

Methods of assessing erosion and erosive potential

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Methods for use *in vitro* are reviewed first, classified under the examination of extracted teeth, chemical and physical methods, digital image analysis, scanning electron microscopy and other potentially useful techniques. Methods for recording erosion in laboratory animals are dealt with next, including techniques that can also be used *in vitro*. Restarski's system and various modifications of it, and digital image analysis. Thirdly the main systems and indices that have been designed for clinical use are arranged under case histories and clinical examination, scales of severity and indices of diagnosis of varying complexity. Finally, some of the main advantages and limitations of the techniques are summarized.

Key words: animal disease models; assessment; clinical markers; indices; techniques

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It must be borne in mind that dental erosion is a clinical manifestation of attack on the teeth by agents in foods and drinks, from regurgitation, or in rare cases from environmental causes. This means that most attention on devising methods of assessment has been focused on scoring methods and indices etc. for use *in vivo*. In the limited amount of research *in vitro* that has been carried out on the process of erosion, there has been a tendency to use methods of assessment originally developed for clinical purposes, and this is reflected in the balance of the references reviewed below, which are based mainly on clinical methods of evaluation, rather than on the needs of research *in vitro*.

Thus what is assessed on extracted teeth or in dental hard tissues used in research may be defined as erosive potential, whereas teeth *in situ* in the mouth are examined for erosion. The methods used for measurement and to standardize the techniques under these two sets of conditions will vary and can be divided into three main groups, which will be considered separately. A major object in devising and adopting a method is of course to ensure reproducibility, so that measurements made at different times and under differing sets of circumstances are comparable.

Two main questions need to be answered:

A. What loss of tooth tissue has already taken place? i.e. What change in morphology can be measured?

B. What loss is likely under prevailing circumstances?

The three chief types of methods are

1. Methods for use *in vitro*.
2. Those suitable for recording erosion in laboratory animals used in research.
3. Techniques for the assessment of erosion in man.

Methods for use *in vitro*

Examination of extracted teeth. If whole human teeth have to be examined for erosion, the methods could be based on those used *in vivo* (see below). A point of difference would be that whereas *in vivo* a part or whole of the entire dentition may be studied and assessed for erosion, observations after extraction may have to be based on single teeth or small groups of teeth. This means that the method of scoring would have to be modified, but care would have to be taken to standardize it so as to achieve comparability. One way of approaching this would be by adhering to set criteria and using the same type of teeth each time (incisors, canines, premolars or molars), obtained from individuals comparable in as many respects as possible.

An additional drawback to using extracted teeth is that their history and the causative erosive forces are unlikely to be known.

Chemical methods. Much of the research on the acidity of drinks and their potential erosiveness

has not used dental hard tissues at all, and has been based solely on measurements of oral or dental plaque pH (1, 2).

A sensitive way of showing small changes indicative of erosion under laboratory conditions is to measure calcium (Ca) and/or phosphorus (P) dissolving from teeth or dental mineral under the influence of potentially erosive acids or other erosive factors. A standard test has to be devised, e.g. exposing a tooth or tooth material to the erosive agent or environment for a set length of time, then taking the erosive medium for analysis (see for example 3 & 4). Initial and final figures are required so that 'blanks' (initial concentrations of Ca and P before any dissolves from the tooth mineral matter) can be subtracted from the final Ca and P levels.

A variety of small-scale and micro-methods exist for the determination of Ca and P in solution. A reliable and sensitive technique for estimating calcium with little interference from other solutes is by atomic absorption spectroscopy (3, 5). P entering solution can be measured by a number of absorptiometric techniques, such as the method of ALLEN (6), in which the intensity of the blue colour of a reasonably stable reduced phosphomolybdate complex is proportional to the concentration of P.

The decalcification of embedded bovine incisors was measured by Ca analysis by atomic absorption spectroscopy by McDONALD & STOOKEY (7), but this technique was subsidiary to their work on recording erosion in rats *in vivo* (see below).

Physical methods. These depend on direct examination and assessment of the condition of a tooth surface or tooth material and need a scale to be set for the purpose of quantifying erosion.

SMITH & SHAW (8) used 4×4 mm windows on extracted primary teeth, which, after exposure to fruit juices, were examined macroscopically before the preparation of 100-µm non-demineralized sections. These were mounted and viewed under polarized light to reveal destruction so that, for instance, what was observed was a complete loss of surface enamel with an irregular lesion surface. Beneath this was a zone with a translucent appearance in polarized light.

Areas of bovine tooth specimens (1×3 mm) were examined by MEURMAN *et al* (5) using three different techniques. Loss of tooth material was measured by profilometry, driving a spherical diamond tip across the surface of the specimen using a load of 5 mg. The analog reading representing the vertical movements of the tip was stored in a microprocessor memory. The recorded profiles were then used to analyse the surface alterations. Surface hardness was also measured by microhardness diamond indentation equipment. The third method of

examination was by scanning electron microscopy (see below).

Surface microhardness was used by LUSSI *et al.* (9) and in later studies to distinguish between erosive factors, using a Knoop diamond under standardized conditions and measuring the length of the indentations.

Indentation techniques have also been used by VAN MEERBECK *et al.* (10), and attention has been drawn to acidic and abrasive mechanisms in the development of cervical lesions (11).

Iodide permeability. As it was already known that the iodide permeability test can detect very early stages of enamel demineralization, it was evaluated as a method for measuring erosion (9) and compared with the SMH (surface microhardness) technique. No clear verdict was given on which of these was superior, leaving the impression that they may be complementary rather than competing with each other.

Digital Image Analysis. Various techniques that had applications both *in vivo* and *in vitro* were developed and published between 1940 and 1990, but with the increasing sensitivity and versatility of computer-aided evaluation and recording equipment, digital image analysis has been found to improve accuracy and reliability, and to reduce time and labour requirements (12). The surface to be examined is set in a fixed position and imaged by a video camera. A feature such as a ridge or other limit of eroded tissue is chosen as a 'threshold' between different zones of attack that can be discerned, and its position is traced and stored, after which the area and depth of the damage to the tooth enamel can be calculated, and statistical comparisons between different treatments can be performed.

Scanning electron microscopy (SEM). A detailed description of the use of SEM is given by ADDY *et al.* (13). Extracted teeth were cleaned in hypochlorite and 70% ethanol, then exposed to various acids and drinks for 5 min. Etching was assessed by sputter-coating the dried specimens with gold, examining them under the electron microscope, and grading the severity of etching on a scale, with 0=no tubules visible to +++=many tubules seen.

A similar method was used by MEURMAN *et al.* (5), with microphotographs taken at magnifications of ×400 and ×2000.

A replica impression technique of 'windows' on extracted teeth, examining castings by SEM after gold sputtering, has been reported recently (14). The model is said to mimic conditions at the tooth surface *in vivo* and to be highly reproducible.

Use of synthetic hydroxyapatite powders/discs. To help overcome problems arising from the variability of extracted teeth and enamel samples of

diverse provenance, the erosive potential of soft drink formulations can be determined using an enamel-like disc composed of ceramic (sintered) hydroxyapatite, measuring weight loss gravimetrically before and after immersion (15).

Other methods. A number of other innovative and sophisticated techniques, that should be capable of measuring erosive potential have been suggested, including 3-D tomography, laser scanning, nuclear magnetic resonance (NMR) and microradiography (e.g. 16), but no literature references on their application in erosion research have been found. Another technique that has been cited as potentially useful in erosion research is the intra-oral cariogenicity test (ICT) (17), in which slabs of enamel are mounted in the human mouth in intra-oral appliances and subjected to cariogenic or erosive conditions, after which demineralization can be recorded by microradiography and changes in surface microhardness.

The main methods of erosive potential assessment *in vitro* and their pros and cons are summarized in Table 1.

Methods for recording erosion in laboratory animals

Based on techniques that can also be used in vitro. Some of the methods reviewed above also have applications when laboratory animals are used in research. For example, the studies on digital image analysis of tooth surfaces were carried out in laboratory rats given various soft drinks and base

materials as drinking fluids (12), subdividing the areas of attack into eroded enamel and exposed dentine, and showing a much improved sensitivity over earlier subjective methods of measurement.

The techniques of SEM can also be employed on specimens obtained from experiments in laboratory animals.

Examination of selected teeth under magnification. A semi-quantitative method for rats' teeth was worked out (18), storing the jaws in ethanol and examining the lingual surface of the first mandibular molar under a magnification of $\times 26$ after staining with Schiff's reagent. Drawings were made, including eroded enamel between the intact area and either the surface border or exposed dentine of the teeth. Eroded enamel was seen when a clear border or slight ridge formation was visible towards the intact area. A detailed description of other features of tooth wear in rats, with pictures, was given. Copies of drawings up to a magnification of $\times 33$ were made, and a computer-coupled graphic analyzer was used to measure areas of intact and eroded surfaces in mm^2 for statistical analysis of the differences between experimental groups.

The grading system of RESTARSKI et al. This was described in a number of papers published in 1945 (19, 20). The heads of rats were autoclaved and the jaws were preserved in 80% ethanol/10% formaldehyde. The molars were examined under a dissecting microscope and graded by an arbitrary system:

0 no effect

1 high polish on lingual enamel

Table 1

Summary of methods for use in vitro – pros and cons.

Method	References	Advantages	Drawbacks
Examination of extracted teeth			Not based on a complete dentition. History of the teeth not known Qualitative.
Chemical methods	1–3, 5, 7	Very sensitive. Easy to study variables, e.g. time and type of acid.	Do not exactly mirror conditions <i>in vivo</i> . Quantitative only.
Physical methods	5, 8, 9–11	Surface hardness data may be relevant to erosion.	Hardness data do not measure extent of hard tissue loss by erosive attack. Qualitative.
Iodide permeability	9	Useful for both erosion and caries.	May use bovine rather than human enamel. Qualitative.
Digital image analysis	12	Very precise and sensitive. Both qualitative and quantitative.	Equipment and programs expensive.
Scanning electron microscopy	5, 13, 14	Mimics the tooth surface. Reproducible. Both qualitative and quantitative.	Very expensive equipment. Skilled labour requirement.
Hydroxyapatite powders/discs	15	Reproducible. Qualitative and quantitative.	Moving away from tooth tissue.
3-D tomography, laser scanning, NMR, microradiography, ICT test	16	All may have potential.	Unproven for erosion studies.

- 2 slight etching of lingual enamel
- 3 mild destruction of lingual enamel; evidence of slight ridge formation at the gingival margin
- 4 moderate destruction of lingual enamel; ridging more definite; some exposure of dentin
- 5 severe destruction of lingual enamel; marked ridge at gingival margin; moderate exposure of dentin
- 6 almost complete destruction of lingual enamel with definite evidence of destruction on other surfaces; marked exposure of dentin with some destruction.

The grades for each half-jaw were averaged and then added to give a total score per rat. A major advantage with this scoring system, according to the authors, was that the results were reproducible. It was therefore employed later in other studies, and has remained in use (see for example 21–23). Other workers adopted methods very similar to it (e.g. 24).

Detailed 'subjective' and 'objective' methods. Two techniques for evaluating erosion in the rat were put forward by HARTLES & WAGG (24). The subjective method made use of a low-power binocular microscope to classify damage to the teeth:

- 0 normal tooth
- 1 lingual enamel highly polished; slight loss of contour of lingual cusps
- 2 slight etching of lingual enamel at gingival margin of exposed crown; slight rounding of occlusal edge of cusps
- 3 beginning of ridge or shelf at gingival margin on the lingual enamel; mild destruction of lingual enamel
- 4 moderate destruction of lingual enamel with some exposure of dentine producing a definite ridge; islands of exposed dentine in centre of cusps
- 5 severe destruction of lingual enamel with exposure of dentine from the gingival margin to the

- occlusal surface; marked ridge at gingival margin
- 6 complete destruction of lingual enamel of exposed crown to form a continuous ridge at the gingival margin; some destruction of dentin

In the objective method, in addition to examining erosion of the tooth surface, the enamel thickness was measured. A method was given of mounting the first and second molars and sectioning them with a diamond cutting-wheel, making a transverse cut across the centre cusp of the first molar and the rear cusp of the second molar. After staining to differentiate enamel from dentin, the buccal and lingual enamel thicknesses were measured, and some hints were given on doing this accurately, along with some sources of minor error.

A simplified system for grading erosion in laboratory rats. This was used on jaws which had been stored in formalin and ethanol and were then examined under $\times 20$ magnification (7). The lingual surfaces of each maxillary molar and the first mandibular molar were separated mesiodistally into three regions, but the second and third mandibular molars were divided into two zones only, so that there were 32 scores for each rat, which could have values of

- 0 no erosion
- 1 slight erosion limited to the enamel
- 2 spot erosion with exposed dentine
- 3 lingual enamel gone (shelf formation)
- 4 erosion extended onto the buccal surface
- 5 massive erosion with reduction in cusp height and tooth volume

Then according to the authors, adding the mean value for each maxillary tooth to that for each mandibular tooth equalled the total mean severity score per rat.

The assessment techniques for use in laboratory animals and their advantages and disadvantages are set out in Table 2.

Table 2

Summary of methods for use in laboratory animals – pros and cons.

Method	References	Advantages	Drawbacks
Digital image analysis	12	Very sensitive. High accuracy. Quantitative data obtainable.	Expensive equipment.
Scanning electron microscopy	5, 13	Good reproducibility.	High equipment and labour costs. May be hard to quantify data.
Examination of selected teeth	18	Very detailed data obtainable.	Semi-quantitative.
Grading system of Restarski <i>et al.</i>	19, 20	Reproducible. Adapted by many others.	Concentrates on lingual enamel. Limited sensitivity.
Modified Restarski systems	21–24	Widely used. Simpler than the original. Quantitative.	Limited sensitivity.
Simplified grading system	7	Rapid. Quantitative	Moderate sensitivity

Techniques for the assessment of erosion in man

Some of the reports published in the literature deal with the generalized subject of tooth wear, which is inclusive of attrition, abrasion and erosion and combinations of these, while other authors have confined their attention to erosion and have attempted to exclude non-erosive damage. Few reports evaluate all three types of wear, most of them dealing with attrition or wear on occlusal surfaces. A source of variation is the way in which damage to the teeth has been recorded and estimated. Some of the papers on the subject give only general guidelines on taking a case history and conducting a clinical examination, whereas others define categories of severity and set out criteria for classifying the extent of erosion. An indication of the different approaches in the various papers published on the subject is given below.

Case histories and clinical examination. Some general advice on how to take case histories and inspect tooth surface loss are given by WATSON & TULLOCH (25). Differential diagnosis for trauma, abrasion, attrition, bruxism and erosion (palatally, 21/12), by making upper and lower study casts to examine tissue loss closely, can lead to a provisional diagnosis of erosion (26). Clinical features of erosion were reviewed by JOHNSON & SIVERS (27), detailing the appearance of the tooth surface as wear/erosion progresses, but again no scale of recording it was put forward.

Instances of erosion with case histories but no systematic classification were given by others (28, 29).

Scales of severity. ECCLES (30) pointed out that earlier classifications of erosion pertained chiefly to the epidemiology of industrial erosion (31, 32), but a different scale with 3 main classes was needed for erosion from dietary and intrinsic causes:

- I Superficial lesions involving enamel only
- II Localized lesions involving dentin for less than 1/3 of the surface
- III Generalized lesions involving dentin for more than 1/3 of the surface:
 - [a] facial surfaces
 - [b] lingual and palatal surfaces
 - [c] incisal and occlusal surfaces
 - [d] severe multisurface involvement

The clinical appearance of these grades was discussed in detail with the aid of photographs, and the point was made that class I lesions are frequently not diagnosed by dentists.

Because earlier indices had required that a diagnosis of the aetiology of the damage had to be made first, a new index was proposed by SMITH & KNIGHT (33). Each surface susceptible to wear was to be assessed visually, so that with 4 per tooth

Table 3

The tooth wear index developed by SMITH & KNIGHT (33).

Score	Surface	Criterion
0	B/L/O/I C	No loss of enamel surface characteristics No change of contour
1	B/L/O/I C	Loss of enamel surface characteristics Minimal loss of contour
2	B/L/O I C	Loss of enamel exposing dentin for <1/3 of the surface Loss of enamel just exposing dentin Defect <1 mm deep
3	B/L/O I C	Loss of enamel exposing dentin for >1/3 of the surface Loss of enamel and substantial loss of dentin, but not exposing pulp or secondary dentin Defect 1–2 mm deep
4	B/L/O I C	Complete loss of enamel, or pulp exposure, or exposure of secondary dentin Pulp exposure or exposure of 2ary dentin Defect >2 mm deep, or pulp exposure, or exposure of secondary dentin

[B=buccal or labial; L=lingual or palatal; O=occlusal; I=incisal; C=cervical]

×32 teeth, there would be a total of 128 surfaces per patient, and it would be straightforward to make a record either clinically or from photographs on a specially-designed chart that could be completed in under 5 min, with the advantage that calculations can be made with the raw scores to produce profiles of tooth wear distribution. The index is set out in Table 3.

Many different facets of the tooth wear index (TWI) were discussed (33), emphasising its efficiency, practicality and reproducibility, but the point should be made that it is not confined to the measurement of dental damage from erosion. Some examples of the use of the index were given along with tables and charts of data, including dietary erosion in a later paper (34). The index has also been applied in cases of erosion from regurgitation (35), including eating disorders (36), with the criteria quoted and the data carefully analysed.

The same TWI method was used by MILLWARD *et al.* to study erosion in 4-yr-old children (37), but as well as recording buccal, occlusal/incisal and lingual surfaces, the children were classified into one of three groups:

Low erosion:

scores of 0 and 1 for all teeth.

Moderate erosion:

any surfaces with scores of 2.

Severe erosion:

any surface with scores of 3 and/or 4.

The incisal edges of primary incisor and canine teeth were excluded, as it was pointed out that

these surfaces are particularly subject to attrition. Another piece of useful information, lacking in some of the other references, was the system of statistical analysis for distinguishing between groups: nonparametric methods using the chi-squared test for independent samples and the kappa statistic for reproducibility data.

The same method was also found to be applicable to children in the age range of 4 to 16.5 yr (38) for estimating erosion in relation to their drinking habits.

The SMITH & KNIGHT method was also used to estimate erosion from low-pH soft drinks, with the aid of photographic records and impressions/study models (39).

Tooth wear in general, with erosion as one component, was evaluated (40, 41) by an index based on RYGE'S system (42), which includes a decision on whether treatment is necessary as part of its criterion. The gradings, illustrated in the earlier reference by exceptionally clear colour photos, were grouped as

- R no visible wear or change in anatomical form
- S limited (normal) wear; limited change in anatomical form
- M considerable wear; obvious change in anatomical form but no need for treatment
- T considerable wear plus marked changes in anatomical form; further damage to tooth and/or surrounding tissues likely
- V excessive wear; extreme change in anatomical form, aesthetics and function; pain on chewing; damage to tooth and surrounding tissues.

The ØILO *et al.* paper (40) also had 12 codes for different degrees of occlusal wear, and the DAHL *et al.* paper (41) subdivided the groupings into three categories of satisfactory degree of wear (R, S and

M); and two categories of degree of wear not acceptable (T and V).

In response to rising concern over dental erosion, an assessment of erosion was included for the first time in the 1993 Survey of Children's Dental Health in the UK (43). Buccal and lingual surfaces of the primary and permanent maxillary incisors were assessed for loss of surface enamel characteristics and/or exposure of dentine or pulp (BA|AB and 21|12), recorded as

Depth 0 Normal

- 1 Enamel only – loss of surface characterisation
- 2 Enamel and dentin – loss of enamel, exposing dentin
- 3 Enamel into pulp – loss of enamel and dentin resulting in pulpal exposure
- 9 Assessment cannot be made

Area 0 Normal

- 1 Less than 1/3 of surface involved
- 2 1/3 up to 2/3 of surface involved
- 3 More than 2/3 of surface involved
- 9 Assessment cannot be made

The findings were included in the survey report, although the methods have been held to be not very discriminating, being based on the examination of only a limited number of tooth surfaces. It has been pointed out that the calibration criteria used are critical to the interpretation of the prevalence data. The dentists who performed the examinations found it difficult to agree in determining the presence of erosion, with wide variations in the measurement of erosion in the enamel, but not when it had reached the dentin.

The chief clinical methods for erosion assessment, with their main good points and drawbacks, are summarized in Table 4.

Table 4
Summary of methods for the assessment of erosion in man – pros and cons.

Method	References	Advantages	Drawbacks
Case histories and clinical examination	25–29	Can be done quickly (but see Drawbacks column).	Not systematic. Not reproducible. Training needed. Could become time-consuming.
Scale of severity	30	More precise.	Scale with only 3 main classes. Not specific for erosion.
Tooth wear index (TWI)	33, 34	Efficient, quick, practical, reproducible. Calculations straightforward.	Not specific for erosion. Needs modification for children.
TWI modified for use in children	37, 38	Detailed statistical analysis of data possible.	Not specific for erosion.
Tooth wear in general	40, 41	Clear divisions and coding.	Erosion acknowledged as only one component.
Assessment of 4 teeth per child for depth and area of erosion	43	Straightforward for large numbers of subjects. Includes both buccal and lingual surfaces.	Not very discriminating. Difficulty of standardization. Calibration criteria need careful setting.

Observations

Erosion has been of interest to the dental profession for many years, and a considerable body of literature on it has built up. It has proved impossible to cover every single published reference within the limits of this review, and it is quite apparent that the value and applicability of the methods of assessment they used vary enormously.

Adhering to the three main categories of techniques, all the methods for use *in vitro*, with the exception of the examination of extracted teeth, are sound and reproducible. For research purposes, chemical methods, digital image analysis and scanning electron microscopy should all be capable of high accuracy. Digital image analysis is the newest and can be computerized, so that a very large amount of data can be generated, stored and analyzed. Chemical methods are extremely sensitive and can be used to demonstrate very slight demineralization in the very earliest stages of erosion.

One advantage of using approaches *in vitro* is that they allow variables such as exposure time and the nature of the erosive agents to be studied individually or in combination, as required. A difficulty is the standardization of specimens, on account of the known variability in susceptibility of teeth to erosion both within and between individuals. Another factor to be taken into account is the species from which the specimen is obtained, as bovine teeth rather than human teeth have been used in some of the research *in vitro*.

All the methods quoted for recording erosion in laboratory animals appear to fulfil the research purposes for which they were devised. RESTARSKI's grading system and the later simplifications of it have all been carefully worked out and used successfully. The well-known method of RESTARSKI *et al.* (19, 20) is almost certainly the one that has been the most widely used, and some of the other systems proposed since then seem to be merely variants of it. One criticism that has been levelled against it is that it concentrates on lingual enamel rather than on the total tooth surface. However, it is likely to be increasingly replaced by digital image analysis on account of improved sensitivity and accuracy, as the equipment for this newer and faster method becomes more widely available and less expensive.

It should be recognized that the oral anatomy and physiology of laboratory animals, often rodents, show distinct differences from those of man, so that great caution is required in extrapolating the results to man.

Much more has been published on techniques for assessment in man. Some references on assessing erosion only by taking case histories and clinical examination are given above. Many dentists

would normally go no further than this, and some are entirely unaware that the scales and criteria of erosion summarized here exist!

One of the chief problems with the grading systems is to decide whether they can be used specifically for erosion, or whether they only record tooth wear in general, as most wear is multifactorial. It is clear that some of the systems were designed to encompass tooth wear from all sources (see Introduction), but one of the main advantages of the scale of SMITH & KNIGHT (33) is its application in evaluating the extent of erosion. Its major purpose is for use in adults, however, and some modification is required when it is to be applied to children (see details of methods above).

Many modifying factors need to be taken into account, such as the quantity and composition of saliva, and whether findings and decisions are based on data from cross-sectional or longitudinal studies. Existing methods of assessment may be adequate for cross-sectional studies but not sensitive enough to record small changes in tissue loss in longitudinal clinical studies, for which accurate surface topographical measurements are required. It is believed that some of the methods fail to take full account of changes in contact angles and the location of erosion lesions. The problem is that many of the reference points on a tooth surface are lost by erosion, rendering comparisons at different times impossible. This may be achieved only when there are restored surfaces which, if they are not susceptible to acid attack, will be left 'proud' by the erosive process and can serve as reference points for further tooth tissue loss. There is also uncertainty over the reliance that can be placed on conclusions drawn from the examination of models and photographs, and great care must be taken in interpreting data from the use of study models to monitor erosion.

Another factor to be considered is the nature of the tooth surface being examined. Use of the intact enamel surface permits investigation under physiological conditions, whereas grinding the surface flat prior to examination can remove surface aprismatic enamel and raise other issues, especially if dentin is exposed.

In summary, dependable methods exist for the assessment of erosive potential *in vitro* and dental erosion *in vivo*, but the prospective user would need to study the literature, get advice from someone experienced in evaluating erosion, and take time to learn the specialized techniques required.

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