliver an impregnated agent. However, floss is used for limited time periods at each individual site making the time for delivery of an agent very short. Toothpicks

and other interdental aids can, in principle, act as devices for the interproximal delivery of agents from mouth rinse, gels, and toothpaste [55].

##### **5.1.6.1 Irrigators**

Irrigators were designed to spray water, under pressure, around the teeth [5]. While irrigation does reduce the debris and loose bacterial content of the interdental spaces and crevices, suitable additives to the irrigation fluid can actively prevent or minimize the destructive actions of pathogenic microbial flora in the oral environment [131]. The elderly, particularly those who are losing some of their manual dexterity, would find this technique useful [23].

##### **5.1.6.2 Sprays**

Sprays have the advantage of focusing delivery on the required site. The dose is clearly reduced and for anti-septics such as chlorhexidine this has taste advantages [5]. Chlorhexidine sprays, usually 0.1–0.2%, can cause extrinsic staining of teeth to which they are applied. None of the other side effects of chlorhexidine have been reported for spray delivery [10].

##### **5.1.6.3 Slow Release Vehicles**

“Slow-release devices” have been developed, among which are “sustained release devices” delivering the drug for <24 h and “controlled-delivery devices” (CDDs) releasing the agent over an extended period of time. A slow-release device, notably a bioabsorbable chip containing 2.5 mg chlorhexidine in a cross-linked hydrolyzed gelatin matrix (PerioChip), was developed. When placed in an isolated pocket, the chip serves as a CDD slowly releasing its chlorhexidine while simultaneously biodegrading, maintaining over a 7- to 10-day period an average concentration of <125 µg/mL in the crevicular fluid reported to be inhibitory to 99% of bacteria isolated from periodontal pockets [47, 177, 243]. The clinical and microbiological data currently available on a combination of mechanical debridement and repeated chlorhexidine

chip administration in comparison to scaling and root planning alone are limited and conflicting as reviewed by Cosyn & Wyn [47].

## 5.2 Chemotherapeutic Agents

### 5.2.1 Bisguanide Antiseptics

Chlorhexidine is the most studied and certainly the most effective antimicrobial in oral use. At physiological pH, chlorhexidine is a large dicationic molecule, (1,6-di(4-chlorophenyl-diguanido)hexane, with the positive charge distributed over the nitrogen atoms on either side of the hexamethylene bridge (Fig. 5.1). Thus, chlorhexidine has the ability to adsorb onto negatively charged surfaces, such as bacterial cells walls, where it exerts its bacteriostatic and bactericidal effects. Chlorhexidine has a wide spectrum of activity encompassing gram-positive and gram-negative bacteria, yeasts, dermatophytes, and some lipophilic viruses [116]. Its activity is greater at alkaline than at acid pH and is reduced in the presence of organic matter. The latter feature may pose a problem with use in subgingival sites containing high levels of serumal proteins [237].

#### 5.2.1.1 Mechanism of Action

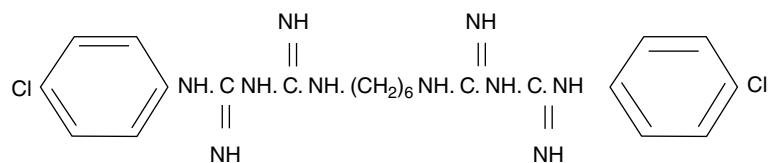
The antibacterial mode of action of chlorhexidine is thought to be as follows. At physiological pH, chlorhexidine is a large dicationic molecule, (1,6-di(4-chlorophenyl-diguanido) hexane, with the positive charge distributed over the nitrogen atoms on either side of the hexamethylene bridge. Thus, chlorhexidine has the ability to adsorb onto negatively charged surfaces, such as bacterial cell walls, and strongly adsorb to phosphate-containing compounds. This alters the

integrity of the bacterial cell membrane and chlorhexidine is attracted toward the inner cell membrane. Chlorhexidine binds to phospholipids in the inner membrane, leading to increased permeability of the inner membrane and leakage of low-molecular-weight components, such as potassium ions ([116] and references therein).

Chlorhexidine also binds to the different surfaces within the mouth (teeth and mucosa) and also to the pellicle and saliva; for example, after a single rinse with chlorhexidine, the saliva itself exhibits antibacterial activity for up to 5 h, whereas persistence at the oral surfaces has been shown to suppress salivary bacterial counts for over 12 h. Thus, although chlorhexidine is able to bind to different anionically charged elements within the oral cavity it also, importantly, maintains its antibacterial activity for several hours. This involved a “reservoir” of chlorhexidine slowly desorbing from all oral surfaces, resulting in a bacteriostatic milieu in the mouth ([116] and references therein).

The antiplaque effect of chlorhexidine can be hypothesized to be as follows. Any bacteria adhering to the tooth surface are challenged by the chlorhexidine at the surface. Depending on the bacterial species, and the amount of chlorhexidine attached to the tooth surface, these microorganisms are either killed (bactericidal effect) or are simply prevented from multiplying (bacteriostatic effect) ([116] and references therein).

The influence of the bacteriostatic effect will increase over time as the concentration of chlorhexidine at the tooth surface decreases, due to desorption into bacteria, saliva etc. The persistent, bacteriostatic effect of chlorhexidine is what makes chlorhexidine the gold standard – plaque is prevented from forming because the bacteria attaching to the tooth surface cannot multiply. This hypothesis means that chlorhexidine must adsorb to the tooth surface and remain there in preference to desorbing into the saliva, while preferentially desorbing from the tooth surface into the bacterial membrane ([116] and references therein).



**Fig. 5.1** Chlorhexidine molecule

### 5.2.1.2 Chlorhexidine Products

CHX has been formulated in a number of products.

#### Mouth Rinses

Aqueous alcohol solutions of 0.12% and 0.2% chlorhexidine mouth rinses are available commercially. The optimum delivery of solution would seem to be those that allow a minimum of approximately 20 mg of chlorhexidine to be delivered throughout the mouth – equivalent to 10 mL of a 0.2% or 15 mL of a 0.12% mouthwash solution – twice daily. Any reduction from these levels would seem to be translated into a reduction in clinical effectiveness [116]. Recently alcohol-free chlorhexidine rinses have become available and have been as effective as one containing alcohol in controlling plaque and reducing gingival inflammation. Therefore, it would seem that its use can be recommended in all patients, but especially in patients for whom the use of alcohol is contraindicated [135].

There is ample evidence to illustrate the superiority of the chlorhexidine mouth rinses in comparison with placebo or control products (as reviewed by [186]). Compared with placebo, chlorhexidine used either in 0.12% or 0.2% concentration demonstrated 35–71% reduction of plaque and 11–39.6% reduction of gingivitis. When chlorhexidine was compared to negative control, significant plaque (21.5–50.3%) and gingivitis (18–30.5%) drops were reported in favor of chlorhexidine [35, 178]. The studies comparing chlorhexidine [95, 105, 130, 178] with other active agents such as essential oils, sanguinarine, and delmopinol seem to agree on the superiority of chlorhexidine [188]. Only one study [35] found no difference between chlorhexidine and Listerine (Pfizer Inc., Morris Plains, NJ, USA). This could be explained by the fact that subjects were requested not to brush the day of the measurements, a fact that may have influenced the plaque scores [188].

Similar results were revealed by the meta-analysis performed by [97] based on the analysis of six-month randomized clinical studies that evaluated both antiplaque and antigingivitis properties of dentifrices or mouthrinses. The 0.12% chlorhexidine mouthrinse had a consistent statistically significant antiplaque (Std. Diff.

= 1.040) and anti-gingivitis effect (Std. Diff. = 0.563) [35, 75, 94, 95, 224, 246].

#### Gel

The effectiveness of the gel would seem to be difficult to explain in terms of an oral bacteriostatic milieu. As previously pointed out, the gel does not penetrate easily to sites away from its application. However, it could be that, during brushing, the gel is carried (through the physical action of moving the brush around the mouth) to binding sites around the oral cavity, from where a slow release will occur. The “equivocal” nature of the gel clinical data merely reflects the relative efficiency with which the gel is transferred around the mouth in different studies [116]. A 1% chlorhexidine gel product is available and can be delivered on a toothbrush or in trays [6]. There is evidence that subgingival chlorhexidine gel administration as a monotherapy temporarily reduces bleeding tendency on probing; a clinical effect coinciding with relevant microbiological changes was also described. To what extent chemical effects contribute to these changes appears to be related to the frequency of gel administration. The expected half-life of a gel within the pocket was investigated and is only about 1 min. Hence, to have a minimum inhibitory concentration (MIC) within the subgingival area, even chlorhexidine with its relatively short killing time requires repeated applications when a gel is chosen as its vehicle: pockets should be irrigated three times within 10 min with a 2% chlorhexidine gel to achieve a 10<sup>2</sup>-fold reduction of the subgingival microflora. There seem to be little to no data indicating that the treatment outcome of scaling and root planning will benefit from the adjunctive subgingival administration of a chlorhexidine gel [45].

#### Tablets

Septofort, are lozenges with 2 mg chlorhexidine gluconate and a mint flavor for disinfecting of the oral cavity and the neck. The preparation is sugar free; it is suitable also for diabetics [87]. Another tablet-form combination of chlorhexidine, fluoride, and xylitol (XYLIHEX) appeared to be as effective antiplaque agents as conventional chlorhexidine rinses [174, 175].

## Varnishes

Currently, three chlorhexidine varnishes are manufactured: Clorzoin<sup>®</sup>, EC40<sup>®</sup>, and Cervitec<sup>®</sup> (Table 5.1). However, none of these varnishes could maintain a significant suppression of *mutans streptococci* for a period of up to 6 months. Therefore, repeated applications are required (for review see [153, 282]). The use of dental varnishes with antimicrobial properties might have potential benefits for patients with chronic gingival inflammation, improving their plaque accumulation and bleeding levels and reducing their gingival index [195]. With regard to measures of periodontitis (pocket depths), there also appeared to be an advantage in favor of people receiving the varnish. The observed effect was greatest in deeper pockets ( $\geq 7$  mm) with differences in pocket reduction estimated to be between 0.62 and 1.37 mm in favor of the groups using the varnish [155]. However, currently available data suggest that in order to achieve a long-term effect, the application of the varnish should be repeated quite frequently. Therefore, patients with fixed orthodontic appliances

might be an interesting population for the evaluation of the effect of monthly varnish applications on gingival health [153]. It was also suggested to use the chlorhexidine varnishes for the prophylaxis of root caries [6].

## Chewing Gum

It has been demonstrated that chlorhexidine can be successfully incorporated into a chewing gum-based delivery system for the reduction of plaque and gingivitis. The advantage of the gum appears to be a longer period of retention in the oral cavity as compared to a mouth rinse and minimal undesirable characteristics such as staining and bitter taste, while maintaining an effectiveness similar to that of chlorhexidine mouth rinses. In fact, chewing two pieces of gum containing 5 mg of chlorhexidine twice a day while not using any other oral hygiene measures for 5 days was showed to be as effective in inhibiting plaque growth and gingival inflammation as was rinsing with chlorhexidine (0.2%)

**Table 5.1** Chlorhexidine varnishes formulation and recommended treatment regimen (Reprinted from [153]. With permission from John Wiley and Sons)

| Varnish               | Components  | Recommended treatment regimen   |
|-----------------------|---|---|
| Clorzoin <sup>®</sup> | 10% Chlorhexidine<br>Sumatra benzoin<br>Ethanol<br>Polyurethane<br>Methylene chloride | A single application during 4 consecutive weeks is recommended:<br><ul style="list-style-type: none"> <li>• The dentition is cleaned, isolated, and dried</li> <li>• The therapeutic varnish is applied to all tooth surfaces by means of a cotton pellet and dental floss and is dried for 15 s with a gentle flow of air</li> <li>• Finally, the teeth are covered with a layer of polyurethane varnish and again dried for 15 s</li> </ul>                             |
| EC40 <sup>®</sup>     | 40% Chlorhexidine<br>Sandarac<br>Ethanol  | A single application of about 10–15 min is sufficient:<br><ul style="list-style-type: none"> <li>• The dentition is cleaned, isolated, and dried</li> <li>• The varnish is locally applied by means of a syringe and is left in place for about 10–15 min</li> <li>• Then, the varnish may be removed by the dentist or is left in place until the following toothbrushing session</li> <li>• This treatment may be repeated 2 times a year or more frequently</li> </ul> |
| Cervitec <sup>®</sup> | 1% Chlorhexidine<br>1% Thymol<br>Ethanol/ethyl acetate<br>Polyvinyl butyral           | 1–3 applications within 10–14 days are recommended:<br><ul style="list-style-type: none"> <li>• The dentition is cleaned, isolated, and dried</li> <li>• The varnish is applied locally by means of a brush and dental floss and is left to dry during 15–30 s</li> <li>• A treatment interval of 3 months is recommended</li> </ul>  |

twice a day [11, 238, 140]. In elderly dependent population the long-term use of a chlorhexidine acetate/xylitol chewing gum (ACHX, Fertin A/S, Vejle, Denmark) was well tolerated, reduced by 91% denture debris and 75% in denture stomatitis and angular cheilitis after 1 year of use, compared with baseline, and may therefore support oral hygiene routines [232, 233]. Cosyn and Verelst [46] indicated that frequent use of a chewing gum as an adjunct to existing oral hygiene measures may reduce plaque levels and gingival bleeding tendency predominantly at lingual/palatal sites in youngsters undergoing fixed orthodontic therapy. However, these clinical parameters do not seem to be additionally reduced when chlorhexidine is incorporated as an active agent. What is more, chlorhexidine increases tooth staining by nearly a factor 5. Hence, there seems to be no indication for a chlorhexidine chewing gum in teenage orthodontic patients when used as an adjunct to normal oral hygiene practices [46].

### Toothpastes

CHX was long time difficult to formulate into toothpaste [6]. However, recent evidence suggested that problems such as inactivation by anionic ingredients contained in toothpaste and the competition for oral retention sites were overcome and 1% and 0.4% CHX-containing dentifrices have been formulated without interactions between chlorhexidine and anionic or cationic ingredients. By adding zinc to a CHX-containing dentifrice, the development of extrinsic dental stain seemed to be decreased [188]. From the studies performed by [217, 277] there is some evidence to support the beneficial use of a CHX-containing dentifrice in comparison with control or placebo products [188].

### Sprays

Sprays containing 0.1% and 0.2% chlorhexidine are commercially available in some countries [6]. Francetti et al. [77] revealed that the efficacy of chlorhexidine spray in the postsurgical control of dental plaque is similar to that of chlorhexidine mouthwash. Tooth staining, however, is significantly lower in the spray group at sites not surgically involved. These effects

might be related to the route of chlorhexidine delivery, as well as the total dose administered that was significantly lower in the spray group with respect to the rinse group. Sprays appear particularly useful for the physically and mentally handicapped groups [6, 78, 79, 119, 120].

### Slow-release Vehicles

A biodegradable chip for the controlled delivery of chlorhexidine directly to the periodontal pocket has been developed. The chip biodegrades and releases chlorhexidine within the pocket over 7–109 days, maintaining an average concentration of chlorhexidine in the gingival crevicular fluid greater than 125 mg/mL for 8 days, and being inhibitory to 99% of bacteria isolated from periodontal pockets. Because it is biodegradable, the chlorhexidine chip does not have to be removed. The chlorhexidine chip is intended for use as an adjunct to scaling and root planning with repeated administration every 3 months in pockets with probing depth more than 5 mm [114, 177, 243] (Fig. 5.2).

A recent systematic review [46] on five RCTs [16, 56, 92, 113, 239] evaluating the surplus value of the chlorhexidine chip when used as an adjunct to SRP



**Fig. 5.2** Clinical aspect of the chlorhexidine chip

revealed that over a 6–9-month period, the use of the chlorhexidine chip in conjunction with SRP significantly reduced PD more than mechanical debridement alone: 0.46 mm additional pocket reduction favoring the test group in the study by [239] and, respectively, 0.30 mm in the study by [113]. The improvements in CAL followed a similar course, yet, to a smaller extent, resulting in a significant additive gain of 0.16 mm in favor of the chlorhexidine chip. Analogue results were found when a comparison was made between a placebo chip and the active chip pointing to a significant additional pocket reduction of 0.26 mm and clinical attachment gain of 0.20 mm, respectively, in favor of the latter. The authors concluded that more RCTs providing clinical and microbiological data are needed to elucidate the surplus value of the chlorhexidine chip as an adjunct to SRP [47].

### 5.2.1.3 Clinical Use of Chlorhexidine

Despite the excellent plaque inhibitory properties of CHX, widespread and prolonged use of the agent is limited by local side effects. Moreover, because of the cationic nature of the chlorhexidine and therefore its poor penetrability, the antiseptic is of limited value in the therapy of established oral conditions, including gingivitis, and is much more valuable in the preventive mode [6]. Chlorhexidine has been found useful in improving oral hygiene and gingival health of several clinical situations (as reviewed by [6, 7, 66]).

- As an adjunct to oral hygiene and professional prophylaxis of periodontal patients [6].
- Improving oral hygiene and gingival health of medically and physically handicapped groups [78, 79].
- In patients with intermaxillary fixation when oral hygiene is particularly difficult [6, 7].
- Medically compromised patients predisposed to oral infections with particular reference to oral candidiasis [6, 7].
- In patients with oral complications associated with cancer therapy (stomatitis, mucositis) chlorhexidine (chlorhexidine gluconate 0.2%) has proved useful. Because of its taste and alcoholic content, it must be applied with an atomizer when the area to be treated is large [256].

- In patients receiving fixed appliance orthodontic treatment [248].
- High-risk caries patients: In children highly infected with *mutans streptococci*, in a 2 years clinical study, the applications of chlorhexidine gel has determined a significantly less number of decayed surfaces compared with other procedures used (topical application of fluoride varnish, Duraphat, or ferric-aluminium-fluoride solution) [137].
- In patients with recurrent oral ulcerations: It has been reported that 0.2% chlorhexidine gluconate mouthwash used three times daily for 6 weeks significantly reduced the total number of minor aphthous ulceration days experienced by patients by approximately 20%. Consequently, the number of days that the patients were free of ulcers during the 6-week period was increased significantly from 17 to 22 days with chlorhexidine, and the interval between successive ulcers was almost doubled [111].
- CHX has been recommended in the treatment of *Candida*-associated infections. However, it was recommended to treat denture stomatitis with specific anticandidal drugs and then to employ chlorhexidine to prevent recurrence [6].
- Oral malodor management: Chlorhexidine rinses (0.2% and 0.12%) in combination with a mechanical approach significantly reduced volatile sulfide compounds levels and mouth and tongue odor by 70–90% (for review see [139, 201, 260]).
- Immediate preoperative (air polishing, ultrasonic scaling, high-speed instruments) chlorhexidine rinsing and irrigation markedly reduces the bacterial load and contamination of the operative area, operator, and staff [6].
- Postoral surgery including periodontal surgery and root planning: chlorhexidine should be used immediately posttreatment and for periods of time until the patient can reinstitute normal oral hygiene [6].
- In the Full-mouth scaling and root planning protocol (the entire dentition in two visits within 24 h, i.e., 2 consecutive days) to reduce the number of subgingival pathogenic organisms. The chlorhexidine regimen, in conjunction with each treatment session, included: (i) brushing the dorsum of the tongue for 1 min with 1% chlorhexidine gel; (ii) chairside rinsing twice with 0.2% chlorhexidine solution for 2 min; (iii) spraying the tonsils four times with a 0.2% chlorhexidine solution;

(iv) subgingival irrigation three times with 1% chlorhexidine gel (repeated after 8 days); (v) instructing the patient to rinse twice daily with a 0.2% chlorhexidine solution for 2 weeks (vi) rinse the mouth and spray the tonsils twice daily with a 0.2% chlorhexidine solution for a period of 2 months after the scaling and root planning (to retard the recolonization of the pockets [200, 251, 255].

CHX, a cation, interacts and forms salts of low solubility and antibacterial activity with anions, such as sodium lauryl sulfate (SLS) and sodium monofluorophosphate (MFP). Both compounds will attach to oral tissues, and both will bind to denature proteins, with a subsequent salt with low antibacterial activity formed in vivo, neutralizing chlorhexidine. To optimize the antiplaque effect of CHX, it seems best that the interval between toothbrushing and rinsing with chlorhexidine be more than 30 min, cautiously close to 2 h after brushing [123].

#### 5.2.1.4 Side Effects of Chlorhexidine

The side effects reported for chlorhexidine include tooth staining, poor taste, taste disturbance, occasionally mucosal erosion, and, rarely, parotid gland enlargement [9] (Figs. 5.3, 5.4). Several mechanisms were proposed for chlorhexidine staining: (i) degradation of the chlorhexidine molecule to release parachloroaniline;



**Fig. 5.4** Brown coloration of the tongue of a patient rinsing twice a day for 4 weeks with a 0.2% chlorhexidine mouth rinse

(ii) catalysis to Maillard reactions; (iii) protein denaturation with metal sulfide formation; and (iv) precipitation of anionic dietary chromogens [6]. The effect may be minimized by limiting the intake of such foods and beverages (tea, coffee) during treatment with chlorhexidine, especially just using the chlorhexidine formulation [116]. Different systems have been introduced in order to reduce the brown pigmentations and other side effects caused by the use of this type of mouthwash, adding to chlorhexidine different products such as peroxiborate, polyvinyl pyrrolidone or sodium metabisulfite, ascorbic acid, and an anti-discoloration system (ADS: sodium metabisulfite and ascorbic acid) [8, 14, 19, 42, 96]. Controversial clinical results were reported regarding the chlorhexidine ADS system. Arweiler et al. [14] suggested that the 0.2% alcohol-containing chlorhexidine mouthwash showed superiority in inhibiting plaque regrowth and reducing bacterial vitality compared with the solution with ADS. In contrast, Cortellini et al. [44] reported that (1) chlorhexidine ADS caused less pigmentation, was burdened by less side effects, and was more agreeable than the control CHX; (2) chlorhexidine ADS was as effective as chlorhexidine without ADS in reducing gingival signs of inflammation in the postsurgical early healing phase; (3) the use of chlorhexidine ADS could be of value in treatment protocols in which the patient compliance with a chlorhexidine mouthwash prescription is relevant.

Hypogeusia (reduced ability to taste things) induced by chlorhexidine concerns specifically salt and bitter, but the ability to recognize either sweet sucrose or sour citric acid is not affected; this inability lasts some days after the interruption of mouth rinses. Incidence of



**Fig. 5.3** Brown coloration of the teeth of a patient rinsing twice a day for 2 weeks with a 0.2% chlorhexidine mouth rinse

dysgeusia (distortion of the sense of taste) is not related to chlorhexidine concentration [80, 107]. Chlorhexidine concentrations in mouth rinses till 0.12% and mucosa exposure not exceeding 1 min twice a day seem the best procedure to protect tastes in clinical practice [149].

### **5.2.2 Quaternary Ammonium Compounds**

This group of cationic surface active agents consists of many members who possess the ability to interact with the bacterial cell membrane, affecting its permeability with subsequent loss of cell content. Although they are bactericidal to both Gram-positive and Gram-negative bacteria, the evidence suggests that they are more effective against the former. Two of the groups, cetylpyridinium chloride (CPC) usually at 0.05%, with and without domiphen bromide and benzethonium chloride, at a similar concentration, have been used in mouthwashes [186]. It is suggested that CPC interaction with bacteria occurs by the disruption of membrane function, leakage of cytoplasmic material, and, ultimately, the collapse of the intracellular equilibrium [103, 219].

Incorporation of cetylpyridinium chloride in dentifrice formulations is difficult because of its poor compatibility of this agent with other dentifrice components and its prolonged use results in stain development [186]. Sheen et al. [229] reported that toothpaste, whilst possessing some plaque inhibitory activity, when used immediately before a CPC mouth rinse adversely affected the plaque inhibitory action of this antiseptic. This in part may explain the reported lack of adjunctive benefits of CPC rinses to normal oral hygiene practices and supports the suggestion, made for chlorhexidine rinses, that their use should follow toothpaste by at least 60 min [229]. However, in the USA, consumers typically rinse their mouths with water in some fashion following toothbrushing to remove the dentifrice slurry from their mouths. It is likely that this regimen may remove the dentifrice excipients, thereby allowing mouth rinses to have the maximum antiplaque effect [273]. Recently, [273] specifically evaluated whether a water rinse imposed between toothbrushing and use of a CPC mouth rinse would be sufficient to remove any potentially interfering dentifrice excipients from the mouth and provide

antiplaque benefits similar to those obtained by waiting for 60 min. after toothbrushing as recommended in a previous study using dentifrice slurry with no brushing [229]. This study demonstrated that the CPC mouth rinse provides significant antiplaque benefits when used as an adjunct to various toothbrushing regimens, including a regimen with CPC rinsing immediately following toothbrushing, versus toothbrushing alone. Brushing with standard toothpaste and rinsing with water before using the CPC mouth rinse was not statistically different from brushing with toothpaste and waiting 60 min. before using the CPC mouth rinse [273]. However, the recommended regimen to enhance the CPC activity while complementing typical oral hygiene practices is to perform a water rinse between toothbrushing and rinsing with the 0.07% CPC rinse [273].

Three recent systematic reviews have evaluated the effect of cetylpyridinium chloride-containing mouth rinses as adjuncts to toothbrushing on plaque and parameters of gingival inflammation [97, 103, 188].

When assessed, the antiplaque efficacy analysis of CPC [97] revealed that four ([13, 145, 224, 246] unpublished data) of the seven studies exhibited statistical significance and three did not [39, 138, 159]). There was a great deal of heterogeneity in both the CPC agents evaluated Cepacol Antibacterial Mouthwash, Combe, White Plains, N.Y., containing 0.05% CPC; Scope Mouthwash, Procter & Gamble, Cincinnati, containing 0.045% CPC; 0.05% CPC-containing mouth rinse and two mouth rinses containing 0.07% CPC, one with an alcohol vehicle and one without alcohol and in the results obtained, with some of the agents exhibiting antiplaque effects and some not exhibiting these effects. When antigingivitis effects were analyzed, similar to the evaluation of the antiplaque effects of mouth rinses containing CPC, [97] found both statistical heterogeneity and a variety of formulations evaluated; the author considered that it was difficult to reach conclusions about this agent, although the results of studies of individual CPC products were similar to those for other types of active agents.

In his review, Paraskevas et al. [186] cited only one 6-month controlled clinical trial [13] who reported plaque reduction of 28.2% and gingivitis reduction of 24% in comparison with control, and failed to identify any RCT supporting the use of CPC as antiplaque or antigingivitis agent.

Recently, Haps et al. [103] screened titles and abstracts of 3,250 papers resulted in eight publications that met the criteria of eligibility for his review [13, 18, 71, 145, 166, 172, 246, 283]. A meta-analysis was performed to compare the effect of CPC mouth rinses to that of toothbrushing in conjunction with a placebo mouth rinse or toothbrushing only. The meta-analysis was performed four times: the plaque parameter for studies  $\geq 4$  weeks (a), the gingival health parameter for studies  $\geq 4$  weeks (b), the plaque parameter for studies  $< 6$  months (c) and the plaque parameter for studies  $\geq 6$  months (d). In all cases, baseline scores were not statistically different. The end scores showed a significant effect for the Quigley and Hein Plaque Index in favor of the CPC group compared to those of toothbrushing only or toothbrushing followed by a placebo rinse [weighted mean difference (WMD):  $-0.50$ ,  $P < 0.00001$ ; test for heterogeneity  $P = 0.002$ ,  $I^2 = 71.6\%$ ]. The heterogeneity was greater for intermediate-length studies ( $I^2 = 68.1\%$ ) than long-term studies ( $I^2 = 58.8\%$ ). One could deduce from these outcomes that a greater effect is observed in long-term studies than in intermediate-length studies. The end scores also displayed a significant effect for the Löe and Silness Gingival Index in favor of the CPC group compared to those of the toothbrushing only or toothbrushing followed by a control rinse group (WMD:  $-0.25$ ,  $P < 0.00003$ ; test for heterogeneity  $P = 0.0001$ ,  $I^2 = 87.0\%$ ) [103]. The authors concluded that the existing evidence supports that CPC containing mouth rinses, when used as adjuncts to either supervised or unsupervised oral hygiene, provide a small but significant additional benefit in reducing plaque accumulation and gingival inflammation.

It has been showed that the CPC antiplaque or antigingivitis potential is limited by the rapidity by which they are desorbed from oral tissue sites. Apparently, they adsorb well initially but do not have sufficient substantivity to maintain an antibacterial effect. Use of high concentrations or more frequent rinsing (four times daily) not only increases efficacy, but also accentuates the undesirable side effects of staining, burning sensation, and transient desquamation of the oral [28, 143].

A nondegradable osmotic slow-release dosage form containing 6.6 mg CPC (MOTS: Mucosal Oral Therapeutic System) and CPC lozenges Cepacol (each 1.6 mg CPC) were also studied for their effect to inhibit new plaque formation and gingivitis, but were less effective compared with the Peridex mouthwash. The

lozenges resulted in more staining at 18 days than MOTS CPC and Peridex [266].

Mouth rinses combining cetylpyridinium chloride with chlorhexidine (CHX 0.05%+CPC 0.05%+zinc lactate 0.14% or chlorhexidine 0.12%+CPC 0.05% nonalcoholic formulation) are available and compare well with established chlorhexidine products (0.2% CHX) in a both significantly decreased plaque and gingivitis indices as well as morning halitosis, with reduced subjective side effects [197–199, 263].

### 5.2.3 Detergents

Sodium lauryl sulfate (SLS) is one of the most widely used synthetic detergents in dentifrices. In general, surface active agents are thought to lower the surface tension, penetrate and loosen surface deposits, and emulsify or suspend the debris, which the abrasives in a dentifrice remove from the tooth surface. Sodium lauryl sulfate is an anionic molecule with high affinity for protein molecules. The concentration of sodium lauryl sulfate in dentifrices usually ranges from 0.5% to 2.0% [17]. It has been showed that SLS has also plaque inhibitory action similar to triclosan. Jenkins et al. [115] compared the magnitude and duration of salivary bacterial count reductions produced by a single rinse of 0.2% triclosan, 1% sodium lauryl sulfate (SLS), and 0.2% chlorhexidine mouthwashes. A group of 16 volunteers (21–42 years old) took part in a single-blind latin-square randomized crossover designed study with balanced residual effects. The results showed that sodium lauryl sulfate exerts not inconsiderable antimicrobial activity in the mouth, which by comparison with saline persisted for the duration of the study. Differences between sodium lauryl sulfate and triclosan were significant in favor of former.

Generally, chlorhexidine is considered as an adjunct to mechanical oral hygiene and used before or after toothbrushing with dentifrice, especially during initial therapy and healing following periodontal surgery. Unfortunately, chlorhexidine and SLS can act as antagonists. The mode of action is based on the ionic attraction of CHX, a cationic bisbiguanide symmetrical molecule, to SLS, a molecule with an anionic nature and high affinity for protein molecules [264].

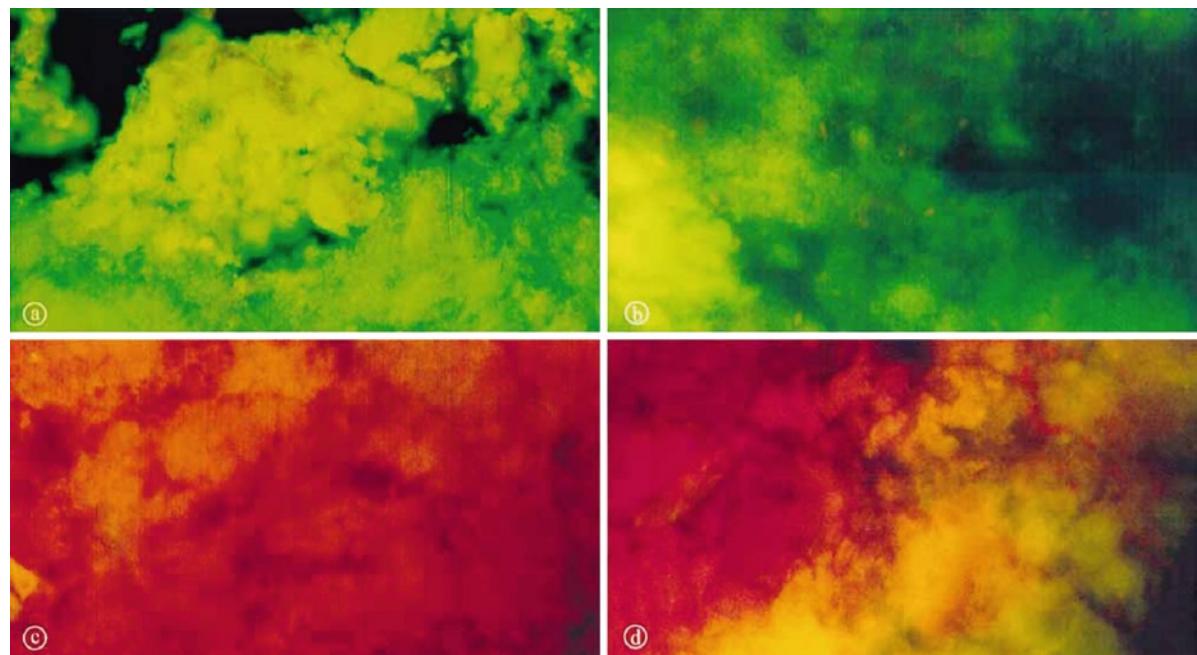
After Barkvoll et al. [17] and Owens et al. [179] studies who revealed that the efficacy of a chlorhexidine rinse was significantly reduced in the environment of SLS, it has been recommended that the time between a chlorhexidine rinsing and toothbrushing with a SLS-containing dentifrice should at least be 30 min, if reduction in the antimicrobial effect is to be avoided, while after 2 h, the neutralizing effect of SLS totally disappeared [123, 264]. It was suggested, in order to optimize the efficacy of a chlorhexidine rinse, that toothbrushing should be performed using no dentifrice or a dentifrice without antagonistic ingredients [264].

In contrast to the previous study, Van Strydonck et al. [264, 265] showed that the antiplaque effect of a chlorhexidine mouth rinse does not appear to be reduced under the influence of a normal toothbrushing exercise with a dentifrice, irrespective of whether the dentifrice contains SLS, or is used before or after the rinse, probably due to a lower intraoral SLS concentration, a shorter contact time of SLS, and water rinsing immediately after brushing with dentifrice.

### 5.2.4 Essential Oils

Phenolic compounds have long been used as antiseptics and disinfectants [112]. Listerine is a combination of the phenol-related essential oils, thymol (0.06%) and eucalyptol (0.09%), mixed with menthol (0.04%), methyl-salicilate (0.06%) plus alcohol (26.9%) [38]. The main antimicrobial mechanism of essential oil mouthwashes are: killing microorganisms by disrupting their cell walls and by inhibiting their enzyme activity, preventing bacteria from aggregating with Gram-positive pioneer species, slow bacterial multiplication, and extract endotoxins from Gram-negative pathogens, reducing bacterial load, slowing plaque maturation, and decreasing plaque mass and pathogenicity [70, 126]. The recommended use is twice daily following the toothbrushing [198] (Fig. 5.5).

Listerine efficacy has been seen in both short- and long-term studies regardless of the level of oral hygiene that were analyzed by several systematic reviews. The 6-month RCT on the essential oils were reviewed by



**Fig. 5.5** (a) Photomicrograph of baseline vital stained plaque sample. Predominantly green staining indicates live bacteria (orig. mag.  $\times 25$ ). (b) Photomicrograph of control plaque sample at 30 min (orig. mag.  $\times 25$ ). (c, d) Photomicrographs of 30-min

plaque samples from essential oil mouth rinse group showing predominantly red staining (d) and complete red staining (c) (orig. mag.  $\times 25$ ) (Reprinted from [182]. With permission from John Wiley and Sons)

Paraskevas et al. [186]. All but one study [129] used a parallel design with professional prophylaxis at start in order to eliminate plaque and calculus. In most of these studies, the product had been used next to unsupervised oral hygiene and was compared either to a placebo or a negative control. Plaque and gingivitis reductions versus placebo ranged between 14.9% and 24.2% and 9.4% and 28.2%, respectively. Reductions versus negative control were 18.8–36.1% and 14–35.9%, respectively [186].

The largest number of studies of mouth rinses that evaluated essential oils (22 studies, four with two active arms) analyzed by [97] showed clearly that this agent is effective as both an antiplaque (Std. Diff. = 0.852,  $P<0.0001$ ) and an antigingivitis (Std. Diff. = 0.762) agent. Four studies [35, 94, 178, 224] compared mouth rinses containing essential oils with chlorhexidine mouth rinses. The studies compared active agents with control agents, as well as with each other. The results showed statistically significant antiplaque effects for both agents in all four studies. In all cases, 0.12% chlorhexidine exhibited greater antiplaque effects than did mouth rinses containing essential oils (the effect for essential oils was about 60% of that for the 0.12% chlorhexidine mouth rinse). The results were similar for gingivitis. The four studies showed a statistically significant advantage for the active agents over the control agents. Mouth rinses containing essential oils had about 60% of the antigingivitis effect of 0.12 % chlorhexidine; however, the difference in antigingivitis effects of the agents was close to, but failed to reach, statistical significance ( $P=0.068$ ) [97].

This review performed by Stoeken et al. [245] aimed to investigate the long-term effect of a mouth rinse containing essential oils (Listerine, Pfizer Consumer Healthcare, Morris Plains, New Jersey, USA) on gingivitis and dental plaque. MEDLINE and Cochrane Central Register of Controlled Trials were searched up to and including December 2006 to identify appropriate studies. Independent screening of titles and abstracts of 566 papers resulted in 11 publications that met the criteria of eligibility. In all studies, essential oils were used as an adjunct to regular daily toothbrushing. A statistically significant reduction in overall gingivitis was noted compared to the control (weighted mean difference [WMD]: -0.32, 95% CI: -0.46 to -0.19,  $P<0.00001$ ; test for heterogeneity:  $P<0.00001$ ,  $I^2=96.7\%$ ). For interproximal sites, the use of the test mouth rinse resulted in significantly more gingivitis

reduction compared to control mouth rinse (WMD: -0.29, 95% CI: -0.48 to -0.11,  $P=0.002$ ; test for heterogeneity:  $P<0.00001$ ,  $I^2=95.18\%$ ), whereas no differences were observed compared to dental floss. With respect to plaque scores, essential oils produced significant overall reductions in plaque (WMD: -0.83, 95% CI: -1.13 to -0.53,  $P<0.00001$ ; test for heterogeneity:  $P<0.00001$ ,  $I^2=96.1\%$ ). Separate analysis for interproximal areas revealed that EO resulted in more pronounced plaque drops compared to the control mouth rinse (WMD: -1.02, 95% CI: -1.44 to -0.60,  $P<0.00001$ ; test for heterogeneity:  $P<0.00001$ ,  $I^2=96.1\%$ ) or the use of floss (WMD: -0.75, 95% CI: -1.15 to -0.363,  $P<0.0002$ ; test for heterogeneity:  $P<0.0002$ ,  $I^2=93.0\%$ ). The authors concluded that when used as an adjunct to unsupervised oral hygiene, essential oils provide an additional benefit with regard to plaque and gingivitis reduction as compared to a placebo or control.

Studies have also shown that essential oil mouthwashes have important benefits in supporting gingival health around implants, following periodontal surgery in the early postoperative phase, while pre-procedural subgingival irrigation and rinsing can significantly reduce the level of bacteremia associated with ultrasonic scaling, for as long as 40 min postrinse (as reviewed by [225]). Essential oil mouthwashes offer additional benefits to both dental practitioners and patients. For example, they control malodor for up to 3 h by killing odor-causing bacteria [40, 193, 201].

Formulations containing essential oils as active ingredients have fewer and generally less troublesome adverse effects than products containing antimicrobial agents. Although the high alcohol content (often as much as 25%) is probably the reason why patients report an initial burning sensation, most accommodate this effect after a few days of use [112]. No mucosal aberrations or development of extrinsic tooth stain at either 6 or 9 months were observed [48, 245].

## 5.2.5 Phenols

Phenolic compounds have long been used as antiseptics and disinfectants. **Triclosan**, a both bisphenol and

nonionic germicide with low toxicity and a broad spectrum of antibacterial activity, is available in dentifrices and mouth rinses. It is also widely used in soaps, antiperspirants, and cosmetic toiletries [38].

The mechanisms of the antimicrobial action are not entirely clear. Due to its hydrophobic and lipophilic nature, triclosan adsorbs to the lipid portion of the bacterial cell membrane and in low concentrations interferes with vital transport mechanisms [219]. It also demonstrated the triclosan's antiinflammatory effect, independent of its effect on plaque formation. Triclosan had a direct inhibitory effect on primary enzymes (cyclooxygenase and 5-lipoxygenase) in the pathways of arachidonic acid metabolism and that this action would lead to a reduction in the formation of proinflammatory metabolites such as PGE<sub>2</sub> and leukotriene B<sub>4</sub>. This was confirmed in cell culture studies in which triclosan inhibited IL-1-induced production of PGE<sub>2</sub> in human gingival fibroblasts [83].

To date there have been no reports of adverse effects on the oral hard or soft tissues that could be attributed to the use of triclosan [24, 58, 66, 112]. No shifts in the microflora of supragingival plaque favoring the growth of either opportunistic or pathogenic bacterial or yeast species have been observed [50, 51, 52, 58, 280].

### 5.2.5.1 Triclosan in Dentifrices

Since it does not bind well to oral sites due to its lack of a strong positive charge, formulations have been developed to enhance its ability to bind to plaque and teeth. These formulations include:

- Combinations with zinc citrate to take advantage of its potential antiplaque and anticalculus properties
- Incorporation of triclosan in a copolymer of methoxyethylene and maleic acid to increase its retention time and
- Combination with pyrophosphates to enhance its calculus-reducing properties [38]

#### Triclosan + zinc citrate

Contradictory results were obtained when antiplaque and antigingivitis effects of triclosan/zinc citrate agent

were reviewed, when both significant [112, 189] and nonsignificant result were reported [97].

#### Triclosan + copolymer

The ultimate goal of a toothpaste with antiplaque and antigingivitis properties is to produce clinically significant results during unsupervised home use between a patient's regular dental visits. So while shorter-term studies provide the initial evidence to support the triclosan/copolymer formulation, it is the result of the six-month studies, which are of most value to the dental team and their patients [24].

A great deal of the evidence for the efficacy of the triclosan/copolymer toothpaste relates to its effects on plaque and gingivitis (Table 5.2) [24]. Four systematic reviews supports very clearly the benefit of triclosan triclosan/copolymer toothpaste for reduction of plaque and gingivitis [57, 60, 186]. Based on the analysis of several 6-month RCT, Gunsolley [97] revealed that the dentifrice composed of 0.30% triclosan, 2.0% Gantrez copolymer exhibited significant results for 14 of the 18 arms (representing 17 studies) and a substantially larger effect (Std. Diff. = 0.823). Because there was statistically significant heterogeneity, the random effects model was used to evaluate the efficacy of the agent. The overall analysis of the efficacy of the triclosan/copolymer agent using a random-effects model resulted in a highly significant ( $P < 0.0001$ ) mean group difference that favored the active agent. Consistent and strong results in support of the antigingivitis effects of these agents were also obtained (12 of the 16 arms demonstrated statistically significant effects). Similar significant plaque and/or gingivitis reductions when compared with NaF control were revealed by Paraskevas [186].

Sixteen trials provided data for the meta-analysis assessing the effectiveness of a toothpaste containing Triclosan and polyvinyl-methyl ether maleic acid copolymer in improving plaque control and gingival health [57].

The meta-analyses for both the Quigley-Hein plaque index (QHPI) and the plaque severity index showed that the triclosan/copolymer dentifrice is effective in reducing plaque compared with a fluoride dentifrice, with a weighted mean difference (WMD) of -0.48 (95% confidence interval (CI) (random effects): -0.64 to -0.32)

**Table 5.2** Summary of significant plaque and gingivitis results from studies of minimum six-month duration, included in systematic review of Davies [59] and the meta-analysis of Gunsolley [97] (Reprinted with permission from Macmillan Publishers Ltd: [24])

| Study                      | Plaque reduction | Gingivitis reduction |
|----------------------------|------------------|----------------------|
| Garcia-Godoy et al. [85]   | 58.9%            | 30.1%                |
| Cubells et al. [49]        | 24.9%            | 19.7%                |
| Deasy et al. [62]          | 32.3%            | 25.6%                |
| Mankodi et al. [148]       | 11.9%            | 19.7%                |
| Denepitiya et al. [64]     | 18.4%            | 31.5%                |
| Bolden et al. [27]         | 17.0%            | 29.0%                |
| Lindhe et al. [136]        | 31.2%            | 26.6%                |
| Triratana et al. [258]     | 32.9%            | 18.8%                |
| Svatun and Saxton [249]    | Not Significant  | 25.0% <sup>a</sup>   |
| Palomo et al. [181]        | 11.3%            | 19.9%                |
| Renvert and Birkhed [204]  | 38.8%            | 18.2%                |
| Kanchanakamol et al. [121] | 12.1%            | Not Significant      |
| Hu et al. [110]            | 16.1%            | 24.3%                |
| McClanahan et al. [154]    | Not Significant  | Not Significant      |
| Charles et al. [34]        | 22.1%            | 20.7% <sup>b</sup>   |
| Tiriratana et al. [258]    | 34.9%            | 25.7%                |
| Mankodi et al. [146]       | 18.7%            | 22.2%                |
| Allen et al. [12]          | 27.9%            | 21.4%                |
| Winston et al. [272]       | Not Significant  | Not Significant      |

All plaque results are statistically significant percentage difference of mean Quigley-Hein Plaque index for triclosan/copolymer toothpaste compared to the placebo toothpaste unless otherwise stated

<sup>a</sup>Gingival Bleeding index

<sup>b</sup>Modified Gingival Index

for the QHPI (0–5 scale) and a WMD of −0.15 (95% CI (random effects): −0.20 to −0.09) for the plaque severity index (0–1 dichotomous index). Similarly, the meta-analyses for the Löe and Silness (0–3 scale) and gingivitis severity indices (0–1 dichotomous index) were both significant, showing a reduction in gingivitis when comparing the triclosan/copolymer dentifrice with a fluoride dentifrice, with WMDs −0.26 (95% CI (random effects): −0.34 to −0.18) and −0.12 (95% CI (random effects): −0.17 to −0.08), respectively. The plaque and gingivitis severity indices demonstrated that the

triclosan dentifrice reduced the proportion of surfaces with heavy plaque by 15% and those with gingival bleeding by 12%. In relative terms, the proportion of sites that had plaque reduced from 0.31 to 0.16, a 49% reduction. Similarly, the proportion of sites with bleeding reduced from 0.24 to 0.12, a 49% reduction. For the Quigley-Hein plaque index, the WMD corresponds to a 23% reduction in plaque when comparing the triclosan dentifrice with a fluoride dentifrice. For the Löe and Silness index, this also corresponds to 23% reduction in plaque when comparing the triclosan dentifrice with a fluoride dentifrice [57].

There have been also a number of randomized, controlled, clinical trials that have investigated the efficacy of a triclosan/copolymer toothpaste as an adjunct to mechanical plaque removal in the control of periodontitis (Table 5.3). Overall, these studies have shown that while good oral hygiene and a rigorous maintenance program alone cannot entirely prevent recurrent disease in highly susceptible patients, daily use of a triclosan/copolymer toothpaste can lead to an improved outcome after treatment. Moreover, it has been showed that a triclosan/copolymer toothpaste is superior to a placebo in limiting the reformation of supragingival calculus. The effect on subgingival calculus formation has not been studied to the present time. A body of available evidence also suggest that the use triclosan/copolymer toothpaste results in an improvement in halitosis (bad breath) (for review see [24] and references therein).

Based on the body of evidence, [24] suggested that dental professionals should consider recommending the twice daily use of a triclosan/copolymer toothpaste to patients who are susceptible to periodontal disease in the knowledge that it will be a useful adjunct to periodontal preventive and supportive therapies [24].

#### Triclosan + pyrophosphate

Gunsolley [97] revealed that although studies of dentifrices containing the triclosan/soluble pyrophosphate agent resulted in marginally statistically significant results, the test for heterogeneity also was significant. The studies were inconsistent, however, because three of the four resulted in nonsignificant results and, thus, did not provide sufficient evidence that this is an effective antiplaque and/or antigingivitis agent.

**Table 5.3** Effect of the home use of triclosan/copolymer toothpaste on periodontal patients (Reprinted with permission from Macmillan Publishers Ltd, [24])

| Study   | Outcomes  |
|---|---|
| Rosling et al. [207] Clinical study of highly susceptible periodontal patients  | Significant difference in mean probing depth change between the groups ( $P < 0.01$ ) over a 3 year period.   |
| Furuichi et al. [82] Clinical study of highly susceptible periodontal patients with $>2$ mm loss of attachment during [207] study | Clinically significant reduction in the number of sites with gingivitis following scaling and root planning in the test group but not in the control group. Control and test groups showed a clinically significant reduction in mean probing depth and a significant gain in attachment following treatment. The changes were significantly greater in the test group. |
| Rosling et al. [208] Microbiological study of highly susceptible periodontal patients (subset of [207])                           | Significant reductions in total viable counts of subgingival microbiota   |
| Ellwood et al. [67] Clinical study of highly susceptible teenagers with high mean probing depth at baseline                       | 50% reduction in attachment loss compared to placebo  |
| Cullinan et al. [50] Clinical study of general adult population   | Significant reduction of number of sites with probing depths $>3.5$ mm  |
| Cullinan et al. [51] Microbiological study of subgingival plaque samples from adults in clinical study                            | The clinical effect was independent of changes in the periodontopathic bacteria   |
| Papas et al. [183] Clinical study of xerostomic patients at high risk of periodontitis  | Clinically significant reduction in probing depths and inflammation in xerostomic patients  |
| Kerdvongbundit et al. [122] Clinical study of smokers with chronic periodontitis  | Clinically significant improvement in plaque, calculus and gingivitis indices   |

### 5.2.5.2 Triclosan in Mouth rinses

Several RCT of 6 months' duration have assessed the effectiveness of a mouth rinse containing triclosan/copolymer [15, 218, 257, 275]. Reductions in plaque and gingivitis ranged from 24% to 36% and from 23% to 46%, respectively, when compared with a placebo rinse [58, 189].

### 5.2.6 Metal Salts

A number of metal ions have been studied for their effects on plaque, and zinc, copper, and tin have been shown to possess plaque inhibitory activity [66]. Results have been somewhat contradictory but appear dependent on the metal salt used, its concentration and frequency of use [6].

Both copper and tin suffer from the local side effect of staining. Some fluoride compounds such as stannous fluoride and amine fluorides also have plaque inhibitory activity, but not as a result of the

fluoride ion itself but rather due to the effect of the stannous ion or the surface-active amine portion of the molecule [66]. Tin combined with fluoride ( $\text{SnF}_2$ ) is a well-known agent that has been used in several formulations including dentifrices, gels [31], sustained release varnishes [244], impregnated dental floss [73], and mouth rinses regimes have been tested throughout the years [189]. Similar to sodium fluoride ( $\text{NaF}$ ), stannous fluoride ( $\text{SnF}_2$ ) has been found to have caries-inhibiting effects in different age groups [20, 247, 271]. Root caries development is a common finding associated with surfaces developing recession in patients once treated for periodontal problems. No difference was reported when the effect of using a dentifrice and mouth rinse containing amine fluoride ( $\text{AmF}$ ) and stannous fluoride ( $\text{SnF}_2$ ) were compared to a dentifrice and mouth rinse, both containing sodium fluoride ( $\text{NaF}$ ), with regard to their root caries experience in a group of periodontal maintenance patients [184, 185].

Furthermore,  $\text{SnF}_2$  has been shown to have plaque inhibiting effects in several clinical trials [186, 271]. The use of  $\text{SnF}_2$  dentifrices results also in a greater

reduction in gingivitis [144, 227], morning breath odor [198], in dentinal hypersensitivity [221, 222] compared to that achieved with conventional dentifrices.

Two recent reviews have evaluated the antiplaque and antigingivitis effect of SnF<sub>2</sub>. Gunsolley [97] revealed that dentifrices with stannous fluoride had statistically significant, but marginally clinically significant, evidence of an antiplaque effect; however, there was both a statistically and clinically significant antigingivitis effect. In another review, performed by Paraskevas & van der Weijden [189], it was concluded that the use of SnF<sub>2</sub> dentifrices results in gingivitis and plaque reduction when compared to a conventional dentifrice [189]. When the combined (dentifrice/mouth rinse) regimen was taken into account, only one study was found that provided evidence of this regimen [160]. This study found no differences between treatment groups in terms of gingivitis and plaque. For both the mouth rinse and the dentifrice/mouth-rinse formulations, more research is necessary in order to gain further insight in the effects of SnF<sub>2</sub> (alone or in combination with AmF) on plaque and gingivitis [189]. A statistically significant increase of the prevalence of staining in comparison with NaF or placebo seemed to be a common finding after the use of different SnF<sub>2</sub> formulations (reviewed by [189] with references therein) due to interactions with dietary chromogens [6].

Stannous pyrophosphate has been introduced in several dentifrices formulations and positive results have been obtained [3, 176].

Although stannous fluoride is the most commonly used metal salt in dentifrices, the potential of the **zinc ion** as plaque inhibitor has also been investigated. The astringency of the zinc at clinically active levels precludes the use of many zinc salts. However, the selection of the citrate salt, which is sparingly soluble in water but more soluble in dentifrice formulation, permits the use of zinc at concentrations sufficient for antiplaque activity while maintaining a palatable product [112].

Zinc salts were included in several toothpaste and mouthwash formulations with antiplaque, anticalculus effects [1, 2, 21, 30, 33, 72, 89, 101, 104, 216, 242, 249, 250] being efficient also in halitosis (bad breath, malodor) reduction due to their capacity to inhibit production of volatile sulfur compounds [171, 205, 211, 253, 279].

In addition to the stannous and zinc salts, **copper** has also been shown to be a potent inhibitor of plaque formation. All the ions have also been reported to inhibit glycolysis, the effectiveness being in the sequence Cu>Sn>Zn. Copper salts have not been used in commercially available mouthwashes or dentifrices because of their unpleasant taste, potentiality to cause stain and potential toxicity [112].

### 5.2.7 Enzymes

Enzymes fall into two groups. Those in the first group are not truly antimicrobial agents but more plaque removal agents in that they have the potential to disrupt the early plaque matrix, thereby dislodging bacteria from the tooth surface. Such agents as dextranase, mutanase, and various proteases, unfortunately, have poor substantivity and are not without unpleasant side effects, notably mucosal erosion. The second group of enzymes employs glucose oxidase and amyloglucosidase to enhance the host defense mechanism. The aim is to catalyze the conversion of endogenous and exogenous thiocyanate to hypothiocyanite via the salivary lactoperoxidase system. Hypothiocyanite produces inhibitory effects upon oral bacteria, particularly streptococci, by interfering with their metabolism. Very few studies have assessed a toothpaste product containing the enzymes and thiocyanate but equivocal results for benefits to gingivitis were obtained and there are no convincing long-term studies of efficacy [6]. The trial conducted by Rotgans & Hoogendoorn [210] was designed to differentiate between enzyme inhibition and mechanical effect of brushing on plaque reduction after using a toothpaste containing amyloglucosidase and glucose oxidase. The double-blind crossover study using nine persons was 50 days long; during the first period the tests persons used their own favorite toothpaste. At day 15 and day 29, new periods were started with experimental pastes. Plaque scores were measured three times a week and no brushing was allowed on the scoring days. Significantly, superior action in reducing plaque index scores was demonstrated by the enzyme-containing dentifrices. Midda and Cookse [161] conducted a double-blind non-crossover study using a split-mouth technique in aim to evaluate the enzyme-containing dentifrices. A total of 150 subjects (20–55 years old) were involved, 135 completed the

trial for the first 3 months and the study is continuing with approximately 60 people for the second 3 months. Significant reduction in gingivitis scores was noted.

### 5.2.8 Natural Products

A considerable number of natural products such as herbs and plant extract have been used in oral hygiene products for many years. Unfortunately, there are little available data and despite the claims, products, particularly toothpastes, have effects against plaque no different to conventional fluoride toothpastes [6].

#### 5.2.8.1 Plant Alkaloids – Sanguinarine

The plant extract, sanguinarine has attracted more attention. Chemically, sanguinarine is described as a benzophenanthridine alkaloid derived from the alcoholic extraction of powdered rhizomes of the blood-root plant, *Sanguinaria canadensis* [91].

Sanguinarine exerts antimicrobial activity against Gram-positive and Gram-negative bacteria, including oral isolates. It reportedly suppresses the activity of several enzymes, possibly through oxidation of thiol groups. The antimicrobial activity is thought to be associated with the lipophilicity of the molecule, being highest at pH 5.4. However, the structure of sanguinarine allows the molecule to function as a metal ligand, and the marketed preparations contain zinc. It may be speculated that potential effects are related to the zinc content, though claims are made that the antiplaque effects are influenced more by the sanguinarine concentration than by the presence or absence of zinc ions [219].

The current formulation contains the extract at 0.03% (equivalent to 0.01% sanguinarine) and 0.2% zinc chloride to enhance the antiplaque effect [186]. The results from studies using either the toothpaste or the mouth rinse have been equivocal [52, 163, 164], but when used in combination, significant reductions in plaque (17–42%) and gingivitis (18–57%) were reported in 6 months RCT [186].

In a double-blind, 4-cell, placebo-controlled, parallel investigation involving 120 subjects, the twice daily use of a sanguinarine and zinc chloride containing dentifrice with and without fluoride in combination with a sanguinarine and zinc chloride mouth rinse resulted in

significantly less plaque accumulation (the percentages of Plaque Severity Index reductions were 33% and 41%), significantly less gingival inflammation, and significantly less bleeding on probing (the percentage reductions in bleeding were 24.7–36.6% at 3 months, and 31.5–41.2% at 6 months for the test groups) when compared to placebo preparations of the same products. The adverse effects included burning sensations and slight mucosal folding with a pebbled, erythematous appearance of the oral mucous [124].

In the second study, 60 subjects with moderate levels of plaque and gingivitis were randomly assigned to active and placebo groups. After baseline evaluations, subjects received a half mouth (split contralateral) supragingival rubber cup polishing, with quadrants randomly assigned within groups. Noninvasive measures of plaque and gingivitis were assessed at baseline, 2, 6, 14, 20, and 28 weeks. Bleeding upon probing was assessed at baseline, 6, 14, and 28 weeks. At 6 months active group's scores were 21% lower than placebo groups for plaque, 25% for gingivitis and exhibited 43% fewer bleeding sites than the placebo group [104].

In patients under orthodontic treatment, a sanguinarine regimen (toothpaste and oral rinse regimen), evaluated during a 6-month period, reduced plaque by 57%, gingival inflammation by 60%, and sulcular bleeding by 45% from baseline compared with placebo group reductions of 27% (plaque) and 21% (gingival inflammation), and an increase of 30% in bleeding index [102].

Briefly, sanguinarine, when used next to normal mechanical plaque control, appears to be an effective inhibitory plaque agent when compared to control or placebo products. One comparative study demonstrated, however, that the plaque reductions achieved are far below the reductions observed with chlorhexidine. Unlike chlorhexidin, it is not able to prevent the development of gingivitis. It seems that mouth rinses containing sanguinarine are more effective in reducing plaque than dentifrice formulations containing this agent [186].

However, two studies have revealed that the use of sanguinarine mouth rinse was a risk indicator for leukoplakia (odds ratio=10.0; 95% confidence interval=2.0, 89.2), with a strong dose-response relation [151, 152].

#### 5.2.8.2 Propolis

Propolis is a resinous material collected by bees from plant buds and exudates, which is employed for construction and repair of the honeycomb. Several

biological activities have been described for propolis, antimicrobial activity against a wide range of microorganisms (bacteria, fungi, and viruses), but also exerts anti-inflammatory, anesthetic, healing, vasoprotective, antioxidant, antitumoral, antiulcer, and hepatoprotective activities [61, 226].

It was demonstrated that propolis and its fractions possess antibacterial activity against several oral anaerobes, including *Actinobacillus actinomycetemcomitans*, *Fusobacterium nucleatum*, *Porphyromonas gingivalis*, and *Prevotella intermedia* [213–215]. In an animal model systemically administered propolis significantly reduced the periodontitis-related bone, when, prevents alveolar bone loss in the rat model [254]. In patients with chronic periodontitis, subgingival irrigation with propolis extract as an adjuvant to periodontal treatment was more effective than conventional treatment both by clinical and microbiological parameters [86].

Other natural products investigated for their potential antiplaque and antigingivitis effects include: green tea [108, 127], tea tree oil [240] as well as various herbal combinations [69, 99, 100, 192, 220, 241, 262].

### 5.2.9 Oxygenating Agents

Oxygenating agents have been used as disinfectants in various disciplines of dentistry, including endodontics and periodontics [6]. Hydrogen peroxide at concentrations acceptable for oral use in humans (<3%) is unstable and relatively difficult to store for long periods. However, commercially available products are available containing the oxidizing agents sodium peroxyborate or sodium peroxycarbonate. For the former peroxyborate product, there is evidence for efficacy in the treatment of acute ulcerative gingivitis [165].

Controversial results were obtained after the use of oxygenating agents in short- and long-term clinical studies. A double-blind crossover study clearly demonstrated that a mouthwash that releases hydrogen peroxide effectively prevents the colonization of filaments, fusobacteria, motile, and curved rods and spirochetes in developing plaque. The mouthwash which was used as the only oral hygiene measure during a 2-week period (three times daily) in addition, markedly reduced the amount of plaque formed and significantly

retarded gingivitis development [270]. Putt et al. [196] reported that a dual-phase hydrogen peroxide/sodium bicarbonate rinse inhibited gingivitis development (49%) and reduced plaque formation (58%) in a 21-days partial-mouth gingivitis model. Bleeding was also significantly inhibited (36%).

Paraskevas [186] identified three 6-month RCT reporting on various formulations containing peroxides. In a 4-year study of 117 subjects, [274] concluded that there was no difference between the salt and peroxide and conventional oral hygiene regimes with respect to clinical measurements in individuals with mild or moderate periodontitis. Rosin et al. [206] compared the gingival health benefits of a thiocyanate/carbamide peroxide toothpaste to that of a triclosan toothpaste in home use in a two-center, randomized, double-blind, parallel-group clinical trial including 140 healthy volunteers who had at least 20 natural teeth with no probing depths greater than 5 mm and a mean gingival index (GI) of 1 or more. At 6 months, in both groups, gingival health improved and plaque scores decreased significantly between baseline and the following examinations. There were, however, no significant differences between the SCN<sup>-</sup> (rhodanide) and carbamide peroxide RCP and the Triclosan group regarding the investigated parameters. Hasturk et al. [106] evaluated the efficacy of a fluoridated hydrogen peroxide-based mouth rinse on gingivitis and tooth whitening in a two-phase study. The first phase (28 days) included the experimental gingivitis phase; the second phase (5 months) was the oral hygiene phase, which included rinsing. Total of 99 subjects were included in the study and were randomly assigned to receive either placebo or test mouth rinse. At 6 months, Eastman bleeding index, modified gingival index, intensity of stain, and extent of stain were significantly reduced in the test group at 6 months compared to baseline ( $P < 0.05$ ), while in the control group, only the Eastman bleeding index was significantly decreased ( $P < 0.05$ ).

In summary, limited evidence exists with regard to the value of these agents in suppressing supragingival plaque formation although some retardation of plaque growth has been noted with the use of oxygenating mouthwashes. In view of the importance of obligate anaerobic bacteria in the development of gingivitis and periodontitis these compounds deserve further investigation [186].

Oral ulcerations were evidenced in humans after the use of hydrogen peroxide rinses [203].

## 5.2.10 Fluorides

The caries-preventive benefits for a number of fluoride salts are well established, but the fluoride ion has limited effect against the development of plaque and gingivitis [6], and little information is available about the use of fluoride in periodontics (as opposed to preventive and restorative dentistry) [180].

The review performed by Gunsolley [97] showed that the dentifrices containing stannous fluoride [142, 145, 147, 154, 191] exhibited a statistically significant, but small antiplaque effect (mean standardized difference between groups = 0.168). (A mean standardized difference is a mean difference in the active agent's effect minus the control agent's effect adjusted by the variability of each study. This standardization accounts for the difference in variability among multiple studies). Moderately consistent results with regard to the antigingivitis effects were demonstrated in all studies [97].

Similar conclusions were presented by Paraskevas [186] who revealed that most of the studies on  $\text{SnF}_2$  dentifrices seem to agree that that this agent provides some benefits with regard to plaque reductions. However, stannous fluoride toothpaste was shown to be more effective in reducing the gingival index than sodium fluoride toothpaste (weighted mean difference of 0.15 (gingival index) and 0.21 (gingivitis severity index) (test for heterogeneity  $P < 0.00001$ ,  $I^2 = 91.1\%$  and  $P = 0.03$ ,  $I^2 = 80.1\%$ , respectively). With regard to plaque reduction inconsistent results existed. On using the plaque index no differences were found, whereas meta-analysis of the Turesky index provided a WMD of 0.31 ( $P = 0.01$ , test for heterogeneity  $P < 0.0001$ ,  $I^2 = 91.7\%$ ) [189].

The use of amine fluoride alone appears to have no favorable effects on plaque and gingivitis. The combination of  $\text{SnF}_2$  with amine fluoride, however, seems to exert some beneficial antiplaque or antigingivitis effects when it is incorporated in mouth rinse formulations or in combination with a dentifrice also containing the same combination, but the evidence is relatively limited [186].

Root caries development is a common finding associated with surfaces developing recession in patients once treated for periodontal problems. Positive influence of amine fluoride/stannous fluoride and of stannous fluoride on the incidence of root caries in periodontal maintenance patients was reported [22, 59, 118, 180, 183, 184, 185, 202].

## 5.2.11 Amino-Alcohols

**Delmopinol** is a morpholinoethanol deviate with amphiphilic characteristics whinge to its high spreading pressure and its high affinity for interfaces, to reach plaque structures rapidly and efficiently. Delmopinol may reduce plaque and gingivitis, despite being almost devoid of bactericidal or bacteriostatic actions in vitro or in vivo, just by interfering with plaque-matrix formation, reducing the adherence of the primary plaque-forming bacteria or of the successional bacteria [4, 234, 235].

The antiplaque and antigingivitis activity of delmopinol was investigated both in short- and long-term studies which were analyzed by a systematic review [186] and a meta-analysis [4].

Paraskevas [186] identified three double-blind randomized 6-month clinical trials [42, 109, 130] describing the changes of plaque and gingivitis indices associated with the use of delmopinol as well as the safety of the use of this agent. All studies have used a similar design and compared delmopinol with a placebo mouth rinse and 0.2% CHX. Plaque reductions for delmopinol ranged between 9.3% and 35% compared with placebo and gingivitis reductions 1% and 18%. The review concluded that studies seem to agree upon the fact that delmopinol reduces plaque more than placebo. Two of the three studies demonstrated also reduction of gingivitis as opposed to placebo. It seems however, that the plaque and/or gingivitis effectiveness of this agent is far below that of chlorhexidine [186].

Similar results were reported by Addy et al. [4] in a meta-analysis on the effects of 0.2% delmopinol mouth rinse in patients with existing gingivitis. Eight double-blind, parallel-group studies were identified. Study durations ranged from 8 to 24 weeks. Five studies ( $n = 913$ ) involved supervised rinsing; three studies ( $n = 467$ ) involved unsupervised rinsing. These sets of trials were analyzed separately and in combination. The meta-analyses of efficacy were based on data obtained at the end of the 2-month studies and the 3-month point of all the other studies. The differences between active and placebo, for the more important outcome variable of BOP, ranged considerably across the studies from less than 10% to greater than 30%. The results of the meta-analyses confirmed the effectiveness of delmopinol 0.2% in the management of plaque and gingivitis when used in conjunction with usual oral hygiene practices [4].

Adverse signs and symptoms after use of 0.2% delmopinol hydrochloride included transitory numbness of the tongue, tooth and tongue staining, taste disturbance, and, rarely, mucosal soreness and erosion. All local side effects were less commonly reported at 6 compared to 3 months. No systemic effects attributable to the agent were observed and no significant shifts in hematological or biochemical parameters occurred [43, 105].

Octapinol is a substituted amino-alcohol with hydrophilic and hydrophobic properties and with limited antibacterial effects. Octapinol retards development of plaque in humans, the suggested mechanisms being that the adsorption to the pellicle-covered tooth surfaces alters the number of exposed charged groups [219]. However, the product was withdrawn due to toxicological reasons [6].

### 5.2.12 Iodine

Iodine is probably the most potent and broad-spectrum antiseptic agent available. Iodine is able to penetrate the cell walls of microorganisms quickly, and iodine's bactericidal effects probably result from a disruption of protein and nucleic acid structure and synthesis [236]. Iodophores ("iodine carriers") exert excellent antimicrobial activity against bacteria, yeasts, viruses, and protozoans with virtually no evidence of resistance development and are generally nonstaining and relatively free of toxicity and irritancy [236].

The best-known and most widely used iodophore is **povidone-iodine** (PVP-I) (Betadine A, Purdue Frederich Company, Norwalk, CT; different trade names in various countries; generic equivalent is available). Povidone-iodine is a loose complex of elemental iodine and the surfactant povidone (polyvinylpyrrolidone) that improves wetting properties and thereby serves to increase the solubility of the iodine while providing a sustained-release reservoir of the element. Povidone was previously used as a plasma volume expander and exhibits little or no toxicity. Povidone-iodine is available in a range of concentrations from 4% to 10% in aqueous and alcoholic (tincture) formulations. A brown or yellow color indicates that the iodophore is still active. Blue povidone-iodine stains on starched linen will wash off with soap and water. Other iodophore stains can be readily removed with

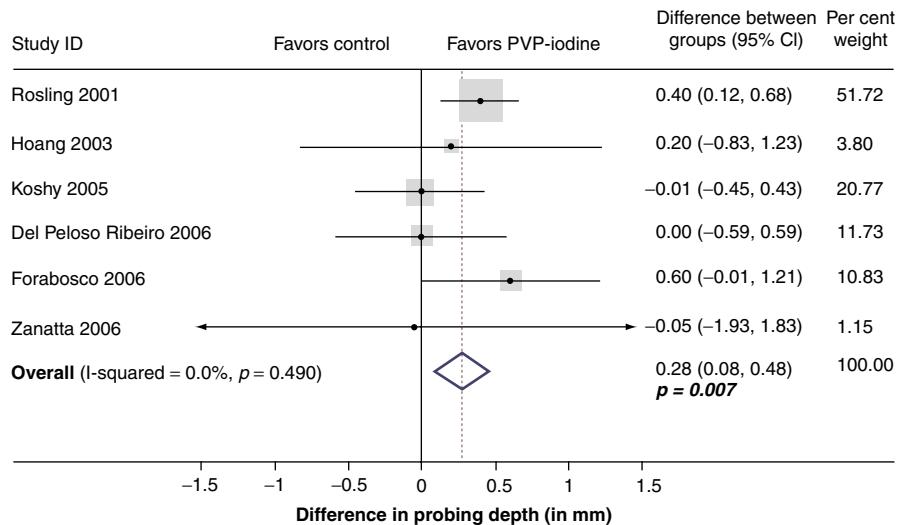
5% sodium thiosulfate solution [236]. PVP-I is microbicidal for Gram-positive and Gram-negative bacteria, fungi, mycobacteria, viruses, and protozoans. Its bacterial activity is due to oxidation of amino (NH<sub>2</sub>), thiol (SH<sub>2</sub>), and phenolic hydroxyl (OH<sub>2</sub>) groups in amino acids and nucleotides. PVP-I also reacts strongly with double bonds of unsaturated fatty acids in cell walls and organelles membranes. PVP-I interacts with cell walls, causing a transient or permanent pore formation. This results in loss of cytoplasmic material and deactivation of enzymes due to direct contact with iodine. PVP-I also was found to cause coagulation of nuclear material without rupturing cell walls [90, 223].

Povidone-iodine in a 5–10% solution seems to overcome the inhibitory effect of serum and can kill the bacteria of experimental biofilms but not on all naturally formed biofilms [236]. Supragingival povidone-iodine (PVP-I) gargle (PVP-I: 0.47 and 0.23% w/v) reduced the viable cell count of eight bacterial strains (*P. gingivalis* ATCC33277 and TDC286, *A. actinomycetemcomitans* ATCC29523 and JP2, *F. nucleatum* No. 2, *Tannerella forsythensis* ATCC43937, *P. intermedia* ATCC25611 and *Streptococcus anginosus* ATCC33397) to below the measurable limit within 15 s [169].

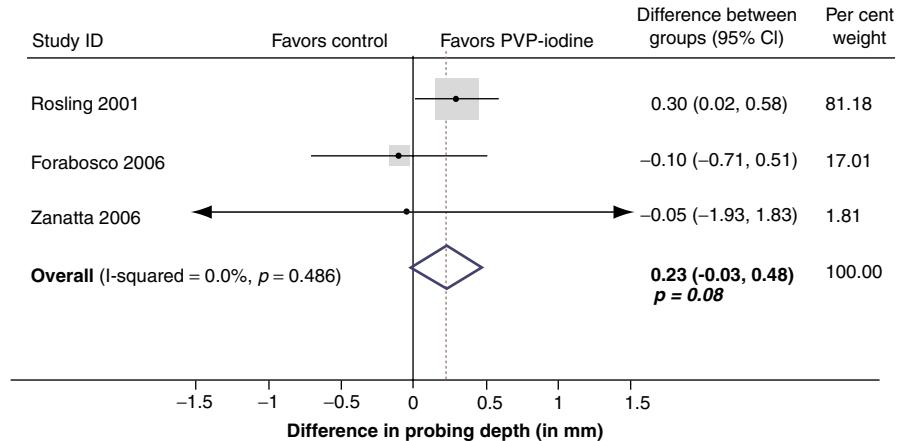
PVP-I is a potent antiseptic and, when used as a component in a rinse with H<sub>2</sub>O<sub>2</sub>, the rinse can decrease the level of gingivitis [41, 90, 150]. The assessment of the additional effect of PVP-I as an adjunct to scaling and root planning compared with water, saline or no rinse in the treatment of chronic periodontitis revealed a small, but statistically significant additional beneficial effect of the adjunctive use of PVP-iodine with enhanced probing depth reductions of 0.28 mm (95% CI: 0.08 to 0.48, *P*=0.007) was found. Effect-size indices ranged from 0.2 to 0.48 (Fig. 5.6). For the 3 months follow-up, the additional effect for PVP-iodine in the meta-analysis was 0.23 mm (95% CI: -0.03 to 0.48) (Fig. 5.7). At the end of the studies, the differences between test and control groups in attachment level changes ranged from -0.13 to 0.95 mm. The mean values of the plaque index varied at baseline from 3.9% to 61% and were reduced equally for test and control sites. Bleeding on probing scores of initially 27–80% dropped without remarkable differences for both groups [212].

For **subgingival irrigation** by means of a syringe, a practical concentration is 10% povidone-iodine providing 1% free iodine with a contact time of at least 5 min.

**Fig. 5.6** Studies investigating an additional effect of povidone-iodine (PVP-iodine), at the study end (Reprinted from [212]. With permission from John Wiley and Sons)



**Fig. 5.7** Studies investigating an additional effect of povidone-iodine (PVP-iodine), 3 months after intervention (Reprinted from [212]. With permission from John Wiley and Sons)



This is generally performed upon completion of each session of scaling and root planning but may also be done prior to mechanical debridement, particularly in medically compromised individuals and in patients with severe gingival inflammation to reduce bacteremia [170, 236]. Hoang et al. [109] pointed out to subgingival irrigation with PVP-iodine as a valuable antimicrobial adjunct to mechanical instrumentation in the management of periodontal infections. At 5 weeks posttreatment, the PVP-I/scaling and root planning group showed a reduction of 95% or more in total pathogens in 43.8% of study sites whereas other treatment groups (scaling and root planning, PVP-iodine and saline alone) revealed similar reductions in only 6.3–12.5% of study sites.

For use with **ultrasonic scalers**, **10% povidone-iodine** is diluted by mixing 1 part of the solution with nine parts or less of water, depending upon patient acceptance [209, 236]. The results of several studies [36, 93, 209] provided some support to the contention that the addition of PVP-I (0.5%) to an ultrasonic device facilitated a significantly better result than ultrasonic debridement with water. The advantage was particularly noted in deep sites ( $\geq 7$  mm) where ultrasonic debridement with PVP-I achieved approximately a 3-mm gain of clinical attachment versus a little less than 2 mm in the other treatment groups. In contrast, more recent reports showed that a concentration of 0.5% PVP-iodine did not add any antimicrobial

effect compared to ultrasonic debridement alone [63, 133, 134, 281].

Povidone iodine has the potential to induce thyroid dysfunction in long-term treatment due to excessive incorporation of iodine. It should not be used during pregnancy or nursing or in individuals at risk for iodine-induced hypothyroidism, including newborns and patients with goiter, Hashimoto's disease, or other underlying thyroid diseases, nor in individuals at risk for iodine-induced thyrotoxicosis, including patients living in areas of iodine deficiency and patients with goiter, Graves' disease or other underlying thyroid disorders [173, 236]. PVP-I should not be used in individuals who are allergic to iodine and its use is contraindicated in pregnant women and nursing mothers [74, 90].

### 5.2.13 Chlorine Compounds

Several antimicrobially active chlorine compounds are commercially available, including hypochlorites, chloramine-T, chlorine dioxide, and various inorganic and organic chlorines. They have a broad spectrum of antimicrobial activity, and many are widely accessible, inexpensive, and fast acting. The likely mechanism of chlorine action is the inhibition of key enzymatic reactions within the microbial cell, protein denaturation, and inactivation of nucleic acids. The active component in hypochlorites is undissociated hypochlorous acid (HOCl). Hypochlorite antiseptic agents in medicine include sodium hypochlorite (aqueous solution) and calcium hypochlorite (solid). Commercial products for the home and health care contain 1% to 15% sodium hypochlorite. The most prevalent products are aqueous solutions of 4% to 6% sodium hypochlorite, which are generally referred to as household bleach. Chlorine compounds are universal antimicrobial agents that are active against a great variety of bacteria, yeasts, and viruses. Sodium hypochlorite is a strong oxidizing agent that possesses numerous attractive properties for antiseptic use, including rapid bactericidal action, ease of use, and very low cost. Disadvantages include irritation of mucous membranes when used in high concentrations, greatly decreased efficacy in the presence of organic matter, corroding effects on some metals, and bleaching of some fabrics. Hypochlorite solutions

gradually lose strength, so that fresh solutions should be prepared daily [236].

A sodium hypochlorite solution for subgingival irrigation at home can be prepared from household bleach that usually contains 5.25%, or 52,500 ppm, of available chlorine. If one part bleach is combined with 49 parts water, the resulting solution will contain an appropriate working concentration of about 0.1% or 1000 ppm of available chlorine. In actual use situations, patients can obtain a working bleach solution by adding 1 teaspoon (5 ml) of household bleach to 250 ml of water (approximately 2 large drinking glasses), and deliver the bleach solution via a commercial oral irrigator at a high pressure setting [236].

Three experimental formulations containing an experimental mouth rinse of (1) Acidified sodium chlorite/malic acid mouth rinse; (2) Acidified sodium chlorite/malic acid mouth rinse buffered; (3) Acidified sodium chlorite/mandelic acid mouth rinse buffered were tested (Alcide Corporation, Redmond, WA. USA) by Yates et al. [278]. The three acidified sodium chlorite (ASC) and chlorhexidine rinses presented similar substantivity and produced similar reductions in salivary bacterial counts, which remained significantly below the placebo control to 7 h. There were no significant differences between ASC and chlorhexidine rinses except at 30 and 60 min when significantly greater reductions were produced by two ASC rinses compared to the chlorhexidine rinse. For plaque area there is no suggestion of any difference in plaque scores between the three acidified sodium chlorite rinses and chlorhexidine ( $P>0.05$ ).

In ASC systems, sodium chlorite generates the microbially active species at an exponentially increasing rate as the pH is lowered. From empirical calculation, an ASC solution at pH 3.0 has only 8.5% of the chlorite available as chlorous acid whereas at pH 2.8 the concentration increases to 12.5%. Thus, when considering the acid-optimization of ASC formulations for potential therapeutic uses, a pH reduction of 0.2 units results in almost 50% greater concentration of the microbially active species. However, to achieve this pH drop, the total titratable acidity must be increased almost 10-fold to that required for a pH 3.0 ASC formulation. Dental erosion could occur as a potential sequel to treatment with such acid-optimized ASC formulations [194].

Recently, a mouthwash containing chlorine dioxide with effects on oral malodor was described [230, 231].

### 5.2.14 Other Antiseptics

**Salifluor** (5-n-octanoyl-3'-trifluoromethyl-salicylanilide), a broad spectrum antimicrobial agent, was investigated for its ability to inhibit dental plaque formation, either alone or in a combination with PVM/MA copolymer and NaF [66]. Three studies performed by [81] were carried out to evaluate the effects of mouth rinses containing 5n-octanoyl-3'-trifluormethylsalicylanilide (salifluor) on plaque and gingivitis. Each trial was performed as a double-blind, randomized and cross-over designed study. Six (control, vehicle control, 0.08%, 0.12%, and 0.2% salifluor and 0.12% chlorhexidine), 3 (control, 0.12% salifluor and 0.12% chlorhexidine) and 3 (control, 0.12% salifluor and 0.12% chlorhexidine) mouthwash preparations were tested in studies 1, 2, and 3, respectively. The findings of study 1, when the volunteers rinsed 2 x daily with various mouthwash preparations for 4 days, indicated that mouth rinses containing salifluor were significantly more effective than control rinses and that (ii) the salifluor mouth rinses were equally effective as the 0.12% chlorhexidine mouth rinse, in retarding 4-day de novo plaque formation. In study 2, after 18 days, the mouth rinse containing 0.12% salifluor retarded de novo plaque formation to the same extent as the 0.12% chlorhexidine mouth rinse at healthy as well as at inflamed sites but the antiplaque effects of the salifluor and chlorhexidine mouth rinses were significantly smaller at sites with inflamed than with healthy gingiva. Study 3 showed that there was no significant difference between the 0.12% salifluor and 0.12% chlorhexidine mouth rinses in retarding de novo plaque formation and the development of gingivitis during a 14-day period of no mechanical plaque control. It was found that salifluor, a highly hydrophobic compound, could not be adequately solubilized with the conventional amount of sodium lauryl sulfate (SLS), the most commonly used anionic surfactant in oral hygiene products, and a combination of salifluor with PVM/MA copolymer and NaF was proposed for increasing its antiplaque effect in mouth rinse and dentifrice formulations [168].

**Hexetidine** is a broad-spectrum antiseptic, active in vitro and in vivo against Gram-positive and Gram-negative bacteria as well as yeasts (*Candida albicans*) [88, 117]. Formulated as a mouthwash, it is available in a number of markets worldwide, with indications for the treatment of a variety of conditions of the

opharynx as well as for management of oral candidiasis associated with denture-induced stomatitis [125, 228]. Hexetidine (0.1%) rinse was effective in inhibiting the development of supragingival plaque and reducing gingival inflammation when used 3x daily for 2 weeks in subjects who temporarily suspended all other oral hygiene measures. For those subjects using hexetidine, a statistically significant inhibition and reduction of supragingival plaque and gingival inflammation with reductions of 6.3%, 33.5%, and 56% for gingivitis, plaque, and gingival bleeding, respectively, were reported [228]. In a later study, Ernst et al. [68] revealed that there were no significant differences after the use of 0.1% chlorhexidine or 0.1% hexetidine, regarding plaque, bleeding, and gingivitis reductions, after 4 weeks of use, with the later one causing less discoloration. The 0.2% hexetidine spray, used as a supplement to regular oral hygiene measures following periodontal surgery showed significant reduction in plaque accumulation and an improvement in wound healing after 28 days of use [25].

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Nonsteroidal anti-inflammatory drugs (NSAIDs) are one of the most commonly prescribed medications. The number of NSAID prescriptions in the USA has been estimated to be 70 million annually; each year, \$6.8 billion are spent on NSAIDs all over the world. The use and cost are expected to increase with time as the life expectancy rises. They are a heterogeneous group of compounds including acetylsalicylic acid (aspirin, ASA). NSAIDs are indicated as analgesics, antipyretic, and anti-inflammatory agents in the treatment of various conditions and diseases. They all seem to share the same pharmacological properties, although their use is often limited to the approved indications by the regulatory agencies. Despite a wide range of pharmacokinetic characteristics, NSAIDs have some common properties. Most NSAIDs are weak organic acids, well absorbed, highly protein-bound, and extensively metabolized. NSAIDs exert their effect with inhibition of cyclooxygenase (COX) enzyme [55,171–174].

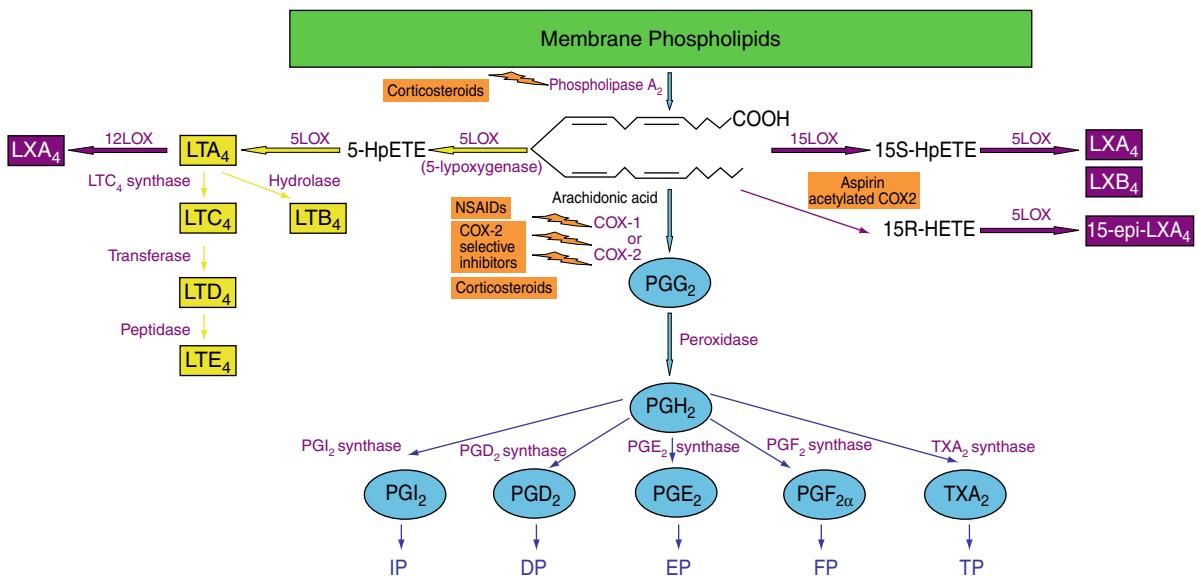
## 6.1 Pathways of Arachidonic Acid Metabolism

Arachidonic acid, an unsaturated 20-carbon fatty acid embedded in cell membranes as a phospholipid ester, is the precursor for PG synthesis. In response to a wide variety of stimuli, free arachidonic acid is released, which is subsequently converted via cyclooxygenase (COX), lipoxygenase, and cytochrome P450 enzyme catalysis to various lipid mediators collectively known as eicosanoids [136,142].

Phospholipids, present in cell membranes, are metabolized to arachidonic acid by phospholipase A2. **In the COX pathway**, the two known COX isoforms convert arachidonic acid to PGG<sub>2</sub> (prostaglandin) and

consequently to PGH<sub>2</sub>. Depending on the type of tissue and stimuli, PGH<sub>2</sub> is further converted by tissue-specific isomerases to five major prostanoids: PGD<sub>2</sub>, PGE<sub>2</sub>, PGI<sub>2</sub> (prostacyclin), PGF<sub>2α</sub>, and TXA<sub>2</sub> (thromboxane A<sub>2</sub>), which bind to specific receptors termed DP, EP, IP, FP, and TP, respectively [10, 28, 42, 55, 95]. Prostanoids (PGs) are end products of fatty acid metabolism produced via the COX pathway. PGs have long been known to behave as important physiological and pathological mediators implicated in a number of therapeutic areas of interest, including inflammation, pain, pyrexia, cancer, glaucoma, male sexual dysfunction, osteoporosis, cardiovascular disease, labor, and asthma [142].

COX enzymes are involved in the production of PGs and TX from arachidonic acid (Fig. 6.1). COX-1 and COX-2 are two isoforms of COX enzyme. It has been proposed that COX-1 and COX-2 subserve different physiologic functions largely because of the striking differences in their tissue expression and regulation (Table 6.1; Figs. 6.2, 6.3). COX-1 displays the characteristics of a “housekeeping” gene, is constitutively expressed in almost all tissues, and is involved in many physiological functions such as regulation of platelet aggregation, gastric mucosa protection, and maintenance of renal function. COX-1 appears to be responsible for the production of prostaglandins (PG) that are important for homeostatic functions, such as maintaining the integrity of the gastric mucosa, mediating normal platelet function, and regulating renal blood flow. In sharp contrast, COX-2 is the product of an “immediate–early” gene that is rapidly inducible and tightly regulated. Under basal conditions, COX-2 expression is highly restricted; however, COX-2 is dramatically upregulated during inflammation. Expression of COX-2 is governed by several biological factors. Growth factors and pro-inflammatory cytokines, such as IL-1 and TNF-α, increase the levels of COX-2,



**Fig. 6.1** Pathways of arachidonic acid metabolism. *COX-1*: cyclooxygenase-1; *COX-2*: cyclooxygenase-2; *PG* prostaglandin; *PGH*<sub>2</sub> prostaglandin H<sub>2</sub>; *PGI*<sub>2</sub> prostaglandin I<sub>2</sub>; *PGD*<sub>2</sub> prostaglandin D<sub>2</sub>; *PGE*<sub>2</sub> prostaglandin E<sub>2</sub>; *PGF*<sub>2α</sub> prostaglandin

*PGF*<sub>2α</sub>; *TXA*<sub>2</sub> thromboxane A<sub>2</sub>; 5-*HPTE* 5-hydroperoxyeicosatetraenoic acid; *LT* leukotriene; *LX* lipoxin; *LOX* lipoxygenase; *HpETE* hydroperoxyeicosatetraenoic acid; *HETE* hydroxyeicosatetraenoic acid

**Table 6.1** Comparison of human cyclooxygenase-1 and cyclooxygenase-2 properties [105] (Reprinted with permission from John Wiley and Sons)

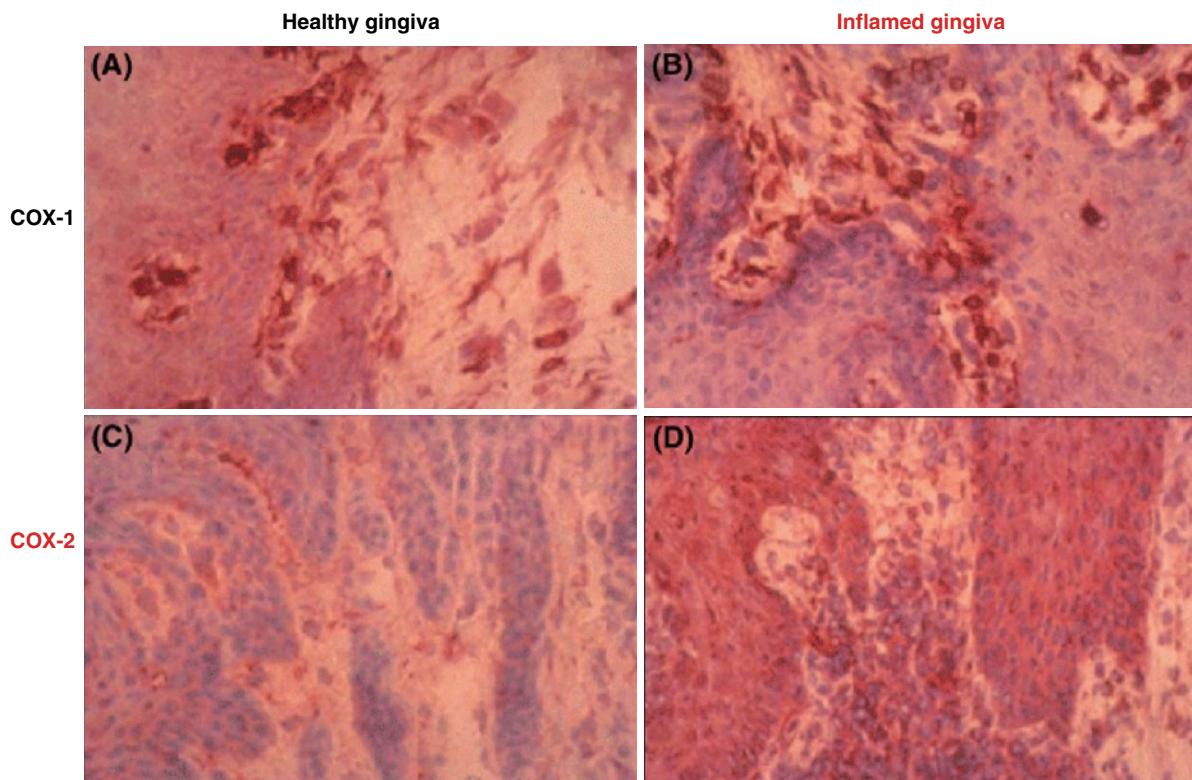
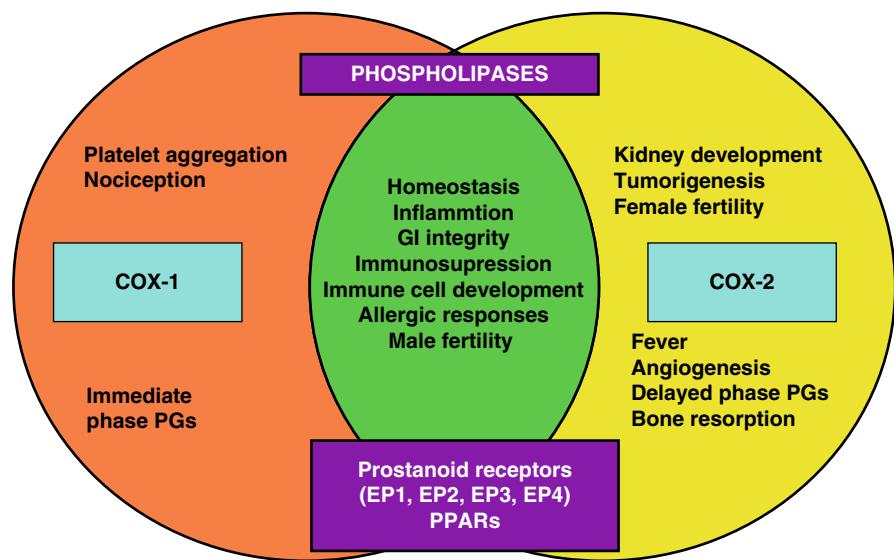
| Properties            | Cyclooxygenase-1                     | Cyclooxygenase-2   |
|-----------------------|--------------------------------------|--|
| Enzyme expression     | Constitutive                         | Inducible  |
| Character of gene     | House-keeping gene                   | Immediate early gene   |
| Locus                 | 9q32–q33.3                           | 1q25.2–q25.3   |
| Size of gene          | 22 kb                                | 8.3 kb   |
| Number of amino acids | 576 amino acids                      | 604 amino acids  |
| 5'-flanking region    | No TATA, GC rich, Sp1                | Nuclear factor-κB, nuclear factor-IL-6, cyclic AMP-response element, E-box, TATA box   |
| Size of RNA           | 2.8 kb                               | 4.6 kb   |
| Expressing cells      | Most cells                           | Not detected in normal conditions. Increased in fibroblasts, monocytes, osteoblasts by IL-1, tumor necrosis factor α, lipopolysaccharide, etc. |
| Glucocorticoid effect | No or slight effect of transcription | Inhibition of transcription  |

whereas corticosteroids and anti-inflammatory cytokines such as IL-4, IL-10, and IL-13 inhibit the expression of COX-2. Constitutive expression of COX-2 in brain, stomach, colon, and kidney of mammals is also reported [2, 31, 55, 63, 129, 130, 170].

Acetaminophen is generally considered an NSAID, but its mechanism of action has not been fully resolved.

It is a weak inhibitor of isolated COX-1 and COX-2, but demonstrates antipyretic and analgesic properties when taken internally. It has no anti-inflammatory actions. Most recently, a splice variant of COX-1 mRNA, retaining intron 1, and given the names COX-3, COX-1b, or COX-1v, has been described. COX-3 mRNA has been isolated in many tissues including

**Fig. 6.2** Tissue distribution and pathological function of COX-1 and COX-2  
(Reprinted from [41]. With permission from Elsevier)



**Fig. 6.3** Immunohistochemical staining of cyclooxygenase-1 (COX-1) and cyclooxygenase-2 (COX-2) proteins in clinically healthy and inflamed human gingiva. In both gingiva, COX-1-immunoreactive cells were detected in subepithelial connective tissue including fibroblasts and endothelial cells and some gingival epithelial cells were slightly immunopositive for COX-1 (a, c). In inflamed gingiva, inflammatory cells were also immuno-

negative for COX-1. However, COX-2 protein was detected in fibroblasts, gingival epithelial cells, endothelial cells, and inflammatory cells in inflamed gingiva, whereas in clinically healthy gingiva, it was slightly detected in gingival epithelial cells and fibroblasts (b, d). Magnification  $\times 400$  (Reprinted from [105]. With permission from John Wiley and Sons) view within article

canine and human cerebral cortex, human aorta, and rodent cerebral endothelium, heart, kidney, and neuronal tissues [61]. In dogs, intron-1 is 90 nucleotides in length and represents an in-frame insertion into the portion of the COX-1 open reading frame encoding the N-terminal hydrophobic signal peptide. This variant produces enzyme protein containing the encoded intron-1 sequence when expressed in insect cells. The activity of the protein is preferentially inhibited by analgesic antipyretic drugs such as acetaminophen, and may explain the therapeutic actions of this class of compounds [19,20]. However, some authors do not consider COX-3 as a distinctive enzyme in humans [12].

In the **lipoxygenase pathway**, the arachidonic acid transforming enzyme 5-lipoxygenase (5-LOX) catalyzes the conversion of arachidonic acid into 5(S)-hydroperoxyeicosatetraenoic acid (5-HPETE) and leukotriene (LT) A<sub>4</sub> (Fig. 6.1). The unstable intermediate LTA<sub>4</sub> can be further converted into LTB<sub>4</sub> by LTA<sub>4</sub> hydrolase or into LTC<sub>4</sub> by LTC<sub>4</sub> synthase, and the LTC<sub>4</sub> synthase isoenzymes MGST<sub>2</sub> or murine MGST<sub>3</sub>. The expression of 5-LO is tightly regulated. Thus, 5-LO is mainly present in mature leukocytes, including granulocytes, monocytes/macrophages, mast cells, and B-lymphocytes or dendritic cells, and the capability of cells to express 5-LO is acquired during cell maturation [160, 187, 188]. Whereas LTB<sub>(4)</sub> is a potent chemotactic and chemokinetic agent for a variety of leukocytes, the cysteinyl-leukotrienes C, D<sub>(4)</sub>, and E<sub>(4)</sub> cause vascular permeability and smooth muscle contraction [188].

Collectively, these molecules make up the cysteinyl *leukotrienes*. Leukotrienes act on target cells, which may be leukocytes, epithelial cells, smooth-muscle cells, or endothelial cells, by interacting with one or both classes of their cognate receptors. B leukotriene receptor 1 (BLT<sub>1</sub>) is expressed primarily on leukocytes and is a high-affinity receptor, whereas B leukotriene receptor 2 (BLT<sub>2</sub>) is expressed more ubiquitously, has a somewhat lower affinity for LTB<sub>4</sub>, and can bind other lipids. The two cysteinyl leukotriene receptors have a broad distribution. All leukotriene receptors activate the Gq class of G proteins, resulting in increased intracellular calcium, the Gi class, resulting in decreased intracellular cyclic AMP (cAMP), or both. These effects, which activate downstream protein kinases, culminate in myriad cellular and tissue responses [127].

Leukotrienes have been identified as mediators of a variety of inflammatory and allergic reactions including rheumatoid arthritis, inflammatory bowel disease, psoriasis, and allergic rhinitis, but their major pathophysiological implication was linked to bronchial asthma. Recently, the 5-LOX pathway has also been associated with atherosclerosis, osteoporosis, and certain types of cancers like prostate cancer [92, 147, 187, 188].

Late in the inflammatory process, when there is a high concentration of cells containing lipoxygenases and corresponding proinflammatory products, such as prostaglandin E<sub>2</sub>, a “class switch” may occur within neutrophils. This class switch gives rise to the synthesis of proresolving molecules through pathways that are spatially and temporally distinct from those involved in the generation of proinflammatory lipid mediators [168, 169]. The term *lipoxins* (LXs) describes the provenance of these mediators: *lipoxygenase interacting products*. The LXs are eicosanoids, and display both anti-inflammatory and pro-resolving bioactions. More recently, other pro-resolving lipid mediators have been described including the resolvins and neuroprotectins. LXs are typically formed **by transcellular metabolism of arachidonate**-involving sequential lipoxygenase activity within an inflammatory milieu. Epithelial-monocyte 15-LOX activity produces 15(S) hydroperoxyeicosatetraenoic acid from arachidonic acid, which can then be converted by neutrophil 5-LOX to generate LXA<sub>4</sub> [5S,6R,15S-trihydroxyl-7,9, 13-trans-11-cis-eicosatetraenoic acid]. Production of LXA<sub>4</sub> by this pathway diverts metabolism of arachidonic acid from biosynthesis of pro-inflammatory LTs. LXA<sub>4</sub> can also be generated by the actions of platelet 12-LOX to convert the 5-LOX epoxide product LTA<sub>4</sub> to LXA<sub>4</sub> and its positional isomer LX<sub>B4</sub>. Aspirin acetylation of COX-2 in endothelial and epithelial cells inhibits the formation of prostaglandins and thromboxanes, while shifting its enzymatic activity toward the generation of 15R-HETE, which is converted by leukocyte 5-LOX to generate 15-epi-LX-designated aspirin-triggered LX that is ATL [144, 155].

The lipoxins produced act as agonists to stimulate the resolution of inflammation and promote the restoration of tissue homeostasis through a number of mechanisms involving the regulation of leukocyte function. These include limiting polymorphonuclear neutrophil (PMN) migration into sites of inflammation, activating

monocytes with a nonphlogistic phenotype (e.g., without the generation of a superoxide anion), and stimulating the uptake of apoptotic PMNs by macrophages [168, 169].

Apparently, using the same metabolic pathways, omega-3 fatty acids in the diet (eicosapentaenoic acid, C20:5; docosahexaenoic acid, C22:6) are the substrate for the formation of a class of molecules that have been termed the resolvins (derived from eicosapentaenoic acid) and protectins (derived from docosahexaenoic acid). These molecules bind to distinct receptors on inflammatory cells, but with some exceptions, their overall actions are the same; the promotion of resolution of inflammation. The main receptors for resolvins are the chemR23 receptors expressed on macrophages and leukotriene-4-receptor 1 substrate for the formation of a class of molecules that have been termed the resolvins and docosatrienes. These molecules bind to distinct receptors on inflammatory cells, but with some exceptions, their overall actions are the same; the promotion of resolution of inflammation. The main receptors for resolvins are the chemR23 receptor expressed on macrophages and leukotriene-4-receptor 1 [154, 156, 168].

## 6.2 Arachidonic Acid Pathway and Periodontal Disease

While COX-1 is constitutively expressed in almost all organs of the human body, COX-2 expression is specifically induced by cytokines. In periodontitis, specific COX-2 expression has been reported in gingival tissues [26, 94, 100, 101, 199–201], suggesting that cyclooxygenase-2 plays a crucial role for prostaglandin E<sub>2</sub> production in periodontal disease [105].

Polymorphisms within the COX-2 gene have repeatedly been implicated as increasing susceptibility to inflammatory diseases [30, 78, 122]. In a European ethnicity population, the rare G-allele of htSNP rs6681231 was associated with aggressive periodontitis prior to and after adjustment for the covariates smoking, diabetes, and gender, with an odds ratio of 1.57 (95% confidence interval 1.18–2.08;  $P=0.002$ ) [148]. In a Chinese population, it was found that the –1195A single nucleotide polymorphisms of COX-2 was a meaningful indicator for severe chronic periodontitis, and the

haplotype AGT was significantly associated with the risk of severe chronic periodontitis [196]. Ho et al. [62] suggested that the –765G to C polymorphism of the COX-2 gene is associated with a decreased risk for periodontitis in Taiwanese population, especially in aggressive periodontitis. The ORs for carriage of the –765C allele (GC+CC versus GG) in aggressive and chronic periodontitis were 0.068 (95% CI: 0.020–0.173,  $P<0.0001$ ) and 0.571 (95% CI: 0.385–0.849,  $P=0.006$ ), respectively. After adjustment for age, gender, and smoking status, the OR was 0.071 (95% CI: 0.017–0.219) and 0.552 (95% CI: 0.367–0.829) for aggressive and chronic periodontitis, respectively.

Prostanoids, including **prostaglandins** and thromboxane, have a variety of roles in physiological and pathological conditions including inflammation, immunological function, ovulation, implantation, cardiovascular disease, and tumorigenesis [105, 189]. Prostaglandin E<sub>2</sub> is formed as a result of the metabolism of arachidonic acid. PGE<sub>2</sub> has been shown to have many proinflammatory effects, including increased vasodilatation, enhanced responsiveness of receptors to painful stimuli, release of collagenase by inflammatory cells, and activation of osteoclasts [86].

Many studies reported that that prostaglandin E<sub>2</sub> levels are elevated in periodontal tissue and GCF in patients suffering from severe forms of diseases (juvenile and refractory periodontitis) compared to healthy controls or patients suffering from a mild form of the disease (gingivitis or chronic adult periodontitis) [35, 37, 51, 102, 111, 112, 115, 116, 118, 202].

Offenbacher et al. [116] analyzed, in a 3-year longitudinal study, the possibility to use PGE<sub>2</sub> values in gingival crevicular fluid as an indicator of ongoing periodontal tissue destruction and as a future predictor of acute periodontal attachment loss. Samples from 41 adult periodontitis patients were collected intrasurgically at the mesiofacial line angle of every tooth excluding third molars. The patients were monitored longitudinally every 3 months until a statistically significant episode of attachment loss, of at least 3 mm in magnitude, occurred at any periodontal site in the patient's mouth. Twenty-four patients did not have any significant attachment loss, which demonstrated a PGE<sub>2</sub> concentration in gingival crevicular fluid of  $50.1\pm7.1$  ng/mL, whereas the 17 patients who had experienced disease activity, demonstrated a mean PGE<sub>2</sub> concentration in gingival crevicular fluid of  $113.4\pm9.0$  ng/mL. These results revealed that PGE<sub>2</sub>

concentration in gingival crevicular fluid can predict clinical attachment level loss 6 months before its occurrence. At the individual sites level, the mean PGE<sub>2</sub> concentration in gingival crevicular fluid at attachment lost sites was elevated compared with contralateral control sites which did not experience attachment loss ( $305.6 \pm 56.5$  ng/mL versus  $65.7 \pm 6.9$  ng/mL). The evaluation of PGE<sub>2</sub> concentration in gingival crevicular fluid as screening test to predict attachment loss demonstrated a sensitivity of 76% and a specificity of 96%. This would indicate that the determination of GCF prostaglandin E<sub>2</sub> levels is a good indicator of inflammatory activity [29].

In addition, various periodontal treatment therapies induced a decrease in GCF prostaglandin E<sub>2</sub> levels [3, 32, 81, 91, 113, 133, 197].

**Thromboxane** is a member of the family of lipids known as eicosanoids. The two major thromboxanes are thromboxane A<sub>2</sub> and thromboxane B<sub>2</sub>. Thromboxane A<sub>2</sub> (TXA<sub>2</sub>), produced by activated platelets, has pro-thrombotic properties, stimulating activation of new platelets as well as increasing platelet aggregation. Thromboxane B<sub>2</sub> is an inactive metabolite/product of thromboxane A<sub>2</sub>. In a ligature-induced periodontitis in *Macaca mulatta*, a statistically significant threefold increase in gingival crevicular fluid levels of prostaglandin E<sub>2</sub> (PGE<sub>2</sub>) and thromboxane B<sub>2</sub> (TxB<sub>2</sub>) were reported at 3 months, as compared to baseline, which positively correlated with increases in redness, bleeding, attachment level loss, and bone loss [114]. Similar results were reported in experimental periodontitis in beagle dogs, where crevicular fluid thromboxane B<sub>2</sub> levels rapidly reached a four- to fivefold peak over baseline at 1 month [117]. The animals with gingivitis-contained gingival TxB<sub>2</sub> levels nearly twice as the normal gingiva, while animals with periodontitis revealed gingival levels of TxB<sub>2</sub> that were 3.5 times greater than clinically normal and twice that of dogs with gingivitis [141]. The selective elevation of both PGE<sub>2</sub> and TxB<sub>2</sub> in ligated sites, compared with levels in spontaneous sites, in the presence of similar levels of LTB<sub>4</sub> and IL-1,8 provides further evidence that these molecules regulate the magnitude of the tissue-destructive response in progressive periodontitis [159].

Human inflamed gingival tissue synthesized significantly larger amounts, compared to normal tissue, of 6-keto-PGF<sub>1</sub> alpha ( $P < 0.05$ ), thromboxane B<sub>2</sub> ( $P < 0.01$ ), PGD<sub>2</sub> ( $P < 0.05$ ), and PGA<sub>2</sub> ( $P < 0.001$ ) [97]. TxB<sub>2</sub> was found only in tissues from deeper periodontal sites

where measured levels were as high as 210 pg/mg. However, only 36% of samples from deep periodontal sites had detectable levels [35]. We found that concentrations of TxB<sub>2</sub> in gingivitis sites are at mean tenfold higher than in the healthy sites and histologically show a consistently intracytoplasmic staining with a significant increase in gingivitis. This argues in favor of a local production of TxB<sub>2</sub>. Concentrations of TxB<sub>2</sub> at periodontitis sites are only threefold higher than in healthy sites and histologically show a stronger staining as for the gingivitis sections, with principally an extracellular localization. Thus, TxB<sub>2</sub> could be released in large quantities in the crevicular fluid when the periodontitis stage has reached [18].

**Leukotrienes** are biologically active compounds that are of importance in host defense reactions, and they have a pathophysiologic role in inflammation and allergic reactions. Leukotrienes collectively induce vascular permeability and chemotaxis of leukocytes and cause intense vasoconstriction and bronchoconstriction [131]. Leukotriene B<sub>4</sub> is a potent mediator of inflammation derived from arachidonic acid by the sequential actions of 5-lipoxygenase (5-LOX) and LTA<sub>4</sub> hydrolase (LTA<sub>4</sub>H). LTB<sub>4</sub> acting through its receptors (BLT1) can cause chemotaxis, degranulation, adhesion, and enhance the survival of neutrophils. Although BLT1 was long known to be a neutrophil chemoattractant receptor, recent studies identified BLT1 expression on macrophages, smooth muscle cells, endothelial cells, activated T-cells, and mast cells considerably expanding the potential role of LTB<sub>4</sub> [96]. Various inflammatory diseases, including asthma, allergic rhinitis, atopic dermatitis, allergic conjunctivitis, rheumatoid arthritis, chronic obstructive pulmonary disease (COPD), obliterative bronchiolitis after lung transplantation, and interstitial lung diseases are associated with increased levels of LTB<sub>4</sub> and/or BLT1 expression and in some of these diseases LTB<sub>4</sub> levels reflect disease activity and are decreased by treatment [119, 134].

Levels of LTB<sub>4</sub> in GCF from adult periodontitis and non-periodontitis sites were evaluated by [38] to demonstrate its possible role in periodontal disease progression. A greater concentration of LTB<sub>4</sub> was found in adult periodontitis sites compared to the non-periodontitis sites. Higher concentrations of LTB4 were revealed in subjects with chronic periodontitis compared to generalized aggressive periodontitis, localized aggressive periodontitis, gingivitis, and healthy groups. Similar

results were reported by [131], as the mean concentration of LTB<sub>4</sub> increased progressively from health ( $39.6 \pm 11.55$  pg/mL) to periodontitis ( $185.2 \pm 54.99$  pg/mL), and the mean concentration of LTB<sub>4</sub> in gingivitis was  $97.1 \pm 37.75$  pg/mL. As [166] showed, in adult periodontitis patients, the LTB<sub>4</sub> concentration was correlated with probing depth and gingival index, while the LTB<sub>4</sub> total amount was correlated with four clinical parameters: plaque index, probing depth, clinical attachment loss, and gingival index.

An increased leukotriene production in periodontitis and in addition detected higher levels of cysteinyl-leukotrienes in GCF from atherosclerotic subjects both with and without periodontitis was revealed by [9]. The authors suggested that high levels of cysteinyl-leukotrienes in the GCF may be a marker of increased risk, not only for periodontitis, but also for the development of atherosclerosis.

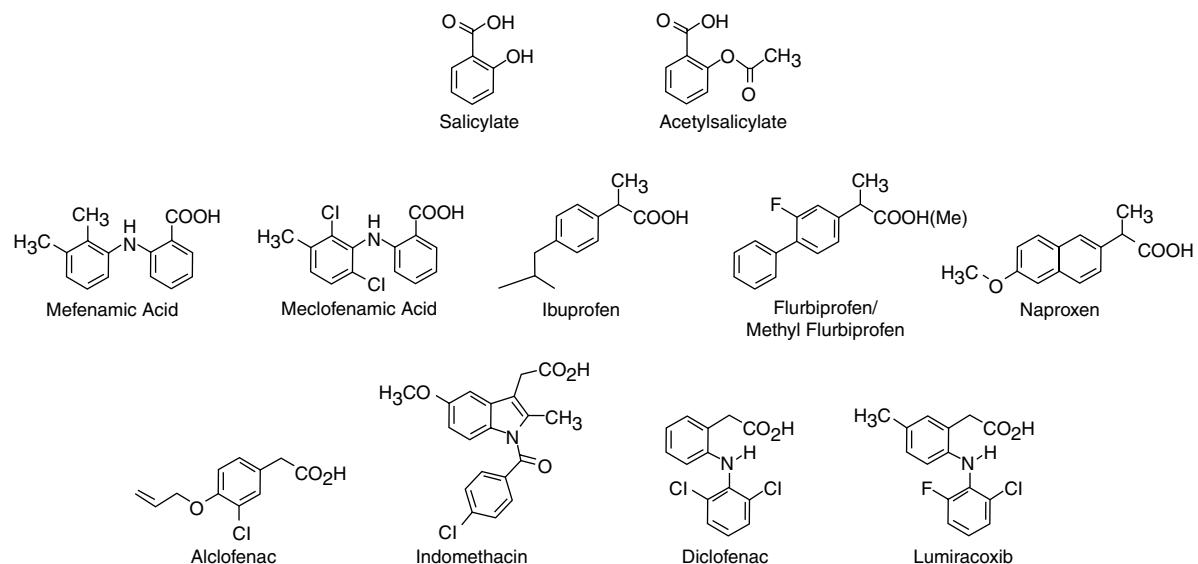
### 6.3 Classification of NSAIDs

NSAIDs are among the most widely used medications in the world because of their demonstrated efficacy in reducing pain and inflammation. Their efficacy has

been documented in a number of clinical disorders, including osteoarthritis, rheumatoid arthritis, ankylosing spondylitis, gout, dysmenorrhea, dental pain, and headache [16, 41].

A variety of NSAIDs can block the enzymatic activity of COX; they vary in their chemical structure and relative ability to block the COX-1 versus the COX-2 isoenzymes (Fig. 6.4). The COX-2 inhibitors also vary in their selectivity for the COX-2 versus the COX-1 enzyme. Highly COX-2 selective are Etoricoxib (Algix, Arcoxia, Tauxib), Lumiracoxib (Prexige), Parecoxib (Dynastat), Rofecoxib (Vioxx, Merck Sharp & Dohme withdrawn from the market September 30, 2004), Valdecoxib (Bextra, Pfizer withdrawn from the market April 7, 2005), while the moderately COX-2 selective are Celecoxib (Artiglog, Artrid, Celebrex, Solexa), Etodolac (Lodine), and Meloxicam (Mobic) [34]. The differences in the biological effects of COX inhibitors are a consequence of the degree of selectivity for COX-2 versus COX-1 and tissue-specific variations in the distribution of COX and related enzymes that convert prostaglandin H<sub>2</sub> into specific prostanoids [5].

For example, several prostanoids, including prostaglandin E<sub>2</sub> and prostacyclin, are both hyperalgesic (elicit an increased sense of pain) and gastroprotective.



**Fig. 6.4** Chemical structures of salicylate, acetylsalicylic acid (aspirin), and several key phenylpropionic and arylacetic acid inhibitors (Reprinted from [16]. With permission, copyright 2010 American Chemical Society)

Thus, nonselective COX inhibition with agents such as aspirin, ibuprofen, indomethacin, and naproxen, which inhibit both COX-1 and COX-2 enzymes, provides effective pain relief for inflammatory conditions, but carries with it a risk for erosive gastritis and gastrointestinal bleeding. Selective COX-2 inhibitors (valdecoxib, rofecoxib, celecoxib, etc.) were developed to minimize gastrointestinal toxicity because of the relative paucity of COX-2 expression in the gastrointestinal tract and the relative abundance of COX-2 expression in inflamed and painful tissues [5].

Shortly after their introduction into the market, it was clear that coxibs (selective COX-2 inhibitors) might cause adverse effects in the renal and cardiovascular systems. Rofecoxib has then been withdrawn from the market by Merck, following the premature cessation of the Adenomatous Polyp Prevention on Vioxx (APPROVe) study, which was designed to determine the drug's effect on benign sporadic colonic adenomas. This study demonstrated a significant increase by a factor of 3.9 in the incidence of serious thromboembolic adverse events in the group receiving 25 mg of rofecoxib per day as compared with the placebo group. Blood pressure was elevated in patients in the rofecoxib group early in the course of the study, but the incidence of myocardial infarction and thrombotic stroke in the two groups began to diverge progressively after a year or more of treatment [23,41].

In the Vioxx Gastrointestinal Outcomes Research (VIGOR) trial, the rate of serious gastrointestinal events among those receiving rofecoxib was half that among those receiving a traditional NSAID, naproxen – 2%, as compared with 4%. However, a significant increase by a factor of 5 in the incidence of myocardial infarction was observed [17]. Celecoxib, a COX-2-specific inhibitor was approved by the US Food and Drug Administration (FDA) for symptomatic treatment of rheumatoid arthritis and osteoarthritis. The aim of the Celecoxib Long-Term Arthritis Safety Study (CLASS trial) was to determine whether celecoxib, a COX-2-specific inhibitor, is associated with a lower incidence of significant upper gastrointestinal (GI) toxic effects and other adverse effects compared with conventional NSAIDs (ibuprofen or diclofenac). In this study, celecoxib, at dosages greater than those indicated clinically, was associated with a lower incidence of symptomatic ulcers and ulcer complications combined,

as well as other clinically important toxic effects, compared with NSAIDs at standard dosages. The decrease in upper GI toxicity was strongest among patients not taking aspirin concomitantly [158]. The Therapeutic Arthritis Research and Gastrointestinal Event Trial (TARGET) aimed to assess gastrointestinal and cardiovascular safety of the COX2 inhibitor lumiracoxib compared with two nonsteroidal anti-inflammatory drugs, naproxen and ibuprofen. Lumiracoxib showed a three- to fourfold reduction in ulcer complications compared with nonsteroidal anti-inflammatory drugs without an increase in the rate of serious cardiovascular events, suggesting that lumiracoxib is an appropriate treatment for patients with osteoarthritis [40,150]. Additional data from randomized controlled trials of COX-2-selective agents have been reported and summarized in meta-analyses, which has reinforced the concern about cardiovascular events with COX-2 inhibitors (coxibs) [5,75].

At present, selective inhibitors of COX-2 appear a rational choice for patients at a low cardiovascular risk who have had serious gastrointestinal events. In these patients, however, a cost-effective alternative is the use of a traditional NSAIDs associated with a co-medication with a low-cost proton pump inhibitor. It would also seem prudent to avoid coxibs in patients who have cardiovascular disease or who are at risk for it [41].

## 6.4 Effect of NSAIDs on Periodontal and Peri-implant Disease Progression

Over decades, several in vivo animal and human clinical studies have evaluated the NSAIDs as inhibitors of the host response in periodontal disease.

### 6.4.1 Animal Studies

As summarized in Table 6.2, it was revealed that NSAIDs delayed the onset and suppressed the magnitude of the acute inflammatory response and decreased the amount of alveolar bone resorption in different

**Table 6.2** Animal studies on adjunctive NSAIDs therapy in periodontal disease

| Reference            | Animal model  | Substrate  | NSAIDs  | Type of administration | Length of the study | Results   |
|----------------------|---|--|---|------------------------|---------------------|---|
| Azoubel et al. [7]   | Experimental ligature-induced periodontitis in rats | Alveolar bone loss   | Etoricoxib (selective COX-2 inhibitor)<br>Indomethacin (nonselective COX inhibitor) | Systemic               | 11 days             | The ongoing ABL was significantly inhibited ( $P < 0.05$ ) by 3 and 9 mg/kg etoricoxib and by indomethacin: control = $4.08 \pm 0.47$ mm; etoricoxib (3 mg/kg) = $1.89 \pm 0.26$ mm; etoricoxib (9 mg/kg) = $1.02 \pm 0.14$ mm; indomethacin = $0.64 \pm 0.15$ mm. Histopathology of periodontium showed that etoricoxib and indomethacin reduced inflammatory cell infiltration, ABL, and cementum and collagen fiber destruction. Animals that received indomethacin presented weight loss starting on the seventh day, and higher mortality rate (58.3%) compared to etoricoxib (0%). In summary, treatment with etoricoxib, even starting when ABL is detected, reduces inflammation and cementum and bone resorption, with fewer gastrointestinal side effects |
| Bezerra et al. [14]  | Experimental ligature-induced periodontitis in rats | Cell influx<br>Osteoclast numbers<br>Alveolar bone loss<br>Cementum integrity  | Indomethacin<br>Meloxicam (type 2 COX inhibitor)                                    | Systemic               | 7 days              | In the nontreated group, there was significant ABL, severe mononuclear influx, and an increase in osteoclast numbers. Significant neutrophilia and lymphomonocytosis occurred at 6 h and at 7 days, respectively, as compared to controls. NSAIDs regimens reduced ABL and histopathologic changes. Neutrophilia and lymphomonocytosis were also significantly reversed. Meloxicam administration resulted in less gastric damage compared with indomethacin treatment  |
| Carvalho et al. [25] | Experimental Ligature-induced periodontitis in rats | ABL evaluated with histological techniques (alveolar bone loss and cellularity), enzyme immunoassay (lipoxin A4), intravital microscopy (rolling leukocytes and endothelial-leukocyte adhesion), | Piroxicam<br>Mangiferin   | Systemic               | 7 days              | Only the group of rats treated with mangiferin was able to significantly reduce ( $P < 0.05$ ) ABL ( $105.5 \pm 33.1$ $\mu\text{m}$ ) when compared to the vehicle-treated group ( $217.6 \pm 18.9$ $\mu\text{m}$ ). Piroxicam at the dose of 20 mg/kg was unable to prevent the appearance of periodontitis 24 h after its induction. After 4 days of daily treatment, both the groups treated with piroxicam and mangiferin significantly reduced ( $P < 0.001$ ) the ABL ( $167.2 \pm 13.4$ $\mu\text{m}$ vs. $162.2 \pm 11$ $\mu\text{m}$ , respectively) when compared to saline-treated animals ( $290.4 \pm 20$ $\mu\text{m}$ )  |

(continued)

**Table 6.2** (continued)

| Reference              | Animal model  | Substrate | NSAIDs  | Type of administration | Length of the study | Results   |   |
|------------------------|---|-----------|---|------------------------|---------------------|---|---|
| Gurgel et al. [52]     | Experimental ligature-induced periodontitis in rats | ABL       | Meloxicam   | Systemic               | 45 days             | The groups that received the COX-2 inhibitor for 15 days ( $3 = 5.83 \pm 2.04 \mu\text{m}^3$ ) and 45 days ( $4 = 3.59 \pm 1.57 \mu\text{m}^3$ ) showed a significant reduction in the volume of bone loss when compared to the groups that received saline solution for the same period ( $1 = 9.01 \pm 3.16 \mu\text{m}^3$ , $2 = 6.86 \pm 3.59 \mu\text{m}^3$ ). After drug withdrawal, no remaining effect was observed. With respect to the posttreatment effect (group 5 = $6.09 \pm 2.66 \mu\text{m}^3$ ), intergroup analysis did not show any significant difference in the volume of bone loss ( $P > 0.05$ ) when compared to the control group ( $6.86 \pm 3.59 \mu\text{m}^3$ , group 2, respectively) |   |
| Holzhausen et al. [64] | Experimental ligature-induced periodontitis in rats | ABL       | Celecoxib   | Systemic               | 30 days             | Groups treated with celecoxib had significantly less bone loss compared to controls ( $P < 0.0001$ ). There was a significant interaction between treatment with celecoxib and time ( $P < 0.03$ ). In both groups treated with celecoxib, the bone loss became significant only after 10 days of ligation placement, while in the control group it was already significant after 5 days. There was no significant difference in bone loss among experimental groups at the end of the experimental period. Infiltration of neutrophils, monocytes, and lymphocytes into the connective tissue of celecoxib-treated rats was significantly reduced compared with that of controls                                   |   |
| Holzhausen et al. [65] | Experimental ligature-induced periodontitis in rats | ABL       | Determination of serum etoricoxib concentrations and white blood cell count | Celecoxib              | Systemic            | 30 days   | Both etoricoxib-treated animal groups presented with significantly lower alveolar bone loss than that observed in the control group at all the studied treatment periods ( $P < 0.05$ ); however, the degree of alveolar bone loss was not statistically different between the groups receiving etoricoxib at either 6 or 12 mg/kg/day. |

|                       |  |   |   |          |   |  |
|-----------------------|--|---|---|----------|---|--|
| Howell et al.<br>[66] | Experimental periodontitis in beagle dogs  | ABL   | Naproxen                                  | Systemic | 7 months  | In dogs treated daily with naproxen, the rate of bone loss in the treatment period was significantly less at 4 months of treatment; however, at 7 months the difference, though lower than pretreatment rate, was not significant. When the percent change in rate of bone loss during the overall 7-month treatment period was compared with pretreatment rate, the control dogs demonstrated a 38% increase in rate of bone loss during the treatment period, contrasting with a 61% decrease in bone loss rate in naproxen-treated dogs. The data indicate that the nonsteroidal anti-inflammatory drug naproxen can significantly inhibit alveolar bone loss in beagles. At 4 months of treatment the rate of bone loss in the naproxen-treated dogs was significantly less than pretreatment, but at 7 months of treatment the rate was no longer statistically significantly less than baseline. This probably reflects a dose response to naproxen treatment for, after 30 days of the treatment period, the naproxen dosage was reduced tenfold due to tolerance by the beagle |
| Howell et al. [67]    | Experimental gingivitis in beagle dogs   | Plaque accumulation<br>Gingival inflammation<br>Bleeding upon gentle probing<br>Tooth staining  | Piroxicam<br>Topical                      | 16 weeks | By week 2, the gingival index in the piroxicam-treated dogs was significantly lower than that of the placebo-treated group and remained so throughout the study, with the exception of weeks 6 and 12 in the topical gel-treated group. Mean percent bleeding sites were also significantly less in the piroxicam-treated groups than in the control dogs. Staining of the teeth increased for all groups over the 16-week treatment period |  |
| Korman et al. [79]    | Preexisting gingivitis and experimental ligature-induced periodontitis in cynomolgus monkeys | Clinical parameters<br>ABL by densitometric analysis of radiographs (CADA)<br>Cultural microbiology of subgingival plaque<br>In situ PMN chemotaxis | Ibuprofen<br>Meclofenamic acid<br>Topical | 20 weeks | Radiographic bone loss was detected in all experimental sites in placebo animals as compared with 67% and 44% for ibuprofen and meclofenamic acid animals, respectively. Mean CADA scores/animal showed a significant loss in bone density for placebo at 6 and 16 weeks, no change for ibuprofen animals, and a significant increase in density for meclofenamic acid animals. All changes were in the absence of any effect on gingivitis |  |

(continued)

**Table 6.2** (continued)

| Reference           | Animal model   | Substrate   | NSAIDs       | Type of administration | Length of the study | Results   |
|---------------------|--|---|--------------|------------------------|---------------------|---|
| Li et al. [93]      | Experimental ligature-induced periodontitis in monkeys     | Plaque formation<br>Gingival redness<br>Edema<br>Bleeding on probing<br>Ramfjord Attachment Level measurements (RAL)<br>GCF PGE <sub>2</sub> , TxB <sub>2</sub> , LTB <sub>4</sub> , IL-1 $\beta$ , and TNF $\alpha$ levels<br>Alveolar bone loss | Ketoprofen   | Topical                | 6 months            | There were no significant differences among groups with respect to gingival indices. Radiographic data demonstrated significant positive effects on bone activity in both groups treated with ketoprofen formulations with improvement over time in the ligature model ( $0.01 \leq P \geq 0.04$ ). The placebo group exhibited bone loss of $1.96 \pm 0.48$ and $1.40 \pm 0.56$ mm per site at 3 and 6 months, respectively. The group treated with ketoprofen cream showed an apparent bone gain of $0.28 \pm 0.41$ and $0.78 \pm 0.47$ mm per site at 3 and 6 months, respectively. The group treated with ketoprofen cream containing vitamin E showed a mean bone loss of $0.41 \pm 0.48$ mm per site at 3 months with improvement to an apparent bone gain of $0.31 \pm 0.44$ mm per site at 6 months. The biochemical data demonstrated early and significant suppression of GCF-LTB <sub>4</sub> by both ketoprofen formulations at 1 month, which preceded the significant suppression of GCF-PGE <sub>2</sub> at 2 and 3 months in the ligature model ( $P < 0.003$ ) and at 2–6 months in the spontaneous model ( $P < 0.02$ ). In summary, ketoprofen at 1% level in suitable topical vehicles can effectively inhibit GCF-LTB <sub>4</sub> and GCF-PGE <sub>2</sub> and positively alter alveolar bone activity in the ligature-induced model of periodontitis in the monkey |
| Nassar et al. [103] | Experimental ligature-induced periodontitis in rats        | ABL   | Meloxicam    | Systemic               | 15 days             | Groups treated with meloxicam, after 5 days, had significantly less alveolar bone loss ( $P < 0.05$ ) when compared with control groups. On the other hand, no significant differences in bone loss were observed after 15 days of treatment with meloxicam   |
| Nyman et al. [107]  | Experimental ligature-induced periodontitis in beagle dogs | Connective tissue attachment loss<br>Alveolar bone loss   | Indomethacin | Systemic               | 28 days             | The placement of ligatures induced an acute inflammatory reaction in the periodontal tissues resulting in loss of connective tissue attachment and alveolar bone. Bone resorption could be observed histologically within 7 days, and radiographically within 2–3 weeks after ligature placement. Daily administration of indomethacin was shown to delay the onset and to suppress the magnitude of the acute inflammatory reaction, and to decrease the degree of alveolar bone resorption  |

|                          |  |  |                                       |                      |          |  |
|--------------------------|--|--|---------------------------------------|----------------------|----------|--|
| Offenbacher et al. [114] | Experimental ligature-induced periodontitis in the rhesus monkey <i>Macaca mulatta</i> | GCF PGE <sub>2</sub> , TxB <sub>2</sub> levels<br>CAL loss<br>ABL    | Flurbiprofen                          | Systemic             | 6 months | In untreated animals there was a statistically significant threefold increase in GCF-PGE <sub>2</sub> and TxB <sub>2</sub> levels at 3 months, as compared to baseline, which positively correlated with increases in redness, bleeding, CAL loss, and ABL. GCF-PGE <sub>2</sub> and TxB <sub>2</sub> levels reached a sixfold peak at 6 months and returned to baseline by 12 months. Flurbiprofen prevented the 3-month rise in TxB <sub>2</sub> , but did not affect the increase in PGE <sub>2</sub> . At 6 months, Flurbiprofen administration caused a dose-dependent inhibition of both PGE <sub>2</sub> and TxB <sub>2</sub> . Probit analysis of the dose-response data revealed that the concentration of Flurbiprofen which caused a 50% inhibition of GCF – TxB <sub>2</sub> level (the IC50 value for TxB <sub>2</sub> synthesis) was approximately two logs lower than the IC50 value for PGE <sub>2</sub> synthesis, i.e., TxB <sub>2</sub> – IC50 = 0.13 vs. PGE <sub>2</sub> – IC50 = 1.35 mg flurbiprofen/kg/day. The slopes of the PGE <sub>2</sub> and TxB <sub>2</sub> inhibition curves were identical, consistent with a similar mechanism, or singular enzyme for the site of action of Fb inhibition of CO activity |
| Offenbacher et al. [117] | Naturally progressing periodontitis in beagle dogs                                     | GCF PGE <sub>2</sub> , TxB <sub>2</sub> levels<br>Alveolar bone loss | Ibuprofen<br>Naproxen<br>Flurbiprofen | Systemic and topical | 6 months | Untreated animals demonstrated that at 1 month, GCF-PGE <sub>2</sub> levels increased twofold over baseline and, by 6 months, had reached a five- to sixfold elevation. GCF-TxB <sub>2</sub> levels rapidly reached a four- to fivefold peak over baseline at 1 month and subsequently dropped to a twofold elevation for the remainder of the study. The rate of ABL in untreated animals increased 38% during the 6-month period, as compared to baseline pretreatment ABL rates. Overall, there was a significant depression in the GCF levels of both PGE <sub>2</sub> and TxB <sub>2</sub> in all NSAID-treated groups. All NSAIDs treatments significantly retarded ABL, ranging from 21.0% to 36.9% of the control ABL rate   |
| Oliveira et al. [120]    | Experimental ligature-induced periodontitis in rats                                    | Vascular endothelial growth factor (VEGF) expression<br>ABL          | Meloxicam                             | Systemic             | 30 days  | A reduction in ABL was observed in the meloxicam-treated group compared to the control group at all periods studied. There was a positive correlation between COX-2 mRNA and VEGF mRNA in the gingival tissues and periodontal disease ( $r=0.80$ ; $P=0.026$ ). Meloxicam significantly reduced the increased mRNA VEGF expression in diseased tissues after 14 days of treatment ( $P=0.023$ ). After 14 days of treatment with meloxicam, an important decrease in VEGF protein expression was detected in diseased tissues ( $P=0.08$ ). Qualitative IHC analysis revealed that VEGF protein expression was higher in diseased tissues and decreased in tissues from rats treated with meloxicam   |

(continued)

**Table 6.2** (continued)

| Reference                   | Animal model   | Substrate   | NSAIDs  | Type of administration | Length of the study | Results  |
|-----------------------------|--|---|---|------------------------|---------------------|--|
| O'uchi et al. [121]         | Experimental ligature-induced periodontitis in beagle dogs | Radiographic and morphometric analyses  | YM175 [disodium dihydrogen (cyclohexylamino) methylene-1, 1-bisphosphonate]<br>Flurbiprofen                                       | Systemic               | 25 weeks            | In placebo-treated animals, the ligation caused a significant decrease in the alveolar bone height by 0.57 and 1.91 mm at 2 and 25 weeks, respectively. YM175 and flurbiprofen tended to prevent bone loss after 15 weeks. Although the ligation elicited no significant change in bone mineral density, it significantly decreased bone volume. YM175 (1.0 mg/kg) and flurbiprofen tended to increase the bone volume   |
| Paquette et al. [123]       | Experimental ligature-induced periodontitis in beagle dogs | Gingival inflammation assessed with a Gingival Index<br>Alveolar bone loss            | Ketoprofen  | Systemic and topical   | 2 months            | Significant differences in soft tissue responses were detected among the treatment cohorts ( $P < 0.05$ ) when groups were compared for changes in gingival indices. Beagles treated with (S)-ketoprofen capsules or placebo dentifrice exhibited similar increases in mean gingival indices; however, beagles treated with 0.3% or 3.0% (S)-ketoprofen dentifrice showed significantly smaller increases in mean scores over the 2 months as compared to placebo-treated animals ( $P < 0.05$ ). From days 1 to 60, cohorts differed significantly in terms of bone loss rates ( $P < 0.001$ ). In particular, beagles treated with systemic or topical ketoprofen exhibited significantly lower mean rates of bone loss compared to placebo treated beagles ( $P < 0.05$ ).  |
| Queiroz-Junior et al. [135] | Experimental ligature-induced periodontitis in rats        | Losses of fiber attachment (FAL)<br>ABL<br>Infiltration of cells into gingival tissue | Indomethacin<br>Celecoxib (selective COX-2 inhibitors)<br>SC236 (selective COX-2 inhibitors)<br>SC560 (selective COX-1 inhibitor) | Systemic and topical   | 11 days             | Celecoxib (3–30 mg/kg) administered systemically reduced FAL and ABL in a dose-dependent manner in diseased animals compared with control (diseased and vehicle-treated) animals. Animals treated systemically with the experimental compound SC236 (12 mg/kg/day) or with indomethacin (2 mg/kg/day) exhibited a significant ( $P < 0.05$ ) decrease in FAL and ABL as well as in leukocyte number present in gingival tissue when compared with control groups. ABL was, however, more sensitive to drug treatment than fiber attachment loss. In contrast, systemic (0.5 or 5 mg/kg/day) administration of SC560, a selective COX-1 inhibitor, was effective in reducing FAL but did not reduce ABL, whereas it was as effective as COX-2 inhibitors in decreasing cell migration into gingival tissue of animals with periodontal disease. Local injection of celecoxib at 60, 120, or 240 µg/site/day, SC236 at 120 µg/site/day, or indomethacin at 100 µg/site/day, preventively (from third to fifth day), significantly reduced FAL and ABL to an extent similar to systemic treatment |

|                            |  |   |                        |  |
|----------------------------|--|---|------------------------|--|
| Saffar & Lasfargues [145]  | Experimental periodontitis in hamsters                                 | Alveolar bone loss<br>Indomethacin<br>Systemic  | 12 weeks               | The systemic indomethacin and calcitonin reduced the extent of bone resorption considerably but not significantly (NS). The reversal phase, the intermediate step between resorption and formation, was decreased by 33% (NS) by indomethacin and 75% by calcitonin ( $P < 0.02$ ). Bone formation was increased by 270% with indomethacin ( $P < 0.05$ ) and by 400% with calcitonin ( $P < 0.03$ ), compared with untreated animals. This exceeded the extent of bone formation activity in control animals  |
| Vardar-Sengul et al. [176] | Experimental <i>Escherichia coli</i> LPS-induced periodontitis in rats | Alveolar bone loss<br>Gingival tissue levels of PGE <sub>2</sub> , PGF <sub>2α</sub> , LTB <sub>4</sub> , and PAF<br>Celecoxib<br>Prophylactic omega-3 fatty acid (P)<br>Therapeutic omega-3 fatty acid (TO3) | Oral gavage<br>14 days | The ABL in the omega-3 fatty acid, celecoxib, and combination groups was less than the loss in the LPS group, but the differences were not statistically significant ( $P > 0.05$ ). Individual administration of celecoxib revealed significant reductions in PGE <sub>2</sub> and PAF levels ( $P < 0.05$ ), while omega-3 fatty acid provided significant reduction in PGE <sub>2</sub> , PGF <sub>2α</sub> , and LTB4 levels compared to the LPS group ( $P < 0.05$ ). Combined administration of celecoxib and omega-3 fatty acid exhibited significantly lower values than those of the LPS group in all the analyzed membrane phospholipid mediators ( $P < 0.05$ ), which approximated the levels in the saline control group ( $P > 0.05$ ) |
| Vardar-Sengul et al. [178] | Experimental <i>Escherichia coli</i> LPS-induced periodontitis in rats | Alveolar bone loss<br>IL-1β Levels in Serum<br>Osteocalcin (OC) in serum<br>C-reactive protein (CRP) in serum<br>Celecoxib<br>Prophylactic omega-3 fatty acid (P)<br>Therapeutic omega-3 fatty acid (TO3)     | Oral gavage<br>14 days | The ABL in the omega-3 fatty acid, celecoxib, and combination groups was less than the loss in the LPS group, but the differences were not statistically significant ( $P > 0.05$ ). The median levels were 47.0 and 52.5 pg/mL, respectively, for the omega-3 fatty acid and combination groups, and these values were significantly higher than those of the LPS and celecoxib groups ( $P < 0.05$ ). Individual and combined administration of celecoxib and omega-3 fatty acid significantly increased OC levels compared to the LPS group ( $P < 0.05$ ). There were no significant differences in serum CRP levels   |

(continued)

**Table 6.2** (continued)

| Reference                  | Animal model   | Substrate  | NSAIDs   | Type of administration | Length of the study | Results  |
|----------------------------|--|--|--|------------------------|---------------------|--|
| Vardar-Sengul et al. [177] | Experimental <i>Escherichia coli</i> LPS-induced periodontitis in rats | Alveolar bone loss<br>Gingival tissue expression of MMP-8, -13, and -14, tissue inhibitor of MMP (TIMP)-1, and laminin (Ln)-5 $\gamma$ 2-chain | Celecoxib<br>Prophylactic omega-3 fatty acid (P)<br>Therapeutic omega-3 fatty acid (TO3) | Oral gavage            | 15 days             | Alveolar bone loss in treatment groups was less than in the LPS group, but the differences did not reach statistical significance ( $P > 0.05$ ). TO3 group exhibited lower MMP-8 expression than the LPS group, but the difference was not statistically significant ( $P > 0.05$ ), whereas in the celecoxib, P + TO3, and combined treatment groups, the expression of MMP-8 was very low and significantly lower ( $P < 0.001$ ) than in the LPS group. MMP-13 expression was similar in all studied groups ( $P > 0.05$ ). MMP-14 expression in the celecoxib and P + TO3 groups was significantly higher than in the saline control (both $P = 0.004$ ) and LPS (both $P = 0.001$ ) groups. LPS injection resulted in a significant decrease in TIMP-1 expression compared to the saline control group ( $P = 0.000$ ). The combined drug treatment, celecoxib, and P + TO3 groups exhibited lower TIMP-1 expression than the saline control group ( $P = 0.002$ , $P = 0.001$ , and $P = 0.001$ , respectively) and similar expression to the LPS group ( $P > 0.05$ ). No significant differences in Ln-5 $\gamma$ 2-chain expression ( $P > 0.05$ ) were found among the study groups |
| Vogel et al. [181]         | Experimental ligature-induced periodontitis in squirrel monkeys        | Alveolar bone loss   | Indomethacin<br>Substituted oxazolopyridine derivative (SOPD)                            | Systemic and topical   | 14 days             | The SOPD significantly inhibited gingival inflammation and loss of attachment as compared to either the placebo or indomethacin groups. Both NSAIDs inhibited bone loss  |
| Weak-Dybvig et al. [185]   | Experimental ligature-induced periodontitis in squirrel monkeys        | Alveolar bone loss<br>The number of osteoclasts per 1 mm of bone contour (osteoclast density)  | Indomethacin   | Systemic               | 14 days             | In animals administered indomethacin prior to and during experimental periodontitis, the gingival inflammatory cell infiltrate appeared less extensive, exhibited no significant loss in alveolar crestal bone height for the duration of the experiment had a significantly lower Percent active formation surface Periodontal ligament surfaces on endosteal surfaces. Administration of indomethacin resulted also in a significantly lower osteoclast density  |

|                       |   |  |                           |          |           |   |
|-----------------------|---|--|---------------------------|----------|-----------|---|
| Williams et al. [194] | Naturally occurring periodontal disease in beagle dogs                    | Alveolar bone loss on standardized radiographs | Flurbiprofen              | Systemic | 12 months | There was a 16% increase in the observed bone loss over the expected bone loss about those teeth which received neither systemic nor local treatment, and a 24% increase of observed bone loss over expected bone loss in those teeth treated with conventional periodontal treatment and no systemic treatment. In marked contrast, in those beagle dogs the with 0.02 mg/kg flurbiprofen alone eases there was a 66% decrease in observed bone loss about those teeth which received no local treatment and a 91% decrease when flurbiprofen administration was combined bined with local surgical treatment  |
| Williams et al. [190] | Naturally occurring periodontal disease in beagle dogs                    | Alveolar bone loss on standardized radiographs | Indomethacin Flurbiprofen | Systemic | 12 months | In the untreated control dogs, the rate of bone loss in the treatment period significantly increased from baseline ( $1.00 \pm 0.08\%$ per month from $0.53 \pm 0.07\%$ per month, $P < 0.001$ ). In contrast, the rate of bone loss significantly decreased from baseline in the flurbiprofen-treated dogs ( $0.17 \pm 0.07\%$ per month from $0.42 \pm 0.08\%$ per month, $P < 0.01$ ). In indomethacin-treated dogs, the treatment period rate of bone loss was less than the pretreatment baseline rate but the decrease was not statistically significant. However, the significant increase in rate in the treatment period that the control dogs demonstrated did not occur                        |
| Williams et al. [191] | Naturally occurring moderate-to-severe periodontal disease in beagle dogs | Alveolar bone loss on standardized radiographs | Ibuprofen                 | Systemic | 13 months | In the untreated control dogs, the rate of bone loss in the treatment period was higher than baseline but not statistically significantly higher. In contrast, the rate of bone loss in the dogs treated with 4.0 mg/kg ibuprofen and 4.0 mg/kg sustained-release ibuprofen was significantly decreased from baseline for the overall 7-month treatment period ( $P < 0.02$ ). The control dogs demonstrated a 38% increase in rate over baseline in the treatment period. In contrast, the sustained release ibuprofen-treated dogs had a striking 70% decrease in rate of bone loss while the 4 mg/kg ibuprofen and 0.4 mg/kg ibuprofen had a 50% decrease in rate of bone loss in the treatment period |

(continued)

**Table 6.2** (continued)

| Reference                | Animal model  | Substrate  | NSAIDs  | Type of administration | Length of the study | Results   |
|--------------------------|---|--|---|------------------------|---------------------|---|
| Williams et al. [192]    | Naturally occurring moderate-to-severe periodontal disease in beagle dogs | Alveolar bone loss on standardized radiographs   | Flurbiprofen  | Topical                | 13 months           | In the untreated control dogs, the rate of bone loss in the treatment period did not change significantly from baseline, although the rate was elevated by 38%. In contrast, the rate of bone loss significantly decreased by 71% from baseline in the flurbiprofen-treated dogs. The untreated control dogs lost ten teeth during the treatment period whereas the topical flurbiprofen-treated dogs lost only one tooth   |
| Williams et al. [195]    | Naturally occurring moderate-to-severe periodontal disease in beagle dogs | Alveolar bone loss<br>GCF levels of PGE <sub>2</sub> , PGF <sub>2α</sub> , TXB <sub>2</sub> , 6K-PGF <sub>1α</sub> | Flurbiprofen<br>Indomethacin                            | Systemic               | 12 months           | In the untreated control dogs, the mean rate of bone loss significantly ( $P < 0.01$ ) increased in the treatment period from the baseline pretreatment rate ( $0.53 \pm 0.07\%$ to $1.00 \pm 0.08\%$ per month). In striking contrast, the rate of bone loss in the flurbiprofen-treated dogs was significantly ( $P < 0.01$ ) decreased to $0.17 \pm 0.07\%$ per month from the pretreatment rate of $0.42 \pm 0.08\%$ per month. In the indomethacin-treated dogs the rate of bone loss did not differ significantly in the treatment period ( $0.32 \pm 0.05\%$ per month) from the baseline rate of bone loss ( $0.42 \pm 0.08\%$ per month). Whereas the effect of indomethacin and flurbiprofen on slowing the rate of alveolar bone loss was not similar, the administration of these two NSAIDs significantly reduced in a similar manner the GCF levels of PGE <sub>2</sub> , PGF <sub>2α</sub> , and TXB <sub>2</sub> ; 6K-PGF <sub>1α</sub> levels were not altered |
| Lasfargues & Saffar [87] | Experimental periodontitis in hamsters                                    | Alveolar bone loss<br>The number of osteoclasts per mm of bone surface   | Indomethacin<br>Calcitonin<br>Indomethacin + Calcitonin | Systemic               | 12 weeks            | Indomethacin decreased the macroscopic bone loss by 25% and the number of osteoclasts per mm by 55% in diseased animals but not significantly. Conversely calcitonin reduced significantly bone loss ( $P < 0.05$ ) and decreased the number of osteoclasts to the control level ( $P < 0.01$ ). The combination of the two drugs had the same effect than calcitonin alone on the two parameters. These results show an effect of indomethacin on bone destruction, but an inhibitory action on prostaglandin synthesis can only be hypothesized. In this model of periodontitis, calcitonin appears more effective than indomethacin  |

|                          |  |   |                              |          |          |   |
|--------------------------|--|---|------------------------------|----------|----------|---|
| Jeffcoat et al. [71]     | Naturally occurring periodontal disease in beagle dogs                                 | Alveolar bone loss<br>Bone-seeking radiopharmaceutical uptake<br>Gingival inflammation  | Flurbiprofen                 | Systemic | 6 months | Follow-up of [187] study after withdrawal of the drug. In flurbiprofen-treated dogs, the rate of bone loss was significantly decreased about both surgically and nonsurgically treated teeth throughout the 12-month treatment period. This decreased rate was sustained through 3 months of the posttreatment period, but was lost 6 months following the termination of flurbiprofen therapy. No similar effect on reducing the rate of bone loss was observed in the placebo-treated dogs. A significant decrease in bone-seeking radiopharmaceutical uptake was observed in the flurbiprofen-treated teeth, which corresponded with the radiographic findings |
| Offenbacher et al. [110] | Experimental ligature-induced periodontitis in the rhesus monkey <i>Macaca mulatta</i> | Clinical measurements included standardized radiographs, Ramfjord attachment level determinations, and assessments of redness, edema, and bleeding on probing | Flurbiprofen                 | Systemic | 6 months | There was a statistically significant inhibition of attachment loss ( $P < 0.05$ ), gingival redness ( $P < 0.05$ ), and bleeding on probing ( $P < 0.05$ ) in ligature-induced spontaneous periodontitis in the flurbiprofen-treated animals at 6 months. The odds of a control ligated monkey undergoing significant attachment loss in 6 months are elevated 29.3-fold, as compared to the flurbiprofen-treated cohort monkey group. Flurbiprofen treatment also significantly inhibited spontaneous attachment loss for 6 months as compared to control monkeys, at $P < 0.05$  |
| Nuki et al. [106]        | Experimental ligature-induced periodontitis in beagle dogs                             | Plaque Index<br>Gingival Index<br>Bacterial culture,<br>Bone-resorbing activity in gingival extracts  | Indometacin<br>Metronidazole | Systemic | 21 days  | No effect of indomethacin administration on ligature-induced bone-resorbing activity of gingival extracts.<br>Neither drug appeared to affect the Plaque and Gingival Index scores of the ligated teeth during the experimental period  |

NSAID nonsteroidal anti-inflammatory drugs; GCF gingival crevicular fluid; PMN polymorphonuclear granulocytes; BOP bleeding on probing;  $PGE_2$  prostaglandin E<sub>2</sub>;  $Tx B_2$  thromboxane B<sub>2</sub>;  $LTB_4$  IL-1 $\beta$ ; TNF $\alpha$ ; Tumor Necrosis Factor  $\alpha$ ; ABL Alveolar bone loss;  $PGF_{2\alpha}$ ; prostaglandin F<sub>2 $\alpha$</sub> ;  $LTB_4$  leukotriene B<sub>4</sub>; PAF platelet activating factor

experimental gingivitis/periodontitis models in rats [7, 14, 25, 52, 64, 103, 120, 135, 176–178], hamsters [145], dogs [66, 67, 71, 106, 107, 117, 121, 123, 191, 192, 194, 195], or monkeys [79, 93, 110, 114, 181].

#### 6.4.2 Human Studies

Adjunctive periodontal clinical trials conducted with a variety of different systemically or locally administered NSAIDs, including flurbiprofen, meclofenamate, ibuprofen, ketorolac, naproxen, and aspirin are summarized in Table 6.3. Several systematic reviews in the field concluded that limited quantitative analysis tended to show a significant benefit related to alveolar bone preservation when NSAIDs were associated with conventional therapy [77, 105, 124, 138, 146, 168, 169]. Otherwise, superior results were not consistently observed when clinical attachment level was used as the outcome measure [77, 138].

The effects of several NSAIDs on pain relief after nonsurgical periodontal treatment and periodontal surgery were evaluated: celecoxib [128], diclofenac [164, 186], diflunisal [89, 98, 99], etodolac [167], flurbiprofen [47], ibuprofen [13, 39, 108, 126, 137, 180], ketoprofen [140], ketorolac [165], and sodium meclofenamate [48]. Therapeutic doses of these NSAIDs, given presurgically or administered immediately following scaling and root planning or periodontal surgery are significantly more effective than placebo in limiting postoperative pain associated with periodontal interventions. However, [157] revealed that ibuprofen taken prior to periodontal surgery increases intraoperative bleeding and should be administered cautiously before periodontal surgeries.

### 6.5 Side Effects of NSAIDs

The use of NSAIDs is associated with many side effects, but their unwanted effects on the gastrointestinal tract, the kidney, and the cardiovascular system are considered as major issues with the use of these important drugs. The emergence of NSAID's side effects depends not only on the patient's condition but also on

the dosage regime that, by itself, is based on the underlying disease. The side effects of NSAIDs are intuitively expected to be less frequent after short-term analgesic doses as compared to when long-term repeated regimens are used [55].

**The gastrointestinal side effects** of NSAIDs have been vastly discussed in the literature since their introduction to the market [55, 82, 84, 85, 184]. Gastric or duodenal ulcers are found at endoscopy in approximately 15–30% of patients taking NSAIDs regularly, while annual incidence of serious upper GI complications (major bleeding, perforation, obstruction) of approximately 1–1.5% and of clinical upper GI events (complications plus symptomatic ulcers discovered on evaluation of GI symptoms or signs) of approximately 2.5–4.5% were reported [84]. American College of Rheumatology ad hoc group on use of selective and nonselective nonsteroidal anti-inflammatory drugs [4] revealed that exposure to nonselective NSAIDs has been associated with a 2.7- to 5.4-fold increased risk of various GI adverse events (Table 6.4). Current NSAID users have a 4.3-fold greater risk of upper GI bleeding compared with controls taking placebo (95%, CI: 3.7–5.0). The case fatality rate for upper GI bleeds is 5%. Among patients with rheumatoid arthritis receiving NSAID therapy, there is a 1.58% hospitalization incidence and a 0.19% per year risk of GI-related death [4].

All patients taking traditional NSAIDs have an increased risk of developing GI complications. A number of clinical characteristics have been associated with an increased risk of complications. The most important include a history of ulcer or GI complications, *Helicobacter pylori* infection prior to NSAID therapy, increasing age, concomitant anticoagulation, concomitant corticosteroid use, and high-dose or multiple NSAIDs (including an NSAID plus low-dose aspirin). Concurrent illness (e.g., severe rheumatoid arthritis, heart disease) has also been reported to increase the risk of GI events [74, 84].

To prevent NSAID-induced gastropathy, three strategies are followed in clinical routine: (i) coprescription of a gastroprotective drug, (ii) use of selective COX-2 inhibitors, and (iii) eradication of *Helicobacter pylori* [11, 162].

After gastrointestinal toxicities, mild and serious **renal complications** are the second biggest concern associated with NSAID use. The renal effects of

**Table 6.3** Clinical studies of adjunctive NSAIDs therapy in periodontal disease

| Study               | No. of patients | Periodontal condition    | Study period | Periodontal treatment  | Outcome   |
|---------------------|-----------------|--------------------------|--------------|--|---|
| Aras et al. [6]     | 34              | Chronic periodontitis    | 6 weeks      | 1. SRP + naproxen sodium (275 mg, 1x/day, 6 weeks<br>2. SRP + placebo  | Except for the GBI, there was a significantly larger change of all clinical parameters in the NSAID group compared to the placebo group after therapy ( $P < 0.05$ ) (GI: Tests from $2.48 \pm 0.14$ to $0.86 \pm 0.11$ , Controls from $2.57 \pm 0.21$ to $1.66 \pm 0.13$ ; PI: Tests from $2.29 \pm 0.19$ to $0.49 \pm 0.09$ , Controls from $2.03 \pm 0.20$ to $1.06 \pm 0.17$ ; PD: Tests from $3.59 \pm 0.20$ to $2.33 \pm 0.13$ , Controls from $3.82 \pm 0.22$ to $2.82 \pm 0.13$ ). When the mean total ELA activity and elastase-like enzyme activity (ELA) concentrations in GCF were analyzed, there was a significant decrease in the NSAID group at baseline and at 6 weeks ( $P < 0.05$ ). However, there was no significant change in the placebo group when ELA levels were compared at the same time period ( $P > 0.05$ ) |
| Azoubel et al. [7]  | 20              | Aggressive periodontitis | 1 month      | 1. SRP + etoricoxib (120 mg/day, 7 days)<br>2. SRP + placebo   | At the end of the experimental period, no statistical differences were observed between the groups in any clinical parameter evaluated. However, all variables presented significant alterations within each group between the beginning and the end of the study, with a decrease in all values, with the exception of $R$ , which increased. Mean CAL decreased from $5.54 \pm 0.47$ mm to $3.59 \pm 0.53$ mm in the test group and from $5.92 \pm 1.10$ to $3.69 \pm 0.80$ mm in the control group ( $P = 0.47$ ). Assessment of the variations in PGE2 levels in GCF revealed a significant difference between the groups after 7 days ( $P = 0.0351$ ); a greater reduction was found in the test group. After 30 days, the groups presented similar results ( $P = 0.40$ )  |
| Bichara et al. [15] | 24              | Intra-bony defects       | 9 months     | 1. GPN group: GTR therapy with barrier membrane and adjunctive Systemic naproxen 500 mg, 2x/day, 7 days<br>2. GA group: GTR therapy with barrier membrane alone                                      | Open defect measurements from baseline to 9 months showed a statistically significant ( $P < 0.05$ ) mean defect fill of $1.96 \pm 1.27$ mm and $2.04 \pm 1.71$ for the GPN and GA groups, respectively. This corresponds to a mean defect fill of 42% and a mean defect resolution of approximately 75% for both groups. The differences between GPN and GA groups were not statistically significant ( $P > 0.05$ ). Defect fill of $\geq 50\%$ was seen in six defects (50%) in the GPN group and in five defects (42%) in the GA group. In summary, the administration of postsurgical naproxen failed to produce osseous healing that was statistically superior to that obtained with polylactide bioabsorbable membranes alone   |
| Brägger et al. [22] | 19              | Chronic periodontitis    | 6 months     | 1. Surgical periodontal therapy (modified Widman flap) + systemic flurbiprofen 50 mg 3x/day for 30 days ( $N = 10$ )<br>2. Surgical periodontal therapy (modified Widman flap) + placebo ( $N = 5$ ) | Standardized radiographs showed minimal remodeling activity after periodontal surgery in both patient groups. A significant reduction of the probing depth and a significant amount of clinical attachment gain was noted at the surgically treated sites irrespective of whether the patients had used flurbiprofen or placebo   |

(continued)

**Table 6.3** (continued)

| Study                  | No. of patients                  | Periodontal condition                                 | Study period | Periodontal treatment   | Outcome  |
|------------------------|----------------------------------|---|--------------|---|--|
| Buduneli et al. [24]   | 12                               | Chronic periodontitis                                 | 10 days      | 1. SRP+systemic meloxicam (7.5 mg, 10 days)<br>2. SRP+placebo   | Both meloxicam and placebo groups showed statistically significant reductions in PBI, PI, and GCF MMP-8 levels on day 10 compared to baseline ( $P < 0.01$ ). No significant differences between groups regarding PBI and PI at day 10 ( $P > 0.05$ ). The GCF MMP-8 concentration on day 10 in the meloxicam group was significantly lower than in the placebo group (30.65 vs. 112.23 ng/ $\mu$ l; $P = 0.016$ )   |
| Cavanaugh et al. [27]  | 55                               | Adult periodontitis                                   | 6 months     | 1. Oral prophylaxis provided every 3 months + ketorolac rinse (0.1%) 2×/day with placebo capsule<br>2. Oral prophylaxis provided every 3 months + 50 mg flurbiprofen capsule 2×/day (positive control) with placebo rinse<br>3. Oral prophylaxis provided every 3 months + 2×/day placebo rinse and capsule ketorolac tromethamine oral rinse | A statistically significant correlation ( $r = 0.73$ , $P = 0.001$ ) exists between GCF PGE <sub>2</sub> concentration and the maximum amount of bone height lost at individual patient study sites. The correlation between GCF IL-1 $\beta$ concentration and maximum bone height lost is also statistically significant ( $r = 0.66$ , $P = 0.005$ ). Over the 6-month duration of the study, both PGE <sub>2</sub> and IL-1 $\beta$ were coordinately expressed in the placebo treatment group as reflected in the significant correlation between GCF concentrations of the 2 mediators ( $r = 0.81$ , $P < 0.001$ ). Treatment of patients with 0.1% ketorolac tromethamine twice daily for 6 months resulted in reductions of PGE <sub>2</sub> in GCF and a negligible correlation between GCF PGE <sub>2</sub> and GCF IL-1 $\beta$ ( $r = 0.42$ , $P = 0.088$ ). This lack of a strong association between the two mediators in the ketorolac treatment group provides a direct biochemical readout of the anti-inflammatory efficacy of ketorolac tromethamine oral rinse in patients with periodontitis |
| Dastoor et al. [33]    | 30                               | Moderate-to-advanced chronic periodontitis in smokers | 6 months     | 1. SURG (Apically positioned flap with osseous recontouring) + CHX + Ibuprofen (600 mg every 6 h for 2 days, and thereafter as needed) + AZ (500, 1×, 3/day) ( $N = 15$ )<br>2. SURG + CHX + ibuprofen + placebo ( $N = 15$ )   | Surgical treatment of moderate (PD=4 to 6 mm) and deep (PD>6 mm) pockets significantly improved clinical parameters of treated and untreated teeth (CAL gain, PD reduction, and reduction of BOP). The additional use of AZM did not enhance this improvement nor did it promote reduction of cross-linked telopeptide of type I collagen levels in GCF. Compared to the control group, the test group had significantly better WHI scores at 1 month, significantly less GI at 2 weeks, and sustained reductions of red-complex bacteria with trypsin-like enzyme activity at 3 months. For nonsurgery teeth, only the test group showed significant gains in overall CAL compared to baseline. In summary, the findings of this pilot study demonstrated that in heavy smokers, adjunctive systemic AZM in combination with pocket reduction surgery did not significantly enhance PD reduction or CAL gain. However, the clinical value of adjunctive AZM may be appreciated by more rapid wound healing, less short-term gingival inflammation, and sustained reductions of peropathogenic bacteria            |
| Drougani & Hirsch [36] | 392 males aged 50 years and over | Chronic periodontitis                                 | 2 years      | 1. Aspirin nonsmokers (300 mg or less per day) ( $N = 51$ )<br>2. Aspirin ex-smokers (300 mg or less per day) ( $N = 102$ )<br>3. No aspirin nonsmokers ( $N = 122$ )<br>4. No aspirin ex-smokers ( $N = 117$ )   | Controlling for age, mean CAL in aspirin takers was significantly less $2.6 \pm 0.08$ mm than non-aspirin-takers $2.9 \pm 0.06$ (se) mm; this association was independent of smoking history. Ex-smokers had significantly more mean CAL $3.9 \pm 0.07$ (se) mm than non-smokers $2.6 \pm 0.08$ (se) mm, irrespective of aspirin status. When most severe score of PAL (MSS-CAL) for each tooth, averaged per subject was analysed, these differences became more pronounced; MSS-CAL in aspirin takers was significantly less $3.9 \pm 0.1$ mm than non-aspirin-takers $4.2 \pm 0.08$ mm. Ex-smokers had significantly more MSS-CAL $4.3 \pm 0.08$ mm than non-smokers $3.8 \pm 0.08$ mm. Aspirin apparently had a protective association on CAL and it is hypothesised that low-dose aspirin may have reduced the rate of attachment loss  |

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| Flemmig et al. [43]  | 60 | Moderate-to-severe periodontitis       | 6 months  | 1. 1x daily adjunctive supragingival irrigation with 300 ml water immediately followed by 200 ml of buffered 0.3% aspirin<br>2. 1x daily adjunctive supragingival irrigation with 500 ml water<br>3. Normal oral hygiene alone                                   | Results at 6 months showed that both supragingival irrigation with buffered 0.3% aspirin and supragingival irrigation with water significantly reduced Gingival Index scores (median 0.1 and 0.35, respectively) and PDs (both median 0.26 mm) compared to the control group. Irrigation with buffered 0.3% aspirin had no significant effect on bleeding on probing compared to the control group  |
| Flemmig et al. [41]  | 30 | Moderate-to-severe adult periodontitis | 12 weeks  | 1. Scaling+systemic aspirin (500 mg, 4x, 6 weeks)<br>2. Scaling+placebo<br>3. Aspirin (500 mg, 4x, 6 weeks)<br>4. Placebo alone  | Adjunctive aspirin administration resulted in synergistic reductions of gingival inflammation and PD as well as CAL gain compared with placebo treatment over the 6-week observation period (interaction: $P>0.05$ ). Only the effect of aspirin was significant in reducing the concentration of elastase-alpha 1-proteinasase inhibitor in GCF GCF E-alpha 1-PI ( $P<0.001$ ), reduction in GCF E-alpha 1-PI concentrations by aspirin may indicate a decreased risk in periodontal disease progression   |
| Funosas et al. [46]  | 33 | Chronic periodontitis                  |           | 1. SRP+placebo<br>2. SRP+1% acetyl-salicylic acid sub-gingival gel applied 48 h after each SRP session<br>3. SRP+1% ketoprofen sub-gingival gel applied 48 h after each SRP session<br>4. SRP+2% ketoprofen sub-gingival gel applied 48 h after each SRP session | All protocols (groups 1, 2, 3, and 4) induced a reduction of PD, plaque, and gingival indices and bleeding on probing. The 1% ASA and 2% KTP protocols (groups 2 and 4) significantly reduced the probing depth variable (ANOVA $P<0.05$ )  |
| Haffajee et al. [53] | 98 | Severe periodontal disease             | 10 months | 1. SRP+SURG+CHX+TET (250, 4x, 30day) then SPT<br>2. 1. SRP+SURG+ CHX + AMOX/CLAV (250, 3x, 30day) then SPT<br>3. 1. SRP+SURG+ CHX + IBU (400 mg 3x/day) then SPT<br>4. 1. SRP+SURG+ CHX + Placebo then SPT   | Subjects receiving antibiotics exhibited significantly more attachment level "gain" ( $0.57 \pm 0.15$ mm) than subjects receiving either ibuprofen or a placebo ( $0.02 \pm 0.10$ mm). The differences between AMO/CLAV and TET groups were not significant, nor were the differences between ibuprofen and placebo. Subjects receiving systemically administered antibiotics had a greater decrease in the number of sites colonized by <i>P. gingivalis</i> , <i>B. forsythus</i> , <i>P. intermedia</i> , and <i>P. micros</i> post-therapy than subjects not receiving antibiotics. In summary, the results of this investigation indicate that adjunctive systemic antibiotics increase periodontal attachment "gain" and decrease the levels of some suspected periodontal pathogens in subjects with evidence of current disease progression |

(continued)

**Table 6.3** (continued)

| Study                | No. of patients | Periodontal condition             | Study period | Periodontal treatment  | Outcome   |
|----------------------|-----------------|-----------------------------------|--------------|--|---|
| Heasman et al. [56]  | 49              | Chronic periodontitis             | 12 months    | 1. SRP + 1% flurbiprofen toothpaste to use<br>2× daily, 12 months<br>2. SRP + Placebo dentifrice   | Both the flurbiprofen and placebo showed significant improvements in the clinical parameters over 12 months and there were no significant differences between the groups. Flurbiprofen-treated patients, however, demonstrated a significantly greater proportion of sites (8.0%) with bone gain when compared to the placebo group (3.3%). There were no significant differences between the groups in the number of sites showing bone loss or no change. In summer, 1% flurbiprofen toothpaste exerts a small, yet significant effect on bone metabolism in the absence of any apparent effects on clinical parameters |
| Heasman et al. [57]  | 21              | Experimental gingivitis           | 28 days      | 1. Systemic flurbiprofen (50 mg b.d. 7 days)<br>2. Placebo   | Results show that flurbiprofen significantly inhibited the development of redness and bleeding ( $P < 0.001$ ) effects that were associated with a significant inhibition of $\text{TxB}_2$ ( $P < 0.05$ ). There were no apparent flurbiprofen effects on $\text{GCF-PGE}_2$ or $\text{GCF-LTB}_4$ during the first 21-day gingivitis model  |
| Heasman et al. [59]  | 47              | Experimental gingivitis           | 27 days      | 1. Systemic flurbiprofen (100 mg, 7 days)<br>2. Placebo  | There were no significant differences at $p = 0.05$ between the groups for plaque indices or gingival crevicular fluid flow. The flurbiprofen group, however, demonstrated greater resolution of gingival inflammation by day 27 when compared to the placebo controls ( $P = 0.04$ ). In summary, these serum concentrations of flurbiprofen are sufficient to produce significant anti-inflammatory effects in the gingival tissues   |
| Heasman et al. [58]  | 24              | Experimental gingivitis           | 17 days      | 100 ml of 10 mm flurbiprofen solution in buffered preservative to one upper quadrant of the mouth. The contralateral quadrant received preservative only | Experimental gingivitis developed in all patients and there were no significant differences between the treatments for Gingival Index or PDs  |
| Heasman et al. [60]  | 25              | Experimental gingivitis           | 27 days      | 1. Oral flurbiprofen (100 mg/day, 6 days) + toothbrushing<br>2. Placebo + toothbrushing<br>3. Oral flurbiprofen (100 mg/day, 6 days) only                | There were no changes in PD throughout the study. A reduction of Gingival Index occurred between days 22 and 27 for all treatment groups. Crevicular fluid flow was also reduced between days 22 and 27 in groups 1 and 2. The differences between the three treatments were very small. It is concluded that systemic flurbiprofen (100 mg/day) can reduce the signs of an experimental gingivitis over 6 days. This effect may be seen when the drug is used alone and as an adjunct to toothbrushing   |
| Jeffcoat et al. [68] | 15              | Rapidly progressive periodontitis | 3 months     | 1. SRP + naproxen (500 mg, 2×/day, 3 months)<br>2. SRP + placebo   | There was significantly less bone loss as determined by analysis of bone height during the 3-month study in the naproxen-treated patients when compared to the placebo-treated patients ( $P < 0.001$ ). Radiopharmaceutical uptake was significantly reduced in the alveolar bone in patients receiving naproxen ( $P < 0.03$ ), whereas no significant change was observed in the placebo-treated patients. Furthermore, the subtraction radiographs showed a significant increase in the proportion of teeth demonstrating bone gain in the naproxen-treated group   |

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| Jeffcoat et al. [69] | 55  | Chronic periodontitis   | 6 months | 1. Oral prophylaxis provided every 3 months + ketorolac rinse (0.1%) 2x/day with placebo capsule<br>2. Oral prophylaxis provided every 3 months + 50 mg flurbiprofen capsule 2x/day (positive control) with placebo rinse<br>3. Oral prophylaxis provided every 3 months + 2x/day placebo rinse and capsule | A significant loss in bone height was observed during the study period in the placebo group ( $-0.63 \pm 0.11$ ; $P < 0.001$ ), but not in the flurbiprofen ( $-0.10 \pm 0.12$ ; $P = 0.40$ ) or ketorolac rinse ( $+0.20 \pm 0.11$ mm; $P = 0.07$ ) groups. Nested ANOVA revealed that ketorolac and flurbiprofen groups had less bone loss ( $P < 0.01$ ) and reduced gingival crevicular fluid $\text{PGF}_2$ levels ( $P < 0.03$ ) compared to placebo. ANOVA suggests ( $P = 0.06$ ) that ketorolac rinse preserved more alveolar bone than systemic flurbiprofen at the dose regimens utilized   |
| Johnson et al. [72]  | 102 | Established gingivitis  | 30 days  | 1. Systemic naproxen (500 mg 2x/day. 30 days)<br>2. Placebo   | When the final index measurements were compared to baseline, the drug had no significant effect on plaque index scores or gingival inflammation. Statistically, naproxen enhanced the resolution of gingival inflammation following removal of microbial plaque. Thus, although this drug does not suppress the inflammation-inducing properties of plaque, naproxen may enhance recovery following plaque removal   |
| Jones et al. [73]    | 10  | Experimental gingivitis | 7 days   | 1. Topical hydroxyethylcellulose (HEC; 3, 5, 10%)<br>2. Topical poly(vinylpyrrolidone) (PVP; 3, 5% polycarbophil (PC; 1, 3, 5%)<br>3. Topical flurbiprofen (5%)   | As determined by the gingival index, the level of gingival inflammation increased in two of the four volunteers treated with the control formulation, but reduced in four out of five subjects treated with the test formulation containing flurbiprofen. Mean gingival crevicular fluid levels were also reduced in four out of five subjects treated with the flurbiprofen-containing formulation, but also in three out of four control subjects. However, the mean gingival crevicular fluid volume of subjects treated with flurbiprofen-treated formulations was significantly lower when compared to that of subjects in the control group. Finally, the plaque indices did not significantly change between week two and three of the study period in either patient group |

(continued)

**Table 6.3** (continued)

| Study              | No. of patients | Periodontal condition          | Study period | Periodontal treatment  | Outcome  |
|--------------------|-----------------|--------------------------------|--------------|--|--|
| Kim et al. [76]    | 47              | Naturally occurring gingivitis | 7 days       | 1. Aspirin, 81 mg 1x/day<br>(N=16)<br>2. Aspirin, 325 mg 1x/day (N=16)<br>3. Placebo (N=15)  | Subjects who took aspirin, 325 mg, had a 10.93-fold ( $P<0.001$ ) higher BOP difference at 1 week than those who took placebo and a 9.31-fold ( $P=0.001$ ) higher BOP between pre- and postvalues than those who took aspirin, 81 mg cross-tabulation between treatment and BOP showed an increase of 50%, 68.75%, and 92.86% in BOP for placebo, aspirin, 81 mg, and aspirin, 325 mg, respectively. Subjects who took aspirin, 325 mg, had a 0.094 lower mean PD difference between pre- and postvalues compared to subjects who took placebo ( $P=0.06$ ). A crossstabulation between treatment and PD showed that 57.14%, 87.50%, and 100% of subjects who took placebo, aspirin, 81 mg, and aspirin, 325 mg, respectively, had a decrease in PD. No statistically significant differences were found in PI between pre- and postvalues for any of the three treatments. Although there were no significant correlations between PGE <sub>2</sub> and 15-epi-LXA <sub>4</sub> or LTB <sub>4</sub> , the subjects treated with aspirin, 325 mg, showed a greater negative association between PGE <sub>2</sub> and 15-epi-LXA <sub>4</sub> ( $r=-0.408$ ) than between PGE <sub>2</sub> and LTB <sub>4</sub> ( $r=-0.153$ ) or aspirin, 81 mg ( $r=-0.142$ ). These results indicated that the group of patients who demonstrated a higher response to aspirin-mediated PGE <sub>2</sub> suppression expressed more 15-epi-LXA <sub>4</sub> in GCF than the group with the lower response. The other eicosanoid, LTB <sub>4</sub> , had a weaker trend toward the level of PGE <sub>2</sub> expression in GCF |
| Kurtis et al. [80] | 42              | Chronic periodontitis          | 10 days      | 1. Nonsmokers: SRP+ daily 100 mg flurbiprofen tablets in a 2 × 1 regimen for 10 days (N=10)<br>2. Nonsmokers: SRP+ placebo tablets in a 2 × 1 regimen for 10 days (N=11)<br>3. Smokers: SRP+ daily 100 mg flurbiprofen tablets in a 2 × 1 regimen for 10 days (N=10)<br>4. Smokers: SRP+ Placebo tablets in a 2 × 1 regimen for 10 days (N=11) | PI and GI scores decreased after therapy for all groups ( $P < 0.05$ ) compared to baseline values. No statistically significant differences were found between baseline and post-therapy results in PD and CAL scores for groups 1 to 4 ( $P > 0.05$ ). Although a statistically significant reduction was observed in GCF volume after the therapy compared to baseline values for groups 3 and 4, no statistical differences were found between baseline and post-therapy results for groups 1 and 2. No statistically significant differences were found between baseline and post-therapy values in GCF levels of PGF2 for groups 1 and 2 ( $P > 0.05$ ). However, statistically significant differences were observed for groups 3 and 4 with regard to baseline and post-therapy GCF levels of PGE <sub>2</sub> ( $P < 0.05$ ). A statistically significant reduction was observed in GCF levels of TBARS after therapy compared to baseline values in group 3 ( $P=0.01$ ). No statistically significant differences were found between baseline and post-therapy values in terms of GCF levels of TBARS in groups 1, 2, and 4 ( $P > 0.05$ ). A statistically significant difference was observed between groups 1 and 3 in terms of post-therapy values of GCF TBARS levels in favor of group 3 ( $P=0.001$ )  |

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| Kurtis et al. [81]   | 58 | Chronic periodontitis | 10 days | 1. Nonsmokers: SRP+ daily 100 mg flurbiprofen tablets in a 2 × 1 regimen for 10 days ( $N=14$ )<br>2. Nonsmokers: SRP+placebo tablets in a 2 × 1 regimen for 10 days ( $N=15$ )<br>3. Smokers: SRP+daily 100 mg flurbiprofen tablets in a 2 × 1 regimen for 10 days ( $N=14$ )<br>4. Smokers: SRP + placebo tablets in a 2 × 1 regimen for 10 days ( $N=15$ ) | PI and GI scores decreased after periodontal therapy for all groups ( $P<0.05$ ) compared to baseline values. No statistically significant differences in PD and CAL scores were found between baseline and after-therapy results for any group ( $P>0.05$ ). Although a statistically significant reduction in GCF volume was observed after therapy for groups 3 and 4, no statistically significant differences were found between baseline and after-therapy results for groups 1 and 2. Statistically significant differences were found between baseline and after-therapy values in GCF levels of MMP-8 for all groups ( $P=0.001$ ). When the MMP-8 levels in GCF of smokers and nonsmokers were compared after therapy, no statistically significant differences were observed between groups 1 and 3 and between groups 2 and 4 ( $P=0.117$ and $P=0.485$ , respectively)   |
| Lawrence et al. [88] | 42 | Untreated CP patients | 22 days | 1. 0.5% ketoprofen (KTP) gel topically applied 2×/day 14.5 days ( $N=9$ )<br>2. 1.0% KTP gel topically applied 2×/day 14.5 days ( $N=8$ )<br>3. 1.0% KTP alternate gel topically applied 2×/day 14.5 days ( $N=8$ )<br>4. 2.0% KTP gel topically applied 2×/day 14.5 days ( $N=8$ )<br>5. 25 mg KTP capsule (positive control) po 14.5 days ( $N=9$ )         | The intra-oral and peroral administrations of ketoprofen appeared safe, given the low incidence of adverse experiences and the absence of significant changes in vital signs and laboratory values during the study. No dose-dependent relationship was noted for ketoprofen with the incidence of adverse events, and there were no serious adverse events. At least 50% of the subjects in each treatment group reported one or more adverse experiences, which in general were of mild-to-moderate severity, and often unrelated to the study medication: headache, oral, and/or gastrointestinal symptoms at similar rates (22–25%) for both the topical and oral formulations. Furthermore, the four tested ketoprofen gel formulations appeared to be well tolerated by this compliant cohort. Results of the soft-tissue surveys indicated an absence of oral mucosal irritancy associated with the use of ketoprofen gels in subjects with moderate or severe adult periodontitis. Systemic ketoprofen administration resulted in significantly higher plasma concentrations compared with four topical ketoprofen formulations |
| Lee et al. [90]      | 19 | Chronic periodontitis | 3 weeks | 1. Mucoperiosteal flap surgery + low-dose flurbiprofen (LDF) alone, 50 mg q.d., 3 weeks<br>2. Mucoperiosteal flap surgery + SDD (20 mg b.i.d., 3 weeks) alone<br>3. Mucoperiosteal flap surgery + a combination of SDD + LDF (combination)  | Short-term therapy with SDD alone produced a significant reduction and LDF alone produced no reduction in host-derived neutral proteinases. However, the combination therapy produced a statistically significant synergistic reduction of collagenase, gelatinase, and serpinolytic (alpha 1-PI degrading) activities (69%, 69%, and 75% reductions, respectively) and a lesser reduction of the serine proteinase, elastase (46%) in gingival tissues   |

(continued)

**Table 6.3** (continued)

| Study                 | No. of patients | Periodontal condition   | Study period | Periodontal treatment   | Outcome  |
|-----------------------|-----------------|---|--------------|---|--|
| Ng & Bissada [104]    | 32              | Generalized moderate periodontitis                              | 24 weeks     | 1. SRP + DOXY (200 mg the first day followed by 100 mg/day, 6 weeks)<br>2. SRP + ibuprofen (800 mg/day, 6 weeks)<br>3. SRP + DOXY + ibuprofen<br>4. SRP + Placebo   | A statistical significance ( $P < 0.05$ ) from baseline data in (1) gains of 0.4 mm and 0.5 mm of CAL for groups 1 and 3, respectively; (2) reduction of 0.7 mm PD for group 3; (3) reduction of 0.4 and 0.1 GI scores for groups 1 and 3, respectively; and (4) gain of 0.5 mm CAL and reductions of 0.4 mm PD and 0.2 GI score for the SRP group when compared to the no SRP group at 24 weeks. In summary, it was concluded that the adjunctive use of systemic doxycycline alone or in combination with ibuprofen results in a statistically significant, yet modest clinical, improvement beyond that obtained by SRP.  |
| Paquette et al. [124] | 42              | Untreated generalized, moderate to advanced adult periodontitis | 22 days      | 1. 0.5% ketoprofen (KTP) gel topically applied 2×/day 14.5 days (N=9)<br>2. 1.0% KTP gel topically applied 2×/day 14.5 days (N=8)<br>3. 1.0% KTP alternate gel topically applied 2×/day 14.5 days (N=8)<br>4. 2.0% KTP gel topically applied 2×/day 14.5 days (N=8)<br>5. 25 mg KTP capsule (positive control) po 14.5 days (N=9) | For patients dosing with 0.5% ketoprofen gel, 2.0% ketoprofen gel or 25 mg ketoprofen capsule, PGE <sub>2</sub> levels remained suppressed throughout the 6 h observation period of day 1. When all time points for day 1 (i.e., 1, 2, 3, and 6 h) were averaged per group, no significant intergroup differences were detected for PGE <sub>2</sub> using an ANOVA procedure. On day 22 (7 days posttreatment), crevicular PGE <sub>2</sub> levels increased and were elevated as compared to baseline levels for the 1.0% and 1.0% alternate gel groups. Furthermore, PGE <sub>2</sub> levels were on average higher than values at the 2 h time point of day 15, the last observation period on drug for all treatments. When PGE <sub>2</sub> responses for all patients (gel formulations and positive control) were pooled and the corresponding means and confidence intervals were calculated, the data indicate significant% decreases in crevicular PGE <sub>2</sub> at 1 and 2 h on day 1 with investigational treatments ( $P < 0.005$ ). In addition, ketoprofen treatments resulted in significant increases in PGE <sub>2</sub> from day 15 (2 h) to day 22 ( $P < 0.005$ ). Concomitant with these reductions in PGE <sub>2</sub> levels, all ketoprofen formulations tended to increase crevicular LTB <sub>4</sub> concentrations. LTB <sub>4</sub> concentrations were elevated above baseline levels for all treatment groups on days 1, 8, and 15 (with the exception of the 25 mg capsule group on day 15). Overall day 1 mean% elevations in LTB <sub>4</sub> ranged from 31.1% to 71.6% for the 1.0% alternate gel and the 1.0% gel respectively. No significant intergroup differences in least square means or percent changes were observed for GCF LTB <sub>4</sub> ; however, when LTB <sub>4</sub> responses for all subjects were pooled, significant elevations (compared to baseline) were detected for all sampling times on days 1, 8, and 15 ( $P < 0.05$ ). These data also indicate that leukotriene levels rose 44% on average following drug withdrawal, from day 15 (2 h) to day 22 |

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| Preshaw et al. [132] | 42 | Moderately advanced chronic adult periodontitis                                | 6 weeks  | 1. Topical ketorolac tromethamine mouth rinse (0.1%) 2×/day, 2 weeks<br>2. Placebo rinse 2 weeks   | 14 days of rinsing with 0.1% ketorolac mouth rinse controlled the elevation of GCF PGE <sub>2</sub> , observed in the placebo group but did not actually reduce GCF PGE <sub>2</sub> concentrations. Changes in GCF PGE <sub>2</sub> levels were not detectable in the 12 h period following first rinsing with ketorolac   |
| Reddy et al. [139]   | 22 | Rapidly progressive periodontitis  | 6 months | 1. SRP + Meclofenamate sodium 50 or 100 mg 2×/day<br>2. SRP + placebo  | Bone change over the 6-month period as assessed by subtraction radiography was the primary efficacy determinant. Specialized software was used to isolate the lesion from the subtraction image and to measure bone change along the root surface. ANOVA using the subject as the unit of analysis revealed a significant dose response ( $P<0.001$ ) with the placebo group having a mean bone loss of $0.42 \pm 0.06$ mm and the low- and high-dose groups having mean bone gains of $0.07 \pm 0.05$ and $0.20 \pm 0.07$ mm, respectively. In summary, these findings indicate that meclofenamate sodium may be a useful adjunct in the treatment of rapidly progressive periodontitis  |
| Rozzman et al. [143] | 54 | Naturally occurring gingivitis   | 7 days   | 1. Aspirin, 81 mg 1×/day ( $N=17$ )<br>2. Aspirin, 325 mg 1×/day ( $N=20$ )<br>3. Placebo ( $N=17$ )   | The coefficient associated with treatment with 81 mg aspirin relative to BOP change from baseline was $4.76$ ( $P=0.008$ ; 95% CI: 1.29, 8.24), and the coefficient associated with treatment with 325 mg aspirin relative to BOP change from baseline was $3.46$ ( $P=0.034$ ; 95% CI: 0.26, 6.65). Treatment with placebo was not associated with any significant change in BOP relative to baseline. Linear regression analysis indicated that while controlling for age, gender, and plaque, “low dose” 81 mg and “regular dose” 325 mg of aspirin demonstrated a statistically significant $5.30$ ( $P=0.001$ ) and $4.13$ ( $P=0.010$ ) increase from baseline, respectively, in percent BOP  |
| Schrodi et al. [151] | 46 | Healthy periodontium and gingivitis  | 7 days   | 1. Aspirin, 81 mg 1×/day ( $N=15$ )<br>2. Aspirin, 325 mg 1×/day ( $N=15$ )<br>3. Placebo ( $N=16$ )   | A statistically significant difference in BOP was found in patients with $\geq 20\%$ of bleeding sites during the visit prior to placebo or aspirin exposure ( $N=11$ ). The group treated with 325 mg aspirin exhibited a moderate yet statistically significant increase in BOP (12.4%) compared to the placebo group (there was no significant difference between the 81 mg aspirin group and placebo). The tendency to bleed was not statistically significant in the group which exhibited $<20\%$ ( $N=35$ ) of bleeding sites during the visit prior to exposure   |
| Sculean et al. [152] | 22 | Periodontitis with at least one intrabony defect with a depth of at least 6 mm | 6 months | 1. GTR with enamel matrix proteins (EMD) + Systemic rofecoxib (12.5 mg 2×/day for 14 days)<br>1. GTR with enamel matrix proteins (EMD) alone | No statistical significant differences in any of the investigated parameters between the two groups were observed at baseline. At 6 months postoperatively, the results show that, in the test group, mean PD decreased from $8.7 \pm 2.0$ mm to $4.7 \pm 2.0$ mm ( $P<0.001$ ) and mean CAL from $9.7 \pm 2.1$ mm to $6.5 \pm 2.1$ mm ( $P<0.001$ ). In the control group, mean PD decreased from $8.6 \pm 1.6$ mm to $4.7 \pm 1.8$ mm ( $P<0.001$ ) and mean CAL from $9.5 \pm 1.6$ mm to $6.5 \pm 2.2$ mm ( $P<0.001$ ). In summary, the systemic administration of a selective COX-2 inhibitor following regenerative periodontal surgery with EMD did not result in additional clinical improvements when compared to treatment with EMD alone |

(continued)

**Table 6.3** (continued)

| Study               | No. of patients | Periodontal condition   | Study period | Periodontal treatment   | Outcome  |
|---------------------|-----------------|-------------------------|--------------|---|--|
| Sekino et al. [153] | 11              | Experimental gingivitis | 14 days      | 1. IBUP group: Systemic ibuprofen (tablets of 200 mg twice daily)<br>2. CHX group: Oral mouth rinse (positive control: 0.1% chlorhexidine digluconate twice a day, for 60 s with 10 ml)<br>3. CTRL group: Oral mouth rinse (negative control: saline) | The increase in the proportion of inflamed gingival units was significantly smaller in CHX than in IBUP and CTRL ( $P < 0.01$ ) and significantly smaller in IBUP than in CTRL ( $P < 0.05$ ). A further analysis of the GI score categories revealed that the percentage of sites that on day 0 were identified as being healthy (GI score 0) but on day 14 had become inflamed (GI score $\geq 2$ ) varied between treatments. Thus, in CHX the proportion of such changing sites was 7% while in IBUP it was 16% and 20% in CTRL. Also, in this comparison, the increase was less pronounced in CHX than in IBUP and CTRL ( $P < 0.01$ )  |
| Toker et al. [163]  | 30              | Chronic periodontitis   | 1 month      | 1. SRP + instructions on daily plaque control + Meloxicam (7.5 mg, 1×/day, 1 month)<br>2. SRP + instructions on daily plaque control + placebo tablets  | There were no significant differences in both the baseline and the 1-month PD, CAL, PI, and BI values between the meloxicam and placebo groups ( $P > 0.05$ ). There was no significant difference between the baseline and the 1-month CAL values of the meloxicam group ( $P > 0.05$ ), but the 1-month PD value was significantly lower than the baseline PD value ( $P > 0.05$ ). While IL-1ra was detected in all GCF samples (100%), IL-1 $\beta$ was detected in 66% of all the samples. There were no significant differences in both the baseline and the 1-month IL-1 $\beta$ and IL-1ra levels between the study groups ( $P > 0.05$ ). There was no significant difference between the baseline and the 1-month IL-1 $\beta$ levels of the meloxicam group ( $P > 0.05$ ). In the meloxicam group, the 1-month IL-1ra levels were significantly lower than the baseline IL-1ra levels ( $P < 0.05$ ) |
| Vogel et al. [179]  | 18              | Experimental gingivitis | 22 days      | 1. Placebo gel to apply topically and placebo capsules<br>2. Placebo gel and capsules containing sulindac<br>3. Topical flucinonide (steroid) gel and placebo capsules  | Only the topical steroid drug significantly inhibited gingival inflammation, while the systemically administered nonsteroidal drug had no apparent effect  |
| Vardar et al. [175] | 30              | Chronic periodontitis   | 3 months     | 1. SRP + systemic nimesulide (100 mg 2×/day, 10 days)<br>2. SRP + naproxen (275 mg 2×/day, 10 days)<br>3. SRP + Placebo (2×/day, 10 days)   | All three groups showed statistically significant reductions in PBI and PI on day 10 and at 3 months ( $P < 0.02$ , and in PD and CAL at 3 months ( $P < 0.02$ , $P < 0.05$ , respectively). In the naproxen group, gingival tissue levels of PG <sub>E</sub> <sub>2</sub> exhibited a significant decrease ( $P < 0.05$ ). However, the decrease of gingival tissue levels of PG <sub>E</sub> <sub>2</sub> in the nimesulide group was insignificant ( $P > 0.05$ ), while a significant increase was observed in the placebo group ( $P < 0.05$ ) on day 10. Both the nimesulide and naproxen groups showed a significant decrease ( $P < 0.05$ ) in PG <sub>E</sub> <sub>2</sub> alpha level, while the placebo group showed a significant increase ( $P < 0.05$ )  |

|                       |     |                                   |           |   |   |
|-----------------------|-----|-----------------------------------|-----------|---|---|
| Williams et al. [193] | 44  | Chronic periodontitis             | 24 months | 1. Subgingival scaling and pumice every 6 months + Systemic flurbiprofen (50 mg, 2×, 18 months)<br>2. Subgingival scaling and pumice every 6 months + placebo | After 12 and 18 months, significantly less alveolar bone loss in flurbiprofen-treated patients compared with controls. At 24 months, rate of bone loss returned to pretreatment levels in both patient groups   |
| Yen et al. [198]      | 101 | Chronic periodontitis             | 12 months | 1. SRP+celecoxib (200 mg, 1×, 6 months) ( <i>N</i> =54)<br>2. SRP+placebo ( <i>N</i> =47)   | When mean PD values from the placebo and celecoxib groups were compared at follow-up visits, sites from the celecoxib group were consistently shallower than in the placebo group in all sites at every follow-up visit. This difference was more pronounced for deep sites where mean PD ranged from 3.8 to 4.49 mm in the celecoxib group and from 5.58 to 6.12 mm in the placebo group. Statistically, the differences in mean PD were significant ( $P<0.001$ ) except at 6 and 9 months for the moderate sites. In deep sites, the celecoxib group showed an average reduction in PD of 3.27 mm at 3 months and 3.84 mm at 12 months, whereas the respective values in the placebo group were 1.89 mm and 2.06 mm. The differences at each time point were statistically significant ( $P<0.001$ ). For moderate sites, the trend was similar to the deep sites, with the changes being of smaller magnitude (1.14–1.25 mm for the celecoxib group versus 0.98–0.99 mm for the placebo group). The celecoxib group also exhibited a greater percentage of sites with $\geq 2$ mm CAL gain and fewer sites with $\geq 2$ mm CAL loss. Both groups showed improved plaque control and BOP scores |
| Abramson et al. [1]   | 21  | Gingivitis and mild periodontitis | 57 days   | 1. Flurbiprofen (100 mg, 3×/57 days) ( <i>N</i> =10)<br>2. Placebo ( <i>N</i> =11)  | GCF-PGE <sub>2</sub> and GCF-TxB <sub>2</sub> levels were significantly reduced in the flurbiprofen treated patients. One week after drug administration was discontinued, GCF levels of PGE <sub>2</sub> and TxB <sub>2</sub> returned to pretreatment levels  |
| Jeffcoat et al. [70]  | 15  | Refractory periodontitis          | 2 months  | 1. Flurbiprofen (50 mg, 2×/day, 3 months) ( <i>N</i> =8)<br>2. Placebo ( <i>N</i> =7)   | Radioispharmaceutical uptake was significantly reduced in the alveolar bone of teeth undergoing active bone loss at the start of the study in patients receiving flurbiprofen ( $P<0.04$ ), whereas no significant change was observed in the placebo-treated patients. There was significantly less bone loss during the 2-month study period in the flurbiprofen-treated patients when compared to the placebo-treated patients ( $P<0.02$ )  |

TBARS thiobarbituric acid reactive substance; BOP bleeding on probing; NSAID nonsteroidal anti-inflammatory drugs; GCF gingival crevicular fluid; CAL clinical attachment level; ASA acetylsalicylic acid; GTR guided-tissue regeneration; CP chronic periodontitis; RPP rapidly progressing periodontitis; EMD enamel matrix derivative; LT<sub>B</sub><sub>4</sub> leukotriene B<sub>4</sub>; PGE<sub>2</sub> prostaglandin E2; IL-6 interleukin 6; IL-*beta* interleukin 1; 15-*epi*-LXA<sub>4</sub> 15-*epi*-lipoxin A<sub>4</sub>

**Table 6.4** Randomized controlled trials of selective nonsteroidal anti-inflammatory drugs gastrointestinal effects

| Author                  | Study name   | Study subjects  | Study length | NSAIDs treatment  | Outcome  |
|-------------------------|--|---|--------------|---|--|
| Bombardier et al. [17]  | VIGOR (Vioxx Gastrointestinal Outcomes Research)                         | 8,076 rheumatoid arthritis patients                     | 9.0 months   | <ul style="list-style-type: none"> <li>• Rofecoxib 50 mg once daily</li> <li>• Naproxen 500 mg 2×/day</li> </ul>  | The study revealed that in patients with rheumatoid arthritis, treatment with rofecoxib at twice the maximal dose approved by the FDA for long-term use resulted in significantly lower rates of clinically important upper gastrointestinal events and complicated upper gastrointestinal events than did treatment with a standard dose (1,000 mg/day) of naproxen. It was also found that the incidence of complicated upper gastrointestinal bleeding and bleeding from beyond the duodenum was significantly lower among patients who received rofecoxib  |
| Silverstein et al. [58] | CLASS (Celecoxib Long-term Arthritis Safety Study)                       | 7,968 rheumatoid arthritis and osteoarthritis patients  | 6 months     | <ul style="list-style-type: none"> <li>• Celecoxib, 400 mg, 2×/day</li> <li>• Ibuprofen, 800 mg, 3×/day</li> <li>• Diclofenac, 75 mg, 2×/day</li> </ul>   | For all patients, the annualized incidence rates of upper GI ulcer complications alone and combined with symptomatic ulcers for celecoxib vs. NSAIDs were 0.76% vs. 1.45% ( $P=0.09$ ) and 2.08% vs. 3.54% ( $P=0.02$ ), respectively. For patients not taking aspirin, the annualized incidence rates of upper GI ulcer complications alone and combined with symptomatic ulcers for celecoxib vs. NSAIDs were 0.44% vs. 1.27% ( $P=0.04$ ) and 1.40% vs. 2.91% ( $P=0.02$ ). For patients taking aspirin, the annualized incidence rates of upper GI ulcer complications alone and combined with symptomatic ulcers for celecoxib vs. NSAIDs were 2.01% vs. 2.12% ( $P=0.92$ ) and 4.70% vs. 6.00% ( $P=0.49$ ). Fewer celecoxib treated patients than NSAID-treated patients experienced chronic GI blood loss, GI intolerance, hepatotoxicity, or renal toxicity |
| Laine et al. [83]       | MEDAL (Multinational Etoricoxib and Diclofenac Arthritis Long-term)      | 34,701 rheumatoid arthritis and osteoarthritis patients | 18 months    | <ul style="list-style-type: none"> <li>• Etoricoxib (60 or 90 mg/day)</li> <li>• Diclofenac (150 mg/day)</li> </ul>   | Overall upper gastrointestinal clinical events were significantly less common with etoricoxib than with diclofenac (hazard ratio = 0.69, 95% CI: 0.57–0.83; $P=0.0001$ ). There were significantly fewer uncomplicated gastrointestinal events with etoricoxib than there were with diclofenac (0.57, 0.45–0.74; $P<0.0001$ ); there was no difference in complicated events (0.91, 0.67–1.24; $P=0.561$ ). The reduction in uncomplicated events with etoricoxib is maintained in patients treated with PPIs and is also observed with regular low-dose aspirin use   |
| Schnitzer et al. [150]  | TARGET (Therapeutic Arthritis Research and Gastrointestinal Event Trial) | 18,325 osteoarthritis patients aged 50 years or older   | 1 year       | <ul style="list-style-type: none"> <li>• Lumiracoxib 400 mg/day (two or four times the recommended chronic dose for osteoarthritis)</li> <li>• Naproxen 500 mg twice daily (maximum therapeutic dose)</li> <li>• Ibuprofen 800 mg three times daily (maximum therapeutic dose)</li> </ul> | The results of the TARGET study show a 79% reduction in ulcer complications in patients taking lumiracoxib compared with two frequently used anti-inflammatory drugs in a non-aspirin population. Similar reductions were noted when lumiracoxib was compared individually with ibuprofen (83% reduction) or naproxen (76% reduction). The yearly incidence of ulcer complications on lumiracoxib was very low (0.26 events per 100 patient-years)   |

NSAIDs give rise to symptoms such as hyperkalemia, sodium retention, acute renal failure, declined glomerular filtration rate, nephrotic syndrome with acute interstitial nephritis, renal papillary necrosis, and edema. Renal side effects of NSAIDs are rare, sometimes transient, and often reversible upon drug withdrawal. The incident rate and the severity of the renal side effect, however, increase in patients with risk factors such as those with diabetes, heart failure, renal dysfunction, and old age. The side effects range from electrolyte retention and reduce glomerular filtration to nephritic syndrome and chronic renal failure. These effects are shared among NSAIDs with evidence of dose and exposure dependency. There is no known predictor for the nephrotoxicity. However, a relationship has been found between high plasma concentration and the renal adverse effect of NSAIDs [55].

Suspicion about an increased risk for **cardiovascular events** associated with the use of COX-2 inhibitors, namely myocardial infarction (MI) and ischemic stroke, was raised during the 2000 VIGOR study, which demonstrated a significantly increased cardiovascular risk among patients taking rofecoxib compared with those taking nonselective NSAID [183]. As a consequence, on September 30, 2004, Merck and Co. voluntary withdrew rofecoxib (Vioxx) from the market due to increased risk of cardiovascular events associated with the drug. It is unclear whether the cardiac toxicities associated with rofecoxib are due to its high COX-2 selectivity. Rofecoxib is the most specific COX-2 inhibitor among the first generation of the class, i.e., negligible COX-1 inhibitory effect [34]. COX-2 inhibitors may increase the incidence of thromboembolic events by impairing the synthesis of PGI<sub>2</sub>, with relatively unopposed platelet thromboxane A<sub>2</sub> activity, although both COX inhibitors may promote thrombus formation by altering normal cholesterol transport [183].

Traditional NSAIDs have long been considered to pose no cardiovascular hazard, except for their hemodynamic effects potentially enhancing the risk of congestive heart failure. In fact, they were often considered potentially cardioprotective because of their inhibitory effects on platelet COX-1. The results corroborate the statistical heterogeneity between coxib-versus-naproxen (yielding a rate ratio virtually identical to that of coxib-versus-placebo) and coxib-versus-non-naproxen NSAID comparisons (now yielding a rate ratio slightly lower than 1.0). Thus, non-naproxen

NSAIDs, particularly high-dose diclofenac and high-dose ibuprofen, appear to share the cardiotoxic phenotype associated with coxibs. Given the size of the cardioprotective effect of low-dose aspirin in low-risk trials (see above), the statistical uncertainty surrounding the point estimate of high-dose naproxen does not allow us to discriminate between a neutral and an aspirin-like cardioprotective phenotype [125].

Prostaglandins play an important role in the regulation of osteoblast and osteoclast functions, and inhibition of prostaglandin production retards bone formation. Therefore, NSAIDs could be expected to have significant consequences in divergent clinical situations where bone formation or remodeling is a contributing factor. NSAIDs are used clinically to prevent ectopic bone formation (also known as heterotopic ossification) (e.g., after total hip arthroplasty or trauma). The efficacy of NSAIDs in the avoidance of heterotopic ossification has been documented in controlled clinical trials, but the inherent risks (e.g., on healing processes and on loosening of prostheses) need further studies. At the same time, NSAIDs are widely used in the treatment of fracture pain, and their inhibitory effects on the ongoing bone healing process have raised concerns. Results of fracture-healing studies in animals treated with NSAIDs or in mice lacking COX-2 gene show that inhibition or deficiency of COX-2 **impairs the bone-healing process**. The limited clinical data also support the assumption that inhibition of COX-2 by nonselective or COX-2-selective NSAIDs delays fracture healing and negatively interferes with spinal fusion in both humans and other animals, whereas the alleged inhibitory effects of COX-2-selective NSAIDs still lack experimental and clinical evidence [45, 50, 54, 109, 182].

By first elucidating the steps involved in the bone-healing process, the mechanism through which NSAIDs serve to inhibit bone healing becomes clearer. Successful bone healing requires an inflammatory response, bone resorption by osteoclasts, and bone formation by osteoblasts. The investigations described previously serve to establish a role for PGs in each of these processes. In other words, PGs fulfill an important position in the course of bone healing. It is not surprising, therefore, that the decreased production of PGs elicited by NSAIDs could potentially impair the bone-healing process [54]. The data acquired so far is, nevertheless, suggestive to recommend alternative pain

treatment modalities in clinical situations where impaired bone healing is a problem, for example, when treating nonunion of fractures, arthrodesis, or osteotomies [182].

Only very few studies assessed the safety and tolerance of the NSAIDs used in clinical periodontal trials [6, 88, 124, 143, 152, 163, 175, 198] and none of the studies reported adverse effects, except for [88].

In a clinical trial with a randomized, partially double-blind, controlled parallel design, Lawrence et al. [88] evaluated the pharmacokinetics and safety of the NSAID, ketoprofen, in gel formulations. Forty-two subjects, aged 35–57 years, with generalized, moderate-to-advanced adult periodontitis were recruited and randomized to one of five treatments over a 14.5-day treatment period: (1) 0.5% ketoprofen gel, (2) 1.0% ketoprofen gel, (3) 1.0% ketoprofen alternate gel, (4) 2.0% ketoprofen gel, and (5) 25 mg ketoprofen capsule. At least 50% of the subjects in each treatment group reported one or more adverse experiences, which in general were of mild-to-moderate severity, and often unrelated to the study medication. There was no statistically significant difference among the five treatment groups in terms of adverse experiences incidence ( $P=0.504$ ). While no serious adverse experiences were recorded for this cohort of subjects, the most frequently cited adverse experiences were headache. All groups reported oral and/or gastrointestinal symptoms at similar rates (22–25%) for both the topical and oral formulations.

NSAIDs and especially selective COX-2 inhibitors are important in the management of postoperative and postfracture pain and lead to considerable reduction of opioid needs. Sculean et al. [152] investigated the effect of rofecoxib (25 mg/day for 14 days) on the healing of intrabony periodontal defects in humans who were treated with an enamel matrix protein derivative and found no inhibitory effect. Dastoor et al. [33] evaluated the adjunctive effects of systemic azithromycin in combination with periodontal pocket reduction surgery in the treatment of chronic periodontitis in smokers. Preoperatively, a 0.12% chlorhexidine rinse and ibuprofen, 600 mg, were given to all patients. Postoperative analgesia was provided through nonsteroidal anti-inflammatory drugs (ibuprofen, 600 mg every 6 h for 2 days, and thereafter as needed). No

adverse drug reactions were reported at any time during the 6 months' study length.

It seems that the short-term administration of NSAIDs delays fracture healing but does not inhibit it and that this effect is both dose-dependent and reversible. It also seems that the effect of NSAIDs is stronger in the early stages of fracture healing, the inflammatory and hematoma stage, and that as soon as NSAIDs administration is discontinued, the normal process of healing is restored and the initially adverse effects are reversed [11, 21, 49].

Reports concluding that chronic use of nonsteroidal anti-inflammatory drugs (NSAIDs) increases the likelihood of cardiovascular adverse events have led patients and clinicians to reduce use. It is intriguing to recollect that the cardiotoxicity of NSAIDs (both COX-2 selective inhibitors and nonselective dual inhibitors have been implicated) would not have been revealed if not for the push to market novel drugs developed to reduce the well-documented gastrointestinal complications of this widely used class. While weighing risks and benefits of interventions is an essential component of clinical decision making, it has become increasingly complex to decide in whom the use of any NSAID – with or without a gastroprotective agent, and with or without concomitant aspirin – is appropriate [149].

Currently, there is insufficient evidence to guide decision making on NSAID use, and as a consequence, it is not surprising that current guidelines and recommendations on the use of NSAIDs [4, 5, 149, 161] come to different, sometimes contradictory conclusions, particularly with regard to aspirin-treated patients at high risk of vascular complications [125] (Tables 6.5–6.7). American Heart Association suggested a stepped-care approach to management of patients with musculoskeletal symptoms; and indicated that the risk for adverse cardiovascular effects that include increased risk for myocardial infarction, stroke, heart failure, and hypertension is likely greatest in patients with a prior history of, or at high risk for, cardiovascular disease. In these patients, use of COX-2 inhibitors for pain relief should be limited to patients for whom there are no appropriate alternatives, and then, only in the lowest dose and for the shortest duration necessary [5].

**Table 6.5** Recommendations of the American College of Rheumatology ad hoc group on use of selective and nonselective nonsteroidal anti-inflammatory drugs aimed at the practicing clinician (Reprinted from [4]. With permission from John Wiley and Sons)

|                        |   |
|------------------------|---|
| Efficacy/effectiveness | If a patient and provider agree to utilize an NSAID for arthritis pain relief and the patient does not respond to one agent, then other agents may be tried. Some patients respond differently to different NSAIDs.<br><br>If a patient and provider agree to utilize an NSAID for arthritis pain relief and the patient is at low risk for toxicity, then the lowest effective dose of the least expensive agent should be considered first line. Low doses of NSAIDs are safer than high doses. There are no compelling data to support the selection of one agent over another if there are no significant toxicity concerns.  |
| Toxicity               | If a patient and provider agree to utilize an NSAID (nonselective or selective) for arthritis pain relief, then the patient should be advised of the potential toxicities and relevant monitoring (complete blood cell count, renal function, liver function, and blood pressure) should be pursued.<br><br>If a patient is taking aspirin for cardioprotective benefit, then selective and nonselective NSAIDs should be avoided. This combination is associated with an elevated risk of GI bleeding. However, if a patient is educated about this risk and wants to take the drugs concomitantly, then a PPI or misoprostol should be added to the regimen.<br><br>If a patient is at moderate-to-high risk of a future cardiovascular event, is taking low-dose aspirin for cardioprotection, and the patient and provider agree that continuous treatment for arthritis pain relief is needed, then the patient should be managed initially with acetaminophen or naproxen. Selective NSAIDs and other nonselective NSAIDs have been associated with an increased cardiovascular risk. Note that naproxen may also confer cardiovascular risk when used intermittently or at low doses that do not inhibit platelet aggregation.<br><br>If a patient and provider agree to utilize an NSAID for arthritis pain relief, and the patient is taking low-dose aspirin for cardiovascular prevention, then continuous use of ibuprofen should be avoided. There is a potential drug-drug interaction between aspirin and ibuprofen that reduces cardioprotective benefit. This may be true for other nonselective NSAIDs, but there are insufficient data to assess the interaction. Selective NSAIDs do not appear to have relevant drug-drug interactions with the anticoagulant effects of aspirin.<br><br>If a patient and provider agree to utilize an NSAID for arthritis pain relief, and the patient has risk factors for GI bleeding, then the patient should be treated concomitantly with either misoprostol or a PPI. These agents will reduce the risk of GI bleeding.<br><br>If a patient has renal insufficiency, then use of both selective and nonselective NSAIDs should be avoided.<br>If a patient has compromised liver function, then the risks of selective and nonselective NSAID use should be carefully considered. Although severe hepatotoxicity with NSAIDs is rare, NSAIDs are associated with liver function abnormalities. Diclofenac should be avoided in patients with liver disease.<br><br>If a patient is fully anticoagulated with warfarin, heparin, or other anticoagulants or is thrombocytopenic, then use of nonselective NSAIDs should be avoided because they can increase the risk of bleeding. |

**Table 6.6** Clinicians guide to anti-inflammatory therapy (Reprinted from [149]. With permission from Elsevier)

|  | No or low NSAID gastrointestinal risk  | NSAID gastrointestinal risk   |
|--|--|---|
| No cardiovascular risk (without aspirin) | Nonselective NSAID (cost consideration)  | COX-2 selective inhibitor or nonselective NSAID + proton-pump inhibitor<br>COX-2 selective inhibitor + proton-pump inhibitor for those with prior gastrointestinal bleeding                               |
| Cardiovascular risk (with aspirin)       | Naproxen*<br><br>Addition of proton-pump inhibitor if gastrointestinal risk of aspirin/NSAID combination warrants gastroprotection | Proton-pump inhibitor irrespective of NSAID<br>Naproxen if CV risk outweighs gastrointestinal risk<br>COX-2 selective inhibitor + proton-pump inhibitor for those with previous gastrointestinal bleeding |

\*Nonselective or selective (low-dose) inhibitor without established aspirin interaction if naproxen is ineffective. Misoprostol at full dose (200 µg four times a day) may be substituted for proton-pump inhibitor

**Table 6.7** Practice points for using NSAIDs (Reprinted from [41]. With permission from Elsevier)

1. When COX-2 inhibitors and NSAIDs are to be used for the management of individual patients, they should be prescribed with the lowest effective dose and for the shortest duration.
2. They should not be prescribed for high-risk patients, e.g., patients with a history of ischemic heart disease, stroke or congestive heart failure, or in patients who have recently undergone CABG.
3. All prescription-strength NSAIDs will now display “black box” label warnings for the potential risk of cardiovascular and gastrointestinal adverse effects.
4. Treatment with NSAIDs alone in patients aged less than 65 years who do not have gastrointestinal risk factors is considered appropriate. Co-therapy with a proton pump inhibitor or treatment with a COX-2 inhibitor was considered unnecessary in these patients.
5. The use of an NSAID alone was considered inappropriate in any patient with previous gastrointestinal event and in those who concurrently receive aspirin, steroids, or warfarin. These patients should receive either an NSAID plus a proton pump inhibitor or a COX-2 inhibitor.
6. Use of a COX-2 inhibitor with proton pump inhibitor co-therapy is appropriate only in patients at very high risk, such as those with a previous gastrointestinal event who are taking aspirin, and those who are taking aspirin plus steroids or warfarin.

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