Resin infiltration is considered a treatment option for initial caries lesions. To prevent enamel lesion s from further demineralization a complete and homogeneous penetration of low-viscosity resins should be accomplished. The aim of this study was to compare the penetration abilities of 3 commercial products: Icon (DMG, Germany), Fluorodose (Centrix Inc. USA) and Tetric flow (IvoclarVivadent, Schaan, Liechtenstein). Artificial white spot lesions were produced in 60 orthodontic extracted human premolars. The samples were randomly divided into 3 groups: F - weekly application of 5% fluoride gel; IC - resin infiltration (Icon1-DMG) and T - treatment with composite resins (Tetric flow). Specimens were studied using confocal microscopy and penetration depths were determined. A good correlation between PC and penetration depth was thereby observed (Pearson correlation coefficient, r=0.820).

Keywords: low viscosity lesions, penetration, white spot lesions, fluoride, initial caries

In modern dentistry, it has been widely accepted that no cavity design or restorative material will cure caries. For sure, once there is a cavity on the enamel surface, surgical intervention will be justified; in these situations, minimally invasive techniques (restricted to the actual damage) reduce the amount of destruction; biomimetic restorative materials (imitating nature while adhesively attached) allow for a satisfying clinical and aesthetic outcome, and, concomitantly the intervention will enable control of the local micro-flora (by modifying the local environment, and thus revealing that operative and restorative dentistry is but a true part of prevention). Nonetheless, minimally invasive approach is late in the disease process and destructive as well, and restorative materials are neither a perfect nor an everlasting replacement for original tooth structure. Thus, in view of the mean lifetime of any restoration type, the original anatomy, strength, and esthetics are lost forever (even with modern preparation concepts like slot, tunnel, or minbox restorations), and this will lead to the continuum of replacement dentistry [1, 2] with repeatedly enlarged restorations and increased damage of hard tissues. Moreover, invasive and even minimally or micro-invasive restorative procedures might be associated with postoperative sensitivity or pathogenic pulp reactions, sometimes requiring highly destructive endodontic treatment solutions.

In recent decades, a much more tissue preserving approach to arrest and control proximal or smooth-surface caries lesions has been studied extensively; this concept aims at occluding the highly porous structures of incipient enamel lesions by means of low-viscosity resins, and has been called penetration, plasticification,(therapeutic) sealing, infiltration, impregnation, noninvasive, or ultraconservative1 technique [3-9]. In medicine, infiltration means the act or process of infiltrating (as of liquids) into the pores or cavities of a substance; thus, this term seems most appropriate to describe the treatment approach using low-viscosity resin mixtures with high penetration capabilities on subsurface enamel lesions.

As long as the initial white spot lesion has an intact surface, the most effective means of caries control is an adequately performed oral hygiene including the complete removal of dental plaque using mechanical means such as tooth brushing and flossing [10]. However, while flossing in particular seems to be a reasonable recommendation for proximal surfaces, its preventive effect has not been supported by evidence up to now, neither with regard to gingival health [11] nor proximal caries, and only the professional use on a supervised basis has been identified to reduce caries risk (in children) [12,13]. This way, arrest of the lesion may be achieved, and remineralization becomes possible; remineralization is the natural repair process for non-cavitated lesions (occurring daily to repair the smallest demineralizations), and it relies on calcium and phosphate ions assisted by fluoride. However, optimal conditions are mandatory to ensure repair or healing by deposition of mineral on existing damaged crystals or nucleation and de novo crystal formation [14].

The use of topical fluorides to enhance remineralization of demineralized proximal enamel has been advocated [10]. Application of fluorine varnish every third month significantly reduced the progression of proximal caries lesions in premolars and molars. The most obvious reduction of caries progression was observed among children with moderate caries risk, while children with high cariesactivity (more than 9 new proximal lesions) did not benefit from proximal caries reduction [15].

From early histological experiments, it is well known that enamel lesions can be imibed, because of the increased micro porosities of the different histological zones. Moreover, these tiny porous openings and widened inter-crystalline spaces act as diffusion pathways for acids and dissolved minerals. With this in mind, it should be possible to infiltrate incipient lesions with other liquids, ie, with low-viscosity resins. Thus, instead of removing the porous canious tissue at a relatively late stage in the disease process, attempts have been made to fill the microporosities of lesions at a much earlier stage of lesion development. This would not only reduce the microporosities (and therefore the access of acid), but also afford some mechanical support to the tissue [16].
Further experiments revealed that viscosity, surface tension, and contact angle on human enamel influence the penetration of restorative resins into acid-etched enamel; however, viscosity of the monomer mixture was not shown to be a limiting factor for the penetration of restorative resin monomers into the pores of etched enamel surfaces [17], and depth of penetration decreased only slightly with changing viscosity. Interestingly, tag lengths (representing the penetration depth) of up to 50 µm or more were observed with composite as well as with non-composite resins on (not carious) phosphoric acid-etched enamel [18-20].

Another factor seems to be the degree of microporosities. Penetration of an unfilled resin into enamel was considerably influenced by the degree of dental hard tissue mineralization. Resin tags in demineralized enamel were significantly longer (some 60 µm) than in other groups, and penetration decreased significantly in remineralized areas or after use of fluoride; however, this was still significantly deeper than in control sites, and remineralized enamel also allowed good penetration of the unfilled bonding agent [21]. Various modifications (additionally to acid concentration and etching time) of the application technique have been proposed to improve the penetration depths in sound but etched enamel.

With orthodontic sealants, similar results could be observed. Light-cured sealant treatment after orthodontic appliance placement significantly reduced or even prevented enamel demineralization [22,23]. In another study, demineralization in the sealant group was reduced significantly, and teeth treated with fluoride varnish exhibited 30% less demineralization than the control teeth. Usually, sealants are applied after acid etching, and removal of surface coating after completion of orthodontic therapy will leave resin tags in formerly etched enamel; these areas have been shown to be caries resistant as well [24]. Therefore, particularly in patients who exhibit poor compliance with oral hygiene and home fluoride use, sealing has been recommended [25].

The aim of the present investigation was to evaluate in vitro the penetration abilities of 3 experimental materials into artificial enamel caries lesions.

**Experimental part**

**Material and method**

Specimens were prepared from 60 human premolars extracted for orthodontic purposes, which were stored in 0.5% chloramine solution (Sigma Aldrich, Buchs, Switzerland) for 7 days followed by storage in distilled water for 14 days after extraction. The teeth were cleaned and sectioned at the enamel-cementum junction using a water-cooled cutting wheel. After demineralization each specimen was randomly assigned to three groups and treated. In the first group (group F) weekly application of 5% fluoride gel (Fluorodose, Centrix Inc. USA) were treated. In the second group (group IC) the enamel was etched for 2 min with 15% hydrochloric acid (Elipar S10, 3 M ESPE, Seefeld, Germany). The surfaces in the last group were etched for 2 min with 15% hydrochloric acid (Icon Dry, DMG, Hamburg, Germany) for 30 s and additional air drying. Then, the low-viscosity resin infiltrant (Icon Infiltrant, DMG, Hamburg, Germany) was applied on the surface for 3 min by means of the sponge applicator provided with the resin infiltration system. After light-curing for 40 s, the infiltrant was applied for further 60 s and again light-cured for 40 s.

A qualified researcher examined all the sections under a microscope. Each section was evaluated and photomicrographs were taken of representative areas that were most often observed. For the photomicrographs, an A377 digital microscope camera with USB port was used. The camera has a CMOS sensor of 2 MPX and a magnification of 20X-800X, manual focus from 0-40 mm and allows a measurement of 0.03 mm. It also has 10 LED lights, the intensity of the light can be manually adjusted.

The operating system was Windows XP.

**Results and discussions**

After 10 s application, the low-viscosity resin infiltrant showed the deepest penetration. Concomitantly, these infiltrants completely infiltrated the lesion body. None of the other materials was able to penetrate the lesion body completely and the commercially available adhesive, Tetric Flow, showed only 40 (30) % penetration of the lesion depth.

Infiltration of enamel caries is a promising therapeutic approach that might bridge the gap between preventive and operative dentistry. It was the aim of the present work to explore and identify the material characteristics which allow rapid infiltration of enamel caries. The knowledge of these material characteristics might be important for the development of a new group of dental resins, known as the infiltrants.

Although an infiltration of the outer layers of the lesion body might inherently delay caries progression, a recent study showed a negative correlation between depth of penetration and progression of caries lesions in a demineralizing environment. Therefore, the aim of any resin infiltration should be the occlusion of preferably as many as possible the pores of caries lesions.

In the present investigation, artificially induced caries lesions were infiltrated. In contrast to natural incipient caries lesions, which are hard to obtain in acceptable numbers, artificial caries lesions can be produced in comparably high numbers. Moreover, due to their relatively uniform structure and lesion depth, the latter seem to be more suitable to the comparison of several materials as compared to natural lesions. However, it must be put into perspective that resin penetration into natural lesions might differ from that observed in artificial caries [26].

The limitation of this study consists in the fact that it was done in vitro. In vivo for example, the pores of an enamel lesion are not as open as assumed. Thus, entrapped air in the depth of the lesions could hamper resin penetration [28]. However, this effect (possibly resulting in deceleration of penetration and incomplete infiltration) could not be observed in the present study. Moreover, the pores of natural lesions are most likely to be contaminated or even occluded by organic materials like salivary proteins or food debris. Organic impurities significantly reduce the surface free energy of enamel, and therefore might impede resin penetration. Finally, it is unlikely that the pores within
enamel lesions are of uniform diameter at different lesion depths. Thus, penetration speed might depend on lesion depth. Viscosity of the monomer mixture was not shown to be a limiting factor for the penetration of restorative resin monomers into the pores of etched enamel surfaces [17] and depth of penetration decreased only slightly with changing viscosity. Interestingly, tag lengths (representing the penetration depth) of up to 50 µm or more were observed with composite as well as with non-composite resins on (non-carious) phosphoric acid-etched enamel [18-20]. In a recent article, similar penetration depths after surface conditioning with hydrochloric acid were found in natural lesions [28]. Some commercially available adhesives (sealants, bonding agents) have been shown to be suitable for infiltration of artificially induced subsurface lesions, as well [29-31], but significant differences could be revealed with artificial lesions, when various resin infiltrants with differing penetration coefficients were used [32]. This has been corroborated with natural lesions recently, thus indicating that resin infiltrants with high penetration coefficients are able to penetrate more deeply into subsurface lesions. A previous SEM study on sealant penetration into etched fissures seems to confirm these observations. Here, a low-viscosity sealant penetrated fully and formed a resin infiltrated layer in enamel beyond the etched depth. However, the high-viscosity sealants used in that study did not penetrate enough to ensure that the acid-etched enamel was infiltrated sufficiently by the sealant to assure good marginal seals [33]. A recent SEM study on in vivo sealed (ClinproSealant, 3M ESPE; with and without a preceding bonding) natural subsurface lesions demonstrated an irregular resin network with twisted and curved tags, while with the sound enamel areas a regular etching pattern was observed. Resin tag lengths were considerably short and ranged from 4.2 to 5.5 µm. No increased penetration depths could be observed after the additional use of a low viscosity adhesive bonding agent (Single Bond, 3M ESPE). No further pretreatment of enamel was performed, and acid etching of the surface zone was done with a phosphoric acid gel [34]. Penetration depths of the sealant were some what higher in another study using the same design; however, pretreatment with a bonding agent resulted in decreased tag lengths [35]. Nevertheless, in both studies, a physical barrier was formed with protective function against exposure of acids from bacterial origin, and cutting off possibly remaining bacteria (within an advanced lesion) from a nutritional supply of fermentable carbohydrates. This was corroborated in a clinical study on sealed (Gluma One Bond, Heraeus Kulzer; or Concise Sealant, 3M ESPE; 18 months, 72 patients) proximal early active lesions. As validated by subtraction radiography, 43.5% of the sealed proximal lesions had progressed during the 18-month study, while 84.1% of the untreated controls (flossing) showed increased demineralization depths [36], thus indicating a reduced (and not an arrested) progression rate for the procedure. Interestingly, deeper test lesions showed lower progression rates (33%) when compared to untreated control sites, thus reemphasizing the results already known from fissure sealants to some extent [37]. From these observations, it might be speculated whether lesion arrest over longer periods after infiltration is due to reduced microorganism viability or physical barrier against acids from bacterial origin [38].

<table>
<thead>
<tr>
<th>Material</th>
<th>Composition</th>
<th>Properties</th>
<th>Table 1 COMPOSITION AND PHYSICAL PROPERTIES OF THE STUDIED MATERIALS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fluoridcse</td>
<td>NaF %</td>
<td>NaF release 3017</td>
<td></td>
</tr>
<tr>
<td></td>
<td>NaF PPM</td>
<td>F release 1365</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Fluoride PPM</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Icon</td>
<td>Icon etch</td>
<td>Elastic modulus 0.9</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Icon Dry: ethanol</td>
<td>Knoop Hardness 6.54</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Icon Infiltrant:</td>
<td>Softening ratio 45.59</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Tri-ethylene-glycol-dimethacrylate-based resin</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tetrc flow</td>
<td>Bis-GMA</td>
<td>Flexural strength 114 MPa</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Urethane dimethacrylate</td>
<td>Flexural modulus 5100 MPa</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Decadiol dimethacrylate 37.6</td>
<td>Density 1.78 g/cm³</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Barium glass filler</td>
<td>Vickers hardness HV 0.530-320</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Ytterbium-tri-flouride</td>
<td>Compressive strength 260 MPa</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Mixed oxide</td>
<td>Depth of cure &gt; 2.0 mm</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Highly dispersed silica</td>
<td>Radiopacity (Bleach L, M, XL)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Prepolymers</td>
<td>Water solubility (7 days) 0.1</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Additives, Catalysts and Stabilizers</td>
<td>Flexural strength 21.0 µg/mm²</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Water absorption (7 days)</td>
<td></td>
</tr>
</tbody>
</table>

Table 2 PERCENTAGE OF PENETRATION DEPTH FOR EACH TYPE OF MATERIALS INTO THE PREVIOUSLY DEMINERALIZED ENAMEL LESIONS

<table>
<thead>
<tr>
<th>Material</th>
<th>Percentage of penetration depth</th>
<th>Standard deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low viscosity</td>
<td>111</td>
<td>21</td>
</tr>
<tr>
<td>resin infiltrant</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adhesive</td>
<td>40</td>
<td>29</td>
</tr>
<tr>
<td>Fluoride</td>
<td>11</td>
<td>4</td>
</tr>
</tbody>
</table>
In another paper was studied the toxic effect of resin-based dental materials [39].

Conclusions

In conclusion, the use of a caries infiltrant might be beneficial as a treatment in demineralized enamel as well as for infiltration of adjacent demineralized enamel not worth to be sacrificed during restorative procedures. For complete resin infiltration of artificial enamel caries lesions within a short period, infiltrants should preferably show a high penetration coefficient.

Aknowledgements: The funding for this study was obtained from the Project for young researchers - Programme II-C3-TC-2015.

References


Fig. 1 Microscopic images of demineralized tooth surfaces and mineralization with different types of materials. Images a, c, e: demineralized lesions; b: mineralization with low viscosity resin infiltrant; d: adhesive infiltrant; f: mineralization with fluoride gel.

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