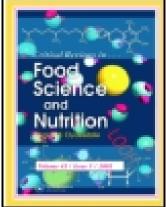
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Antimicrobial Activity of Coriander Oil and Its Effectiveness as Food Preservative

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Antimicrobial activity of coriander oil and its effectiveness as food preservative

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Abstract

Foodborne illness represents a major economic burden worldwide and a serious public health threat, with around 48 million people affected and 3000 death each year only in the USA. One of possible strategies to reduce foodborne infections is the development of effective preservation strategies capable of eradicating microbial contamination of foods. Over the last years, new challenges for the food industry have arisen such as the increase of antimicrobial resistance of foodborne pathogens to common preservatives and consumers demand for naturally-based products. In order to overcome this, new approaches using natural or bio-based products as food preservatives need to be investigated. Coriander (*Coriandrum sativum L.*) is a well-known herb widely used as spice, or in folk medicine, and in the pharmacy and food industries. Coriander seed oil is the worldøs second most relevant essential oil, exhibiting antimicrobial activity against Gram-positive and Gram-negative bacteria, some yeasts, dermatophytes and filamentous fungi. This review highlights coriander oil antimicrobial activity and possible mechanisms of action in microbial cells and discusses the ability of coriander oil usage as a food preservative, pointing

out possible paths towards the successful evolution for these strategies towards a successful development of a food preservation strategy using coriander oil.

Keywords: foodborne disease, Coriandrum sativum L., natural antimicrobials, food preservation

Introduction

Illness and death caused by the ingestion or contact with contaminated food are a constant threat to public health security as well as socioeconomic development throughout the world (Kuchenmuller et al., 2009). For instance, CDC estimates that each year roughly 48 million Americans succumb, 128,000 are hospitalized and 3,000 die of foodborne diseases (http://www.cdc.gov/foodborneburden/2011-foodborne-estimates.html). In 2012, identified a total of 19531 laboratory-confirmed cases of infection that resulted in 4563 hospitalizations and 68 deaths (2013). The major pathogens, both bacterial and parasitic, causing foodborne illness were Salmonella (39.94%), Campylobacter (34.78%), Shigella (10.95%), Cryptosporidium (6.32%) and non-Shiga (non-STEC O157) and Shiga toxin-producing E. coli (STEC O157) (5.54%) (Figure 1a). Regarding foodborne illness resulting in hospitalization, data showed that the percentage of incidence is very similar to the hospitalization for each pathogen (Figure 1b). Nonetheless, when observing the percentage of deaths resulting from each pathogen, it can be observed that the mortality rate (percentage of hospitalizations resulting in death) is higher for Listeria (11.21%), Vibrio (10.91%) while only approximately 1% of major pathogens hospitalizations, such as Campylobacter and Salmonella, resulted in death (Figure 1c). According to the last WHO report, in England and Wales, six foodborne pathogens are responsible for 93% of all known-ethiology foodborne infection cases: non-typhoidal Salmonella, Campylobacter, Yersinia, C. perfringens, STEC non-O157 and norovirus (Adak et al., 2002). In Europe, salmonellosis is still the most frequently reported foodborne disease though its incidence is decreasing. On the other hand, campylobacteriose incidence is increasing throughout Europe, being the most common gastrointestinal pathogen in the Netherlands,

England and Wales, Scotland, Finland, Denmark, Norway, Sweeden, Iceland and Switzerland (Tirado and Schmidt, 2001). All these findings regarding the incidence of foodborne microbial illness have raised awareness for the global burden that foodborne diseases represent. For this reason, new ways to prevent and control foodborne infections are in demand. Over the past decades, due to the enrichment of multiple-antimicrobial resistant microorganisms, both pathogenic and commensal, much effort is being made in designing and developing more effective ways to avoid foodborne microbial infections in humans (Teuber, 1999). One of the chosen ways to reduce or even eliminate microbial contamination in foods is the addition of preservatives, synthetic or natural, supplemented directly to foods or incorporated in the food packages. The most common chemical preservatives in use are weak acids (e.g. acetic, lactic, benzoic and sorbic acids), hydrogen peroxide and chelators (citric acid, disodium and calcium salts of ethylenediamine- tetraacetic acid) (for a more detailed review see (Brul and Coote, 1999)). Weak acids inhibit the outgrowth of bacterial and fungal cells (Hirshfield et al., 2003), hydrogen peroxide generates biocidal singlet oxygen species (Cerioni et al., 2013; Toivonen et al., 2012) and chelators play a role in disrupting the biogenesis of a normal cell wall (Marvin et al., 1989; Prachayasittikul et al., 2007). Although these compounds have been used extensively in food preservation, they can lead to the development of microbial resistance through preservative degradation by specific enzymes, selective permeability to preservatives, among other mechanisms (Table 1) (Parry-Hanson et al., 2010; Plumridge et al., 2008; Ravirala et al., 2007). As a consequence of the antimicrobial resistance to chemical preservatives, over last years, researchers have focused on the study and exploitation of natural products antimicrobial activity for use as preservatives, becoming increasingly accepted that natural based products are

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inherently better tolerated in the body and have innate advantages for the food industry over synthetic chemical preservatives (Kadan et al., 2013). Plant antimicrobials are phytochemicals that play an important role in the plant defense mechanisms against pathogens and predators. These phytochemicals are generally grouped in phenolic compounds (e.g. phenols, phenolic acids, quinones, flavonoids and tannins), terpenoids and essential oils, alkaloids, lectins and polypeptides. The addition of these phytochemicals to foods (revised in (Sultanbawa, 2011)) can extend the shelf-life of products, due to their known antimicrobial and antioxidant properties. On the contrary of chemical preservatives, where toxic effects have been described and are still debated, the use of phytochemicals can even result in beneficial health effects for the consumer. In recent years, there has been a renewed interest on the essential oils extracted from plant materials (leaves, flowers or buds, bulbs, seeds, rhizome and fruits, among others), since they contain a wide range of natural components, such as phenolic compounds, terpenes, esters, aldehydes, ketones, acids, isoflavonoids and aliphatic alcohols. Most of essential oils used possess a cGenerally Recognised As Safeo (GRAS) status as designated by the American Food and Drug Administration (Smith et al., 2008) and have a broad spectra of antimicrobial action against different pathogenic and spoilage microorganisms (Hammer et al., 1999; Si et al., 2006). Some of the most widely used essential oils are the ones extracted from the coriander plant (Coriandrum sativum L.), either from the seeds or from the leaves of the plant (Matasyoh et al., 2009; Telci et al., 2006b). Coriander and coriander essential oils have a vast range of biological activities, such as antimicrobial, anti-oxidant, anti-diabetic, anxiolytic, anti-epileptic, antianti-mutagenic, depressant, anti-inł ammatory, anti-dyslipidemic, anti-hypertensive, neuroprotective and diuretic (revised in detail in (Sahib et al., 2012)). In view of the potential

uses of *Coriandrum sativum L*. essential oils in the food industry, we provide a comprehensive review about coriander antimicrobial activities and discuss its potential application in the food industry as a preservative, added directly to foods or incorporated in a packaging material. Since essential oil antimicrobial activity is largely dependent on its composition, the phytochemistry of coriander essentials oils will also be briefly discussed.

1. Coriander

The erect, glabrous, annual plant coriander (*Coriandrum sativum L*.) is grown especially in the Indian sub-continent but also in Europe, Northern Africa and Asia for culinary, aromatic and medicinal uses. Its herbage, generally designated as cilantro, is used in many foods. The ground seeds of coriander are used as spice, for instance, in curry powders. The essential oil extracted from coriander fruits (generally referred as seeds) is a main flavouring agent in many food items, such as gin, breads, soups, sauces, canned goods and candy.

The species has a very wide range of distribution and a formal taxonomy distinguishing three subspecies and ten botanical varieties (Diederichsen, 1996a; Diederichsen and Hammer, 2003).

1.1. Cultivar and Chemotaxonomy

Infraspecific studies of variation in C. sativum L. have been performed, mostly based on morphological characteristics in conjunction with a few chemical features. Over the years, coriander subspecies classification has suffered some alterations. In 1981, coriander was divided in two types, based on fruit size: large fruits (designated var. *vulgare* Alef., also called var. *sativum*) and small fruits (var. *microcarpum* DC) (Purseglove et al., 1981). Large fruited plants generally grow in tropical and subtropical areas whereas small-fruited plants are produced in temperate regions (Purseglove et al., 1981). Later, in 1990, a new classification was proposed by

Ivanova and Stoletova based on the division of coriander production in four geographic centres, each one corresponding to a different subspecies of coriander: *indicum* Stolet. (India), *sativum* (Northern Africa), *asiaticum* Stolet. (Central Asia) and *vavilovii* Stolet. (Abyssinia) (Lopez, 2006). More recently, in 1996, Diederichsen proposed six ecogeographic coriander types: Near Eastern, Indian, Central Asian, Syrian, Caucasian and Ethiopian (Diederichsen, 1996). Based on the extensive analysis on morphological variation and essential oil composition performed in the previous work (Diederichsen, 1996), in 2003, a new study analysing several parameters (patterns of phenological and morphological traits, seed essential oil and faty acid content) recommended the division in three subspecies: *sativum*, *microcarpum* DC and *indicum* (Diederichsen and Hammer, 2003).

Regarding coriander botanical varieties, Ivanova and Stoletova first described the existence of 9 varieties: sativum, africanum Stolet., asiaticum Stolet., anatolicum Stolet., afghanicum Stolet., vavilovii Stolet., arabicum Stolet., indicum Stolet. pygmaeum Stolet (Diederichsen and Hammer, 2003). Due to some inconsistencies in nomenclature rules and new data on chemical analysis of volatile and fixed oils in the fruit, this division was revised in 2003 by Diederichsen and Hammer. This study proposed the division in 10 varieties based on 20 descriptors selected from phenology traits (4 characters), vegetative plant parts (7 characters), generative plant parts (5 characters) and volatile and fixed oil in the fruits (4 characters) (Diederichsen and Hammer, 2003). In this new division, four coriander botanical varieties previously proposed by Ivanova and Stoletova (sativum, africanum Stolet. and asiaticum Stolet.) were maintained, the microcarpum (DC.) variety proposed by Hegi in 1926 was included and six new varieties: syriacum Diederichsen (var. nova), vavilovii (Stolet.) Diederichsen (comb. nova), indicum Stolet.

ex Diederichsen (var. nova), *bhutanense* Diederichsen (var. nova), *omanense* Diederichsen (var. nova) and *pygmaeum* Stolet. Ex Diederichsen (var. nova) were described (Diederichsen and Hammer, 2003). This diversity of coriander types and other parameters can influence the yield and composition of the essential oil. (Telci et al., 2006b). For instance, it was described that var. microcarpum DC. yields almost two times more essential oil than var. vulgare and contains more oxygenated monoterpenes and a higher linalool content together with a lower monoterpene content (Telci et al., 2006b).

1.2. Coriander essential oil

International Organization for Standardization (ISO) (ISO DIS9235.2) defines an essential oil as ãa product made by distillation with either water or steam or by mechanical processing of citrus rinds or by dry distillation of natural materialsö, meaning that an extract can only be called essential oil if it is obtained by steam or hydrodistillation. In recent years, new extraction processes such as supercritical or subcritical (Eikani et al., 2007) extractions with water or other solvents (e.g. CO₂) (Grosso et al., 2008) have been described for the obtention of allegedly called õessential oilsö. This designation raised great controversy among scientists working in the field since, by the ISO definition, the extracts obtained by these techniques cannot be designated as essential oils. Hydrodistillation has a different mechanism of extraction (mainly distillation) while other water extraction and Soxhlet extraction processes consist mainly in the dissolution and/or solubilisation of the essential oil in the solvent (extraction process) (Eikani et al., 2007). Due to this definition, for the purpose of this review, we will only take into account studies performed with coriander oil obtained by steam or hydrodistillation.

1.2.1. Oil composition

Coriander seeds contain up to 1% of essential oil with linalool being the main component, accounting for approximately 60% of the oil & components. Typical coriander oil compositional analysis (Table 1) includes linalool (57-72%), terpinen-4-ol (0.1-0.5%), -terpinene (4.2-9.3%), p-cymene (1.12-4.0%), limonene (0.3-3.7%), -pinene (1.63-4.81%), -pinene (<0.1-0.6%), camphene (<0.1-0.92%), myrcene (0.18-1.3%), sabinene (0.1-0.3%) camphor (0.2-6.4%), geranyl acetate (1.0-5.0%) and linallyl acetate (1.25-7.1%), among other terpene hydrocarbons and oxygenated compounds. Coriander essential oil is also regulated by ISO (ISO 3516:1997) that lists 65-78% linalool, -pinene (3-7%), -terpinene (2-7%), camphor (4-6%), limonene (2-5%), geranyl acetate (1-3.5%), geraniol (0.5-3%), -terpineol (0.5-1.5%) and myrcene (0.5-1.5%) as the main constituents of coriander fruit oil. Further constituents can be linally acetate, borneol, citronellol, nerol, terpinene-4-ol, among other monoterpene hydrocarbons and aliphatic aldehydes (Ziegler, 2007). When compared to coriander seed oil, coriander leaves oil contains a very low percentage of linalool (<1%) and -pinene (<0.1%) and a higher percentage of aldehydes and alcohols, accounting for about 56 and 36% of all oil constituents, respectively (Matasyoh et al., 2009), while typical seed oil constituents such as -terpinene, camphor, limonene, geraniol and myrcene were not detected in coriander leaves oil (Matasyoh et al., 2009).

1.2.2. Factors affecting coriander oil composition

Climatic conditions, geographic position of the growth region, agrotechnology of growing along with the vegetation state of plants at the time of harvest (Msaada et al., 2009; Msaada et al., 2007; Telci et al., 2006a) can influence both the qualitative composition and individual component contents in the isolated essential oil (Misharina, 2001). In terms of growth region

variations, it is observed that Indian regions produce coriander oils with lower amounts of linalool and higher ester contents when compared to European and Russian oils (Anwar et al., 2011). Regarding the stage of fruit maturity, it was seen that the percentage of monoterpene alcohols, particularly linalool, is higher in the final stage of maturation while monoterpene esters and aldehydes decrease throughout the maturation stages (Msaada et al., 2007).

Essential oil yield and quality can also be enhanced by the choice of fertilizers. Fertilizing techniques such as mycorrhization of coriander plants led to a 43% increase in essential oil concentration in fruits as compared to control fruits and the quality of the essential oil obtained with mycorrhizal plants was judged to be of superior quality (Kapoor et al., 2002).

Besides these pre-harvest factors, some post-harvest factors should also be considered such as the length and conditions of oil storage. Misharina in 2001 described that oil exposure to daylight, at room temperature, when compared to dark storage, caused many changes in its composition over a 12 month period (Misharina, 2001). For instance, some monoterpene hydrocarbons (-terpinene, -pinene, -myrcene, sabinene, -thujene and ocimene) disappeared completely, the levels of -pinene, limonene and linally acetate suffered a six-fold decrease whereas *p*-cimene, and 4-terpineol, camphor and linalool oxides increased up to 28 times the initial value (Misharina, 2001). On the other hand, when stored in the dark for a year, the oil composition was comparable to that of fresh oil samples (Misharina, 2001). These changes might be related with chemical modifications induced by light-initiated oxidative processes (Misharina, 2001).

1.2.3. Coriander oil toxicity

On of the utmost important aspects of natural products, especially those which are readily available to the consumers, is safety. Until recent years, people assumed that the consumption of natural products was harmless, but there are recent, abundant evidences that described severe adverse effects and even deaths associated with the use of dietary natural supplements and nutriments. Concerning coriander oil toxicity, there are only a few studies available on the oil itself or its main component, linalool, and the information on extensive studies conducted on in vitro or in vivo models is scarce (see (Burdock and Carabin, 2009) for a more detailed revision). The values for acute oral toxicity (LD₅₀) of coriander oil in rats ranged from 2.48 to 6.14 g/kg while the values for linalool, in Osborne-Mendel rats, were greater than 2.79 g/kg (Jenner et al., 1964). As coriander oil is mainly composed of linalool, accounting for up to 80% of the oils composition, linalool toxicity studies suggested that coriander oil toxicity can be attributed to linalool (Khani and Rahdari, 2012). When applied in an undiluted form, coriander oil is found to be irritating to the skin; however, when diluted (1% in lotion or 6% in petrolatum) it produced no irritation to the human skin (Burdock and Carabin, 2009; Casetti et al., 2012). Other studies performed with linalool corroborate the oils results, since diluted solutions of linalool (20% in petrolatum or Vaseline) did not produce irritation in the human skin (Burdock and Carabin, 2009). A short-term coriander oil consumption study described kidney, liver and stomach lesions in rats without any effects on survival, body weights or food consumption for dose levels superior to 160 mg/kg/day (Letizia et al., 2003). These adverse effects on the liver caused by coriander oil might also be related with the induction of peroxisomal enzymes in rat liver by linalool (Roffey et al., 1990). When studying developmental and reproductive coriander oil toxicity, it was found that only high doses of oil (500-1000 mg/kg/day) produce a decrease in

gestation and in the viability of pups (http://legacy.library.ucsf.edu/tid/rpd11e00/pdf;jsessionid=E475FB367C64F56F4703F2C1ADF0 6B5B.tobacco03). Both coriander oil and linalool did not present any immunotoxicity in mice. The vast majority of coriander genotoxicity assays are conducted with coriander extracts (Reyes et al., 2010) and not with the essential oil. One of the few studies performed with coriander oil on hamster fibroblasts revealed that the oil was not clastogenic (Ishidate et al., 1984). Regarding linalool genotoxicity, although some mutagenic effects have been described in the past (Burdock and Carabin, 2009), the most recent studies refer that this compound is devoid of mutageneicity (Di Sotto et al., 2008; Di Sotto et al., 2011). Coriander oil did not produce sensitization reactions, due to the fact that it probably does not contain the protein component responsible for the allergic reaction to coriander extracts (Burdock and Carabin, 2009). *In vitro* studies also proved that linalool has a significant citotoxic effect against HepG2 (human hepatocellular carcinoma) cells, with a 50-100% decrease in viability (Usta et al., 2009) and that coriander oil has a prooxidant activity on lipid peroxidation in the Fe²⁺/H₂O₂ system of induction (Samojlik et al., 2010). Coriander oil also showed some haemolytic activity against human red blood cells, causing a 46% hemolysis at a concentration of 3.2% (Duarte et al., 2013).

Overall, coriander oil and its major constituent, linalool, have low acute oral and dermal toxicity in laboratory animals and did not present other significant adverse effects both *in vitro* or *in vivo*, making it a safe ingredient to be used as a food ingredient or, perhaps, a food preservative, due to its remarkable broad antimicrobial activity.

1.2.4. Coriander oil antioxidant activity

The antioxidant activity of a compound can be shown by different assays, such as the ability to scavenge free radicals, the inhibition of lipid peroxidation or chelation of transition metals ions (Samojlik et al., 2010). The antioxidant activity of coriander oil is generally attributed to the presence of phenolics compounds (Samojlik et al., 2010), although some studies suggest that that the total antioxidant capacity of the oil was not directly related to the total polyphenolic compounds but seemed more dependent on the content of flavonoids and tannins (Sriti et al., 2011). Notwithstanding the good radical scavenging activity of coriander oil (Samojlik et al., 2010; Singh et al., 2006), some prooxidant activity on lipid peroxidation have also been described (Samojlik et al., 2010), meaning that this oil should be carefully used.

2. Antimicrobial activity of coriander essential oil

Antimicrobial susceptibility tests can be classified as diffusion, dilution or bioautographic methods (Burt, 2004). The CLSI methods for antibacterial susceptibility testing have been adopted by researchers, with slight modifications, to test antimicrobial and antifungal activity of essential oils (Duarte et al., 2012; Hammer et al., 1999; Soares et al., 2012). Due to the absence of a standardized protocol to test essential oil antimicrobial activity, the comparison between results from published works is complicated, since the outcome of the test can be affected by numerous factors (for a more detailed revision refer to (Burt, 2004)). When evaluating essential oil activity, the term minimum inhibitory concentration (MIC) is cited by most researchers as a measure of the antimicrobial performance of the oil (Delaquis et al., 2002; Duarte et al., 2012; Silva et al., 2011a; Silva et al., 2011b). Associated with MIC value determination, the authors sometimes refer also the minimum bactericidal or lethal concentrations (MBC or MLC) (Silva et al., 2011a; Silva et al., 2011b). So far, there is no consensus regarding the acceptable inhibition

level for natural compounds when compared to standard antimicrobials. For instance, Duarte and co-authors proposed several degrees of bioactivity for natural compounds based on the minimal inhibitory concentration (MIC) values: strong activity (up to 0.5 mg/mL), moderate activity (0.51-1.0 mg/mL) and weak activity (above 1.1 mg/mL) (Duarte et al., 2005). Coriander oil has shown a broad microbial activity against bacteria and fungi using diverse antimicrobial activity assays such as agar dilution, well or disc diffusion and broth macro or microdilution that will be summarized in the following sections.

2.1. Antibacterial activity

Coriandrum sativum essential oil proved to be effective against a wide range of foodborne and clinically relevant Gram-positive and Gram-negative bacteria using several assays such as disk diffusion, agar or broth dilution (Table 2). Using disk diffusion assays, the amount of coriander oil loaded onto the paper disks able to cause bacterial growth inhibition ranged from 24 mg to 0.09 µg. In these studies, a wide range of bacterial species were inhibited such as Shiga and non-Shiga toxin producing *E. coli*, several *Pseudomonas* species, *B. gladioli*, *X. campestris*, *B. megaterium*, *Y. enterocolitica*, *L. monocytogenes*, *S. typhimurium*, methicillin-sensitive *S. aureus* and *P. mirabilis*. The results also showed that coriander oil seemed more active against Grampositive than Gram-negative bacteria. For instance, Lo Cantore and collaborators referred that, although coriander oil was able to inhibit bacterial growth of Gram-positive and Gram-negative bacteria, an higher activity was verified against Gram-positive bacteria and some species of the Gram-negative bacterium *Xanthomonas* and lower activities were obtained for *Pseudomonas* spp. (Lo Cantore et al., 2004). Contradicting these results, other study showed that coriander oil was effective against Gram-negative bacteria but had no effect on the Gram-positive bacterium

L. plantarum (Elgayyar et al., 2001). Using both agar and broth dilution methods, is possible to assess the level of bacterial susceptibility through the determination of MIC values and even MBC values, using broth dilution methods. Due to the limited aqueous solubility of coriander oil, as a result of the high percentage of hydrophobic constituents, the oil is commonly solubilised in culture medium containing a small percentage of DMSO, Tween 20 or Tween 80 (Hammer et al., 1999; Rattanachaikunsopon and Phumkhachorn, 2010; Silva et al., 2011b). Overall, the results obtained by these two methods are very similar, with MIC values ranging from 0.03 to 1.6% (v/v). The results obtained by these assays corroborated some of the results obtained with disk diffusion assays, showing coriander oil effectiveness against, for example, C. methillin-sensitive methicillin-resistant S. vancomycin-resistant jejuni, and aureus. Enterococcus, K. pneumoniae, non-Shiga and Shiga toxin producing E. coli, A. baumannii, among other pathogens. Some of these studies proved that Gram-positive bacteria were more susceptible to coriander oil. Delaquis and co-authors demonstrated that coriander oil was effective against L monocytogenes and S. aureus but had no effect on P. fragi or S. typhimurium (Delaquis et al., 2002). On the other hand, other studies revealed a high activity of coriander oil against Gram-negative bacteria. For example, it was found that the Gram-negative bacterium C. jejuni was very susceptible to coriander oil (MIC values of 0.03-0.06%) (Rattanachaikunsopon and Phumkhachorn, 2010) and, also, that coriander oil was able to have a bactericidal effect on Gram-negative bacteria and not on Gram-positive bacteria, such as B. cereus and E. faecalis (Silva et al., 2011b).

It is widely accepted that the antimicrobial activity of essential oils depends on major constituents and their concentrations. The inhibitory effects of essential oils are mainly due to

their major components but the small amounts of minor components might also contribute to the antimicrobial activity. It has been described that different compound classes have different antimicrobial activities: phenolic compounds possess the highest antimicrobial activities followed by alcohols, aldehydes, ketones, ethers and hydrocarbons (Ferdes and Ungureanu, 2012). The presence of the hydroxyl (-OH) group on phenolic compounds is thought to be responsible by these compounds antimicrobial activity (Ferdes and Ungureanu, 2012). Some of coriander oil constituents such as linalool, -pinene, p-cymene, -terpinene, limonene and linalyl acetate proved to be effective against several Gram-positive and Gram-negative bacteria (Cristani et al., 2007; Di Pasqua et al., 2006; Ozek et al., 2010; Sonboli et al., 2006; Trombetta et al., 2005). However, the susceptibility levels obtained for these compounds seemed to be slightly higher than those obtained for the essential oil, meaning that the oils antimicrobial activity is more than a õsum of its partsö probably due to complex interactions between all individual components.

2.2.Antifungal activity

Some studies showed that coriander essential oil had antifungal activity against several types of fungi, such as yeasts, dermatophytes and filamentous fungi using agar and broth dilution as well as disk diffusion assays (Table 3). Although some of the studies have focused on the oil antifungal activity against *Candida* species (Furletti et al., 2011; Hammer et al., 1999; Silva et al., 2011a), probably due to its clinical relevance, the oil also inhibited the growth of other fungi such as *M. canis*, *A. niger*, *S. cerevisiae*, *G. candidum* and *K. fragilis* (Delaquis et al., 2002; Elgayyar et al., 2001; Soares et al., 2012; Toroglu, 2011). Coriander oil was able to inhibit *Candida* spp. growth at concentrations ranging from 0.0008% to 0.4%, *S. cerevisiae* at a

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concentration of 0.13%, *M canis* at concentrations of 78-620 µg/mL (0.009-0.07%) and the other fungi were inhibited at 1.74 or 24 mg of coriander oil per disk. Some of these studies also evaluated the fungicidal activity of coriander oil yielding MLC values ranging from 0.017 to 0.4% (Silva et al., 2011a; Soares et al., 2012).

2.3. Other antimicrobial activities

So many years of increasing applications of antimicrobial agents have created a situation leading to the rise on the number of multiple-antibiotic-resistant bacteria and fungi (Teuber, 1999). To fight these new õsuperbugsö, researchers have focused their attention on the possible combination of natural compounds with common antimicrobials, expecting that the natural compound could potentiate the antibiotic activity or even reverse the resistance to the antibiotic. In fact, some researchers have evaluated the potential synergism or additive effect of coriander oil against common antibiotics and antifungal drugs. Despite some results stating that coriander oil had an antagonistic effect with some antibiotics against both Gram-positive and Gramnegative bacteria (Toroglu, 2011), others revealed that coriander oil had synergetic effects with gentamicin and ceftriaxone for *S. aureus*, with ceftriaxone for *M. smegmatis* (Toroglu, 2011) and with chloramphenicol, ciprofloxacin, gentamicin, tetracycline against *A. baumannii* (Duarte et al., 2012). The oil@s combination with piperacillin and cefoperazone or gentamicin yielded only an additive effect against *A. baumannii* (Duarte et al., 2012) or *S. aureus* (Toroglu, 2011), respectively.

It is known that the organization of microbial cells in biofilms increases their resistance to antimicrobials and disinfectants (Duarte et al., 2013), probably as a consequence of the delay in antimicrobials diffusion through the biofilms matrix, of the altered growth rate of biofilms cells

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or other physiological modifications in cells while growing in a biofilm (Donlan and Costerton, 2002). Most studies on the antimicrobial activity of coriander oil have focused on planktonic cells; notwithstanding, the ability of microorganism to form biofilms points out the need to evaluate the oils anti-biofilm activity. A study evaluating the action of coriander oil on biofilms formation by Candida albicans showed a clear effect of the oil on the formation of biofilms, characterized by an increased lag phase and a decrease in biofilms growth at a concentration of 0.125 mg/mL (Furletti et al., 2011). Another study evaluated coriander oil ability to inhibit the formation or eradication of Acinetobacter baumannii biofilms (Duarte et al., 2013). The authors concluded that the oil was able to inhibit biofilm formation at concentrations ranging from two to four times the MIC value (0.2-1.6%), causing an 85% reduction on total biofilms biomass and metabolic activity. When applied to pre-formed biofilms, the same oil concentrations were able to eradicate pre-formed biofilms, leading to a 75-90% reduction on total biomass and metabolic activity after 24h of incubation (Duarte et al., 2013). These results encourage the use of coriander oil as an anti-biofilm compound, since, typically, antibiotics are about 1000 times less effective against cells in biofilms than planktonic cells (Melchior et al., 2006).

Another important feature of essential oils is their volatility but there is a general lack of scientific information about the effectiveness of essential oils in the vapour phase. The evaluation of the antimicrobial activity of essential oils in the vapour phase has been gaining interest in the last years. Studies by Lopez and co-authors revealed that, in general, essential oils and some oils constituents are less effective in the vapour phase; for example, camphor, 1,8-cineole, *p*-cymene and limonene were not effective against common foodborne pathogens while linalool showed a slight growth inhibition of *S. choleraesuis* and *C. albicans* (Lopez et al., 2005;

Lopez et al., 2007b). This is due to the fact that lipophillic molecules in the aqueous phase associate to form micelles, suppressing the attachment of essential oilsø components to the microorganisms, whereas the vapour phase allows free attachment, thus increasing the oilsø effectiveness (Martinez-Abad et al., 2013).

2.4. Mode of action

Despite the large number of studies addressing the antibacterial activity of essential oils against microorganism, the mechanisms involved are still not fully understood. Unveiling the mode of action of essential oils on bacterial and fungal cells is of great relevance towards their successful application in the pharmaceutical and food industries.

Coriander oil acts in a similar way against Gram-positive and Gram-negative bacteria: its action seems to be bactericidal as a consequence of membrane permeabilization and subsequent loss of all cellular functions such as metabolic activity, membrane polarization and efflux activity within 30 min of incubation (Silva et al., 2011b). Although the global mechanism of action was the same, due to the differential bacterial susceptibility results obtained by Silva et al., it was suggested that coriander oil might have a distinct action on Gram-positive and Gram-negative bacteria (Figure 3) taking into account the differences on the cellular envelopes of the two bacterial types and the oil composition (Silva et al., 2011b). The authors proposed that, at low oil concentrations, in Gram-positive bacteria, the oil caused a thickening of the cell wall, due to the action of the alcohols (Linhova et al., 2010) (mainly linalool) present in the oil (Silva et al., 2011b). It was also suggested that, in Gram-negative bacteria, (+)-linalool, in a similar manner to (+)-menthol (Casabianca et al., 1998; Trombetta et al., 2005), was able to establish a strong interaction with the outer membrane since it presents a strong negative charge conferred by

lipopolysaccharides, thus disrupting this structure and enabling a facilitated access of the oil to the intracellular space (Silva et al., 2011b). This oiles action on the bacterial membrane is also corroborated by other studies that revealed that some oil constituents such as limonene, linally acetate, p-cymene and -terpinene were able to elicit homeoviscous adaptation (Di Pasqua et al., 2006) in bacteria due to these compounds ability to interact with the phospholipids of Grampositive and Grampositive bacteria membranes (Cristani et al., 2007; Di Pasqua et al., 2006; Trombetta et al., 2005), thus perturbing the lipid fraction of the plasma membranes which could result in increased permeability to test compounds.

Regarding the antifungal mechanism of action, coriander oil inhibits germ tube formation on yeasts such as *C. albicans*, even at concentrations bellow the MIC value (Figure 3) (Silva et al., 2011a). Similarly to what was verified by Silva and co-authors for bacteria, coriander essential oil seems to target the fungal membrane (Figure 3), since, after 30 min of contact, membrane permeabilization can be seen in all fungal cells, causing the leakage of large intracellular components such as DNA (Silva et al., 2011a).

3. Essential oils in food preservation

The use of essential oils as food preservatives is not controversial since they are plant-based materials and have long been used in culinary and folk medicine throughout the world. However, the possibility of applying the oils in an industrial scale for food preservation still remains unclear. Their applicability is determined by two key factors: their effectiveness in terms of product and the acceptance level of the consumers to the modified product. In addition, consumersø approval is based on the sensory qualities of the end-product and the fact that no sensitization or allergic reaction is induced by its consumption (Michalczyk et al., 2012).

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It is well known that the antimicrobial efficacy of essential oils is greatly reduced in food systems when compared to in vitro work, as the presence of fats, carbohydrates, proteins, salts and pH strongly influence the activity of the oils (Burt, 2004). Also, in a complex biological system such as food various interactions may occur between the additives used, food constituents and the food matrix (Michalczyk et al., 2012). Additionally, food is generally colonized by many different species of bacteria and their interactions should be considered, since they can be mutually antagonistic or even synergetic (Gram et al., 2002). Therefore, the disturbance of this equilibrium by the addition of a food preservative must be taken into account. As a result, larger amounts of essential oils are required to be added in food systems for attaining the same antimicrobial properties, thus causing major shifts in the organoleptical properties of the food item (Busatta et al., 2008), exceeding the sensorially acceptable level (Michalczyk et al., 2012). The direct incorporation of essential oils in foods was one of the early approaches for food preservation using these compounds. Several essential oils such as the ones obtained from sage, oregano, marjoram and thyme were incorporated in meat and fish products to improve these foodsø shelf life (Lucera et al., 2012). These studies proved that the addition of essential oils to foods, alone or in combination with modified atmosphere packaging (MAP) (Sellamuthu et al., 2013) were able to improve the sensory shelf-life of the products due to a reduction in microbial growth, thus exerting a bacteriostatic effect (Lucera et al., 2012).

With the new breakthroughs in the packaging industry, with the development of active and intelligent packaging systems, it was though that the incorporation of the essential oil in a plastic or biopolymer (polysaccharide-based, protein-based, lipid-based or composites) film could overcome the addition of such high amounts of essential oils to foods, as a result of the gradual

release of the oil from the film onto the food surface (Cran et al., 2010; Zivanovic et al., 2005). As a result of their properties such as their low cost, good processability, mechanical and physical features (Kuorwel et al., 2011), plastic films have been described for the development of antimicrobial films with essential oils, proving to be effective in the improvement of the shelflife of several foods such as meat, salads (Muriel-Galet et al., 2013). Nowadays, biopolymerbased films are being preferred over plastic films since they can be edible, are biodegradable and some have intrinsic antimicrobial properties as is verified for chitosan films (Cha and Chinnan, 2004; Zivanovic et al., 2005). Another relevant type of films are the ones involved in paper packaging, since many food and drink packages are made of several papers and boards (Rodriguez et al., 2007). Several studies have successfully incorporated clove, cinnamon and oregano essential oils in paper packaging materials for the inhibition of fungal growth in fresh fruits (Rodriguez-Lafuente et al., 2010; Rodriguez et al., 2007). The data obtained proved that paper packages containing cinnamon oil were very effective against several fungi such as C. albicans, A. flavus, Rhizopus stolonifer and Alternaria alternata (Rodriguez-Lafuente et al., 2010; Rodriguez et al., 2007; Rodriguez et al., 2008) showing a bacteriostatic action as no complete growth inhibition was achieved.

The use of films based on antimicrobial polymers can further reduce the amount of essential oil incorporated while still being effective as preservation method (Wang et al., 2011). One of these strategies is based on the synergistic effect observed between some films, such as chitosan films and essential oils (Khanjari et al., 2013) and between two or more combined essential oils (Goni et al., 2009), allowing a reduction in the amount of incorporated essential oil (Wang et al., 2011). Additionally, the oil quantity incorporate in the packaging material can be reduced if we consider

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the vapour-phase activity of the oil, a parameter greatly influenced by the oil polarity (Licciardello et al., 2013), since the MIC concentrations in the vapour phase can be 30-100 times lower than in the liquid phase (Martinez-Abad et al., 2013). Due to the vast array of film materials available for the development of antimicrobial packages, when designing a new active packaging, one should investigate the kinetics of the oil release from the film, as the antimicrobial activity of the film might depend on its ability to release the oil (Gutierrez et al., 2010; Mercier et al., 2002).

Overall, when analysing the data available, the antimicrobial activity of active films containing essential oils, seems to be similar to the one obtained *in vitro*: antimicrobial films containing essential oils are, in general, very effective against yeasts and molds and more active against Gram-positive bacteria (Ghasemlou et al., 2013; Lopez et al., 2007a; Martinez-Abad et al., 2013) than Gram-negative bacteria, with *P. aeruginosa* being one of the most resistant bacterium to essential oils (Lopez et al., 2007a).

3.1. Coriander oil in food preservation

Coriander oil is extensively used as a food additive or adjuvant in all sorts of foods, such as alcoholic beverages, tobacco, candy, pickles, dairy products, chewing gum, meat sausage and pickles with use levels ranging from 0.1 to 100 ppm. The in vitro effectiveness of coriander oil against numerous foodborne pathogens such as *S. aureus*, *C. jejuni*, Shiga and non-Shiga toxin producing *E. coli*, *L. monocytogenes*, *Y. enterocolitica* and *S. thypimurium*, conducted to the exploitation of this oil and major constituent, linalool, applicability in food preservation. The efficacy of coriander oil and linalool in inhibiting microbial growth in foods was tested through direct addition and incorporation into films (Table 4). The direct addition of coriander oil (0.02%)

v/w) to minced beef caused a reduction in *Enterobacteriaceae* counts and was able to inhibit undesirable sensory changes due to meat spoilage, although myoglobin oxidation was not prevented (Michalczyk et al., 2012). In another study, the direct addition of coriander oil to ground chicken meat and beef (0.5% v/w) resulted in complete *C. jejuni* cell death after 30 min of contact time, whilst 2 and 4-log reduction on bacterial loads were obtained for lower oil concentrations (0.1 and 0.25%) (Rattanachaikunsopon and Phumkhachorn, 2010). Although these two strategies were successful, when coriander oil was incorporated into a chitosan film, its microbial efficacy against *L. monocytogenes* and Shiga-toxin producing *E. coli* was limited and lower than the one obtained with other essential oils, as the inhibitory effects of essential oil incorporated into the chitosan film were lower than the ones of the pure oil (Zivanovic et al., 2005).

So far, the only strategy described for the use of linalool in food preservation was based on the development of low-density polystyrene (LPDE) films containing this compound for the reduction of microbial contamination in Cheddar cheese (Suppakul et al., 2008). It was observed that linalool (0.34% w/w) LPDE films were able to cause a significant reduction in total aerobic bacteria, thus reducing natural microbial contamination in cheese samples (Suppakul et al., 2008). The films were also able to reduce microbial counts in artificially contaminated cheese with *L. innocua* or *E. coli*. In fact, even after long-term storage (1 year), these films continued to inhibit E. coli growth (Suppakul et al., 2011). Furthermore, sensory analysis of these films revealed that linalool, at the percentage used, may not present a problem in the alteration of the sensory properties of the food item (Suppakul et al., 2008).

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Although food deterioration can be caused by microbiological contamination, there is a significant part of this process that results from chemical alterations in the food product. Among chemical processes, one of the most relevant is oxidative deterioration by the degradation of fats and pigments. Therefore, the antioxidant activity of essential oils, together with their antimicrobial activity, plays a key role on preventing food spoilage and improving the shelf-life of foods (Bentayeb et al., 2007; Lopez de Dicastillo et al., 2011; Nerin et al., 2006). Due to its described antioxidant activity, coriander oil was tested as an antioxidant in food products in order to increase their oxidative stability. Coriander oil was effectively used as an antioxidant for the preservation of Italian salami, being able to reduce lipid oxidation to a further extent than a synthetic antioxidant, hence improving the shelf-life of the product with no significant alterations on the sensory profile (Marangoni and de Moura, 2011a; Marangoni and de Moura, 2011b). In another study, coriander oil was used to prevent the deterioration of ghee: although its antioxidant activity was not so effective as the one of the synthetic antioxidant during storage; during frying, the addition of coriander oil proved to result in the highest antioxidant activity (Patel et al., 2013).

Conclusions and future trends

An attractive application of essential oils and their constituents is in foods to prolong their shelf-life by controlling growth and survival of microorganisms. The organoleptic impact of essential oils and their components in food products is still currently restraining their application to foods. Synergistic interactions of essential oils with other preservatives, their incorporation into antimicrobial packaging films and their combined use with other preservation technologies such

as irradiation or modified atmosphere have been proposed to reduce the sensory alterations caused by essential oils.

Among all the essential oils with described antimicrobial properties, several coriander oil features make it a an attractive and valuable choice for the development of natural-based food preservation techniques such as its antioxidant activity, broad antimicrobial effectiveness and bactericidal activity within 30 min of contact against several foodborne pathogens and its safety as a food ingredient. Also, the numerous biological properties of coriander oil can also be advantageous to consumers to promote their health, thus adding value to the food item. Although coriander oil is the second most used essential oil worldwide, there is still a need for more research and clinical studies to evaluate the safety of its consumption and prove its biological activities in vivo. Also, for the use as food preservative, its efficacy against common foodborne fungi, such as Fusarium spp., Aspergillus ochraceus, Penicillium verrucosum, Aspergillus flavus and Aspergillus parasiticus and vapour-phase antimicrobial activities should also be evaluated. Overall, the research on coriander oil application or linalool as food preservatives is still scarce, yielding very different outcomes in terms of success. So far, only the direct addition of coriander oil to meat, its incorporation in a chitosan film and the incorporation of linalool in a LPDE film were described. Due to the vast array of technologies for food packaging and preservation made available over recent years, the possibility of using coriander oil as food preservative still needs extensive investigation in order to improve its efficacy in food media and reduce the undesirable alterations in the organoleptic properties of foods.

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Table 1: Variations of some oil components of *Coriandrum sativum* according to different locations.

	Country	Iran	Pakistan	Russia	Georgia	Tunisia	Canada
	Oil yield (%)	0.36	0.15	na	na	0.37%	0.44
	alpha- pinene	3.3	1.63	4.81	4.66	3.4	4.5
	alpha- thujene	nd	0.02	0.07	0.18	0.3	0.5
	beta-pinene	0.4	0.23	0.62	0.48	t (<0.1)	t (<0.1)
	borneol	nd	0.18	na	na	0.6	0.1
	camphene	na	0.02	0.63	0.92	0.3	0.5
	camphor	0.2	0.38	2.96	4.45	2.9	6.4
	gamma-	9.3	4.17	6.1	6.81	8.4	8.9
	terpinene						
Oil	geranyl	na	4.99	1.03	2.55	1.8	2
compos	acetate						
ition	limonene	0.3	0.26	2.51	3.7	1.4	2.6
	linalool	70.1	69.6	69.75	68	71.6	57
	linalool oxide	na	na	1	1.48	na	na
	linalyl acetate	na	na	1.25	1.76	1.9	7.1
	myrcene	0.2	0.18	0.8	1.23	0.6	1.3
	p-cymene	2	1.12	4	2.48	1.4	2.1
	sabinene	0.2	0.12	0.22	0.3	0.1	0.1
	terpinen-4- ol	0.1	0.39	0.04	0.16	0.3	0.5
nap.	OI ndo	(Ebrahim i et al., 2010)	(Anwar et al., 2011)	(Misharina,	, 2001)	(Msaada e	t al., 2009)

naData not available, ndCompound not detected

Table 2: Literature review on coriander oil antibacterial activity.

Strain	Oil concentration	Method	Reference	
Escherichia coli				
Pseudomonas syringae		Disk diffusion		
Pseudomonas cichorii				
Pseudomonas corrugata				
Pseudomonas agarici				
Erwinia carotovora			(Lo Cantore et al., 2004)	
Agrobacterium	217.5-6960 µg/disk			
tumefaciens				
Burkholderia gladioli	MS, GISH			
Xanthomonas campestris				
Bacillus megaterium				
Clavibacter michiganensis				
Curtobacterium				
flaccunfaciens				
Rhodococcus fascians				
Streptococcus pyogenes				
Streptococcus viridans				
Methicillin-sensitive				
Staphylococcus aureus			(Casetti et al., 2012)	
Methicillin-resistant	0.04-0.5 % (v/v) Agar dilution			
Staphylococcus aureus		Agar dilution		
Enterococcus faecalis				
Enterococcus faecium				
Vancomycin-resistant				
Enterococcus				
Escherichia coli				
Klebsiella pneumoniae				
Escherichia coli O157:H7			(Delaquis et al.,	
Listeria monocytogenes	0.23-0.4%	Broth dilution	2002)	
Staphylococcus aureus			,	
Acinetobacter baumannii			(Hammer et al., 1999)	
Aeromonas sobria				
Enterococcus faecalis		Agar dilution		
Escherichia coli	0.25-1%			
Klebsiella pneumoniae				
Pseudomonas aeruginosa				
Salmonella typhimurium				

Serratia marcescens			
Staphylococcus aureus			
Staphylococcus aureus			
Escherichia coli O157:H7			
Yersinia enterocolitica	24 mg/digle	Disk diffusion	(Elgayyar et al.,
Listeria monocytogenes	24 mg/disk	DISK UIITUSIOII	2001)
Salmonella typhimurium			
Pseudomonas aeruginosa			

Table 2 (cont.)

Strain	Oil concentration	Method	Reference	
Campylobacter jejuni	0.03-0.06%	Broth dilution	(Rattanachaikunso pon and Phumkhachorn, 2010)	
Escherichia coli				
Pseudomonas pyocyaneus				
Yersinia enterocolitica	2 μL/disk	Disk diffusion	(Toroglu, 2011)	
Bacillus megaterium				
Streptococcus faecalis				
Corynebacterium diphteriae				
Staphylococcus aureus			(Singh et al., 2002)	
Streptococcus		Disk diffusion		
haemolyticus	0.09 and 4.5 μg/disk			
Bacillus subtilis				
Pseudomonas aeruginosa				
Escherichia coli				
Klebsiella spp.				
Proteus vulgaris				
Acinetobacter baumannii	0.1-0.4%	Broth dilution	(Duarte et al., 2012)	
Bacillus cereus				
Enterococcus faecalis	0.1-1.6%	Broth dilution	(Silva et al., 2011b)	
Staphylococcus aureus				
Methicillin-resistant				
Staphylococcus aureus				
Pseudomonas aeruginosa				
Klebsiella pneumoniae				

Escherichia coli
Salmonella typhimurium
Acinetobacter baumannii

 Table 3: Literature review on coriander oil antifungal activity.

Strain	Oil concentration	Method	Reference
Saccharomyces cerevisiae	0.13%	Broth dilution	(Delaquis et al., 2002)
Candida albicans	0.25%	Agar dilution	(Hammer et al., 1999)
Aspergillus niger Geotrichum candidum Rhodotorula	24 mg/disk	Disk diffusion	(Elgayyar et al., 2001)
Saccharomyces cerevisiae Klyveromyces fragiliis	2 μL/disk	Disk diffusion	(Toroglu, 2011)
Microsporum canis Candida spp.	78-620 μg/mL	Broth dilution	(Soares et al., 2012)
Candida albicans Candida krusei Candida parapsilosis Candida dubliniensis Candida tropicalis	0.007-0.5 mg/mL	Broth dilution	(Furletti et al., 2011)
Candida albicans Candida tropicalis	0.05-0.4%	Broth dilution	(Silva et al., 2011a)

Table 4: Effect of coriander oil antimicrobial efficacy in foods.

Type of Food / Microorganis m	Compoun d	Delivery mode	Main findings	References
Minced beaf	Coriander oil	Direct addition	Inhibition of undesirable spoilage sensory changes Inhibition of Enterobacteriaceae growth No changes in myoglobin oxidation	(Michalczyk et al., 2012)
L. monocytogenes E. coli O157:H7	Coriander oil	Incorporation into a chitosan film	Low microbial efficacy Lower microbial efficacy of essential oil after incorporation into the film	(Zivanovic et al., 2005)
Ground chicken Ground beef (C. jejuni)	Coriander oil	Direct addition	Complete bacterial death at a concentration of 0.5% v/w after 30 min	(Rattanachaikuns opon and Phumkhachorn, 2010)
Cheese	Linalool	Incorporation into a LPDE film	Significant reduction in total aerobic bacteria Maintenance of cheese sensory properties E. coli growth inhibition after long-term storage	(Suppakul et al., 2008; Suppakul et al., 2011)

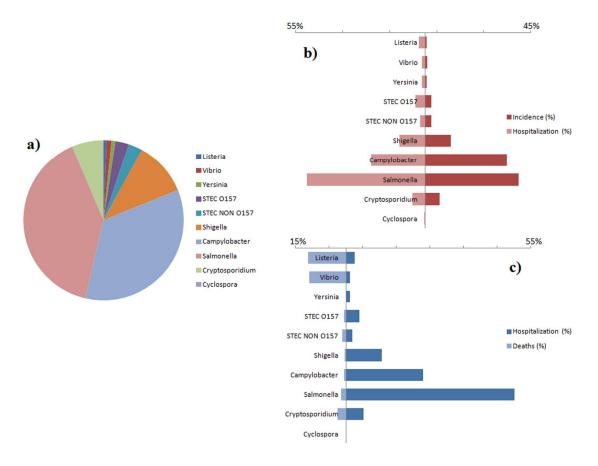


Figure 1: 2012 data on foodborne infections in the USA: a) number of cases of bacterial and parasitic infection; b) percentage of incidence and hospitalizations by pathogen; c) percentage of hospitalizations and deaths in hospitalized patients by pathogen. Foodborne Diseases Active Surveillance Network, United States, 2012 (2013)

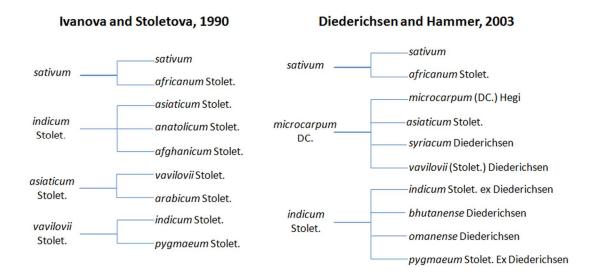


Figure 2: *Coriandrum sativum* infraspecific data according to Ivanova and Stoletova (1990) and Diederichsen and Hammer (2003).

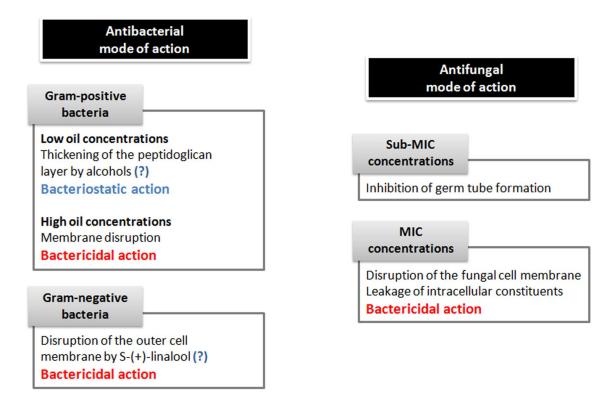


Figure 3: Coriander oil mechanism of action on bacterial and fungal cells.