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ORIGINAL ARTICLE



# Effect of resin infiltration and microabrasion on the microhardness, surface roughness and morphology of incipient carious lesions

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**ABSTRACT**

**Objective:** The effects of resin infiltration and microabrasion on incipient carious lesions by surface microhardness, roughness and morphological assessments, and resistance to further acid attack of treated lesions were evaluated.

**Material and methods:** Eighty artificially-induced incipient lesions were randomly divided into five groups ($n = 16$): resin infiltration with an adhesive resin (Excite F, Ivoclar Vivadent, Schaan, Liechtenstein), resin infiltration with a resin infiltrant (Icon, DMG, Hamburg, Germany), microabrasion without polishing (Opalustre, Ultradent, South Jordan, UT, USA), microabrasion with polishing (Opalustre, Ultradent, Diamond Excel, FGM, Joinville, SC, Brazil), and distilled water (control group). All specimens were exposed to demineralization for another 10 d. Microhardness, roughness and morphological assessments were done at baseline, following initial demineralization, treatment and further demineralization. Data were analysed by the Kruskal–Wallis, Friedman's and Bonferroni tests ($p < .05$).

**Results:** Enamel lesions treated with resin infiltrant and microabrasion demonstrated similar hardness values, with a nonsignificant difference compared with sound enamel. Resin infiltration demonstrated lower roughness values than those of microabrasion, and the values did not reach the values of sound enamel. Further demineralization for 10 d did not affect the hardness but increased the roughness of infiltrated and microabraded enamel surfaces. Polishing did not influence the roughness of microabraded enamel surfaces. After resin infiltration, porosities on enamel were sealed completely. The surface structure was similar to that of the enamel conditioning pattern for microabraded enamel lesions.

**Conclusions:** Within the limitations of this study, the icon infiltration and microabrasion technique appeared to be effective for improving microhardness. Icon appeared to provide reduced roughness, although not equal to sound enamel. Further research is needed to elucidate their clinical relevance.

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## Introduction

A new minimally invasive approach has been recently described to treat noncavitated approximal and smooth surface carious lesions [1,2]. This infiltration concept is based on the penetration of low-viscosity resins, so-called infiltrants, in noncavitated caries lesions, the surface of which is previously conditioned with a strong acid [1,2]. The infiltrants are capable of penetrating the porous structures of the lesion body, creating a diffusion barrier for cariogenic acids and thereby preventing lesion progression [1]. A resin material with a low viscosity and a high penetration coefficient has been developed and marketed under the name Icon caries infiltrant, since 2009 [3]. The technique requires conditioning of the hypermineralized enamel surface with 15% hydrochloric acid for two minutes [4]. This removes enamel surface approximately 34–58 μm in depth [2,5], to obtain resin penetration into the lesion [3].

Such microinvasive treatment of incipient caries has been recently shown to significantly reduce mineral loss after demineralization challenge and to arrest artificial enamel lesions compared with untreated lesions [6]. It has been confirmed by randomized placebo-controlled clinical trials that resin infiltration is effective at arresting the progression of noncavitated approximal carious lesions [7–9].

Microabrasion is also used in the microinvasive treatment of incipient carious lesions as an alternative technique for noncavitated lesions [10–12]. The technique has been widely used to remove superficial noncarious enamel defects and superficial dysmineralization defects, and has been supplemented with tooth bleaching techniques since its introduction in 1986 [13,14]. The mechanism of treatment has been explained by the removal of the outermost regions of enamel by mechanical abrasion. The simultaneous chemical erosion presumably results from the creation of a smooth and lustrous surface (abrasion effect) [11,13]. This treatment approach has been advocated for the management of white spot lesions caused by orthodontics [10–12]. The technique involves repeated applications of an acidic and abrasive compound to the demineralized enamel surface [11,12,15]. The commercial product name of Opalustre, which contains approximately 6.6% hydrochloric acid and silicon carbide abrasive particles, has been introduced to the market [12,16].

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The technique was reported to result in the loss of 25–200 μm of enamel thickness, when 5–10 applications of microabrasive systems were applied, meaning it can be considered a safe and conservative procedure [11,12]. The Opalustre technique might be effective and long-lasting for the removal of white spot lesions and for the production of more regular and smooth surface without causing significant tooth structure loss.

In spite of the fact that these favorable results may have been achieved by using microinvasive treatment techniques for incipient caries lesions to establish minimally invasive dentistry, several other evaluations and comparisons need to be carried out. Limited *in vitro* studies evaluating these two microinvasive treatment methods (resin infiltration and microabrasion) have been performed to date [16–18], especially considering their effects on surface quality of enamel surface of incipient carious lesions. To date, there is no evidence that polishing has a positive effect on surface quality of microabraded incipient lesions. There is also a lack of evidence for the effect of further demineralization on incipient carious lesions treated during the treatments. Therefore, the objectives of this *in vitro* study were to compare the effects of resin infiltration (using adhesive resin or resin infiltrant) and microabrasion (with and without polishing) on incipient carious lesions by assessing surface qualities in terms of surface microhardness, surface roughness, and morphological assessments, and to evaluate the resistance to new acid attack of the incipient carious lesions after microinvasive treatment.

The null hypotheses tested were that (1) there is no difference regarding surface microhardness (1a) and surface roughness (1b) of the lesions between the two microinvasive treatment techniques; and (2) new acid attacks do not alter surface microhardness (2a) and roughness (2b) of incipient lesions after microinvasive treatment.

## Materials and methods

### Specimen preparation

Eighty enamel specimens (5 × 4 × 3 mm) were prepared from the labial surfaces of bovine incisor crowns by cutting at the middle of the crown using a rotary diamond disc (Thin-flex X927-7, Premier Dental Products Co., Ontario, Canada) mounted on a dental micromotor handpiece. After embedding in epoxy resin (Epofix Resin, Struers, Erkrath, Germany), specimen surfaces were ground flat with sequential aluminium oxide abrasive papers (500-, 1200-, 2400- and 4000-grit; FEPA-P, Struers, Ballerup, Denmark) in a polishing device (Labopol-5, Struers, Ballerup, Denmark) for 20 s each. Specimens were then inspected under a 40× stereomicroscope (S4E, Leica Microsystems, Wetzlar, Germany) to ensure that they were free of caries, fracture or other defects.

### Artificial caries formation (initial demineralization)

Artificial incipient carious lesions were created by specimen storage in 5 L of a demineralization solution containing 50 mM acetic acid, 3 mM calcium chloride dihydrate, 3 mM potassium dihydrogen phosphate, 10 mM potassium hydroxide, 6 μM methyl hydroxy diphosphonate, and distilled water (pH 5.0, 37 °C) for 10 d [19]. The pH of the solution was monitored daily and, if necessary, adjusted with either hydrochloric acid or potassium hydroxide.

To determine the demineralization degree of enamel specimens, laser fluorescence measurements were performed at four-time intervals using a caries detection tool (Diagnodent pen, Kavo, Biberach, Germany): at baseline, following initial demineralization, treatment, and further demineralization. Before each session, the instrument was calibrated against the ceramic standard supplied by the manufacturer. The surface was dried with compressed air for 5 s before measurements, and the type B probe, which is for smooth surfaces, was used. The labial window area was carefully scanned using the probe by holding the tip in close contact with the enamel surface and tilting the tip around the measuring site in order to collect the fluorescence from all directions. At each sample surface, four sides were determined: these were the upper and lower middle points of the square mesially and distally. Each point was scanned twice, and the highest value from the two readings was registered. Laser fluorescence measurements were recorded, and the lesions were considered as enamel lesions according to the Diagnodent pen readings (values range from 14 to 20) [20].

### Microinvasive treatments and further demineralization

Eighty artificially-induced incipient lesions were randomly divided into five groups according to the treatment methods: ($n = 16$ per group); resin infiltration with a two-step etch and rinse adhesive resin (AR; Excite F, Ivoclar Vivadent, Schaan, Liechtenstein), resin infiltration with a resin infiltrant (RI; Icon, DMG, Hamburg, Germany), microabrasion without polishing (MA; Opalustre, Ultradent, South Jordan, UT, USA), microabrasion with polishing (MAP; Opalustre, Ultradent; Diamond Excel, FGM, Joinville, SC, Brazil), and immersion in distilled water (CON; control group). The general composition and application procedures of the materials used for each group are shown in Table 1.

For further demineralization, all specimens ($n = 80$) were immersed in a freshly prepared demineralization solution (pH 5.0) at 37 °C for the same amount of time (10 d) as for initial demineralization after application of treatments.

### Surface microhardness

The microhardness measurements were performed at baseline and following initial demineralization, microinvasive treatment, and further demineralization. Three indentations, spaced 100 μm from each other, were made on the enamel surfaces using a Vickers indenter, with a static load of 200 g for 15 s coupled with a microhardness tester (Duroline M, Metkon Instruments Inc., Bursa, Turkey). The average of the three readings was taken as the mean Vickers hardness number (VHN, kgF/mm$^2$) of the specimens.

### Surface roughness

The surface roughness measurements of the specimens were performed using a stylus profilometer (Surftest SJ-210, Mitutoyo, Sakado, Japan) to an accuracy of 0.01 μm, a range of 2.00 and 0.25 mm cut-off. For each specimen, three measurements were performed in four evaluation periods as stated above for microhardness measurement, and average roughness values (Ra, μm) were recorded.

### Statistical analysis

After the microhardness and the roughness values were calculated, statistical analyses were applied to each group at all evaluation periods. The Kruskal–Wallis test was used to verify differences between groups (intergroup comparisons), and Friedman's test was used to analyse differences between evaluation periods (intragroup comparisons). The statistical significance level was set at 0.05. All statistical analyses were performed with SPSS software version 21.0 (SPSS Inc., Chicago, IL).

### Surface morphology

Three enamel specimens were selected from each group in four evaluation periods, and a scanning electron microscope (Vega II, Tescan, Cambridge, England) was used to analyse the surface morphology of the specimens. The specimens were ultrasonically cleaned in deionized water for 20 min, dried for 24 h in a dessicator, and the surface morphology of enamel specimens was analysed. The most representative images were archived for illustration.

## Results

### Laser fluorescence measurement

The values (median and interquartile ranges) obtained with the laser fluorescence device were $0.00 \pm 0.25$ at baseline for all groups (sound enamel). There were no statistically significant differences in the values among the five groups (Kruskal–Wallis test and Bonferroni post hoc test, $p > .05$). After initial demineralization, laser fluorescence values were $16.13 \pm 0.94$ for the adhesive resin group (AR), $16.00 \pm 1.00$ for the resin infiltrant group (RI), $16.50 \pm 0.50$ for the microabrasion without polishing group (MA), $16.50 \pm 0.25$ for the MAP group, and $16.10 \pm 1.00$ for the control (CON) group. There were no statistically significant differences in the values between the five groups after initial demineralization (Kruskal–Wallis test and Bonferroni post hoc test, $p > .05$). The differences between baseline and initial demineralization values were statistically significant in all of the treatment groups (Friedman's and Bonferroni *post hoc* test, $p = .000$).

### Surface microhardness

The median and interquartile ranges of the surface microhardness values (VHN) are presented for each group and evaluation period in Tables 2 and 3. The microhardness values were significantly increased for both infiltrated and microabraded enamel surfaces in comparison with the values for demineralized enamel surfaces, and resulted in a statistically nonsignificant difference compared with sound enamel, except for Excite F (Friedman's test, $p < .05$). All of the treated groups were statistically different from the control. Specimens treated with Icon and microabrasion used with

**Table 1.** Materials used for each group, their composition and application procedure.

| Groups | Material | Composition | Application procedure |
|---|---|---|---|
| Resin infiltration (adhesive resin, AR) | UltraEtch + Excite F | *Etchant*: 35% phosphoric acid, cobalt aluminate blue spinel, cobalt zinc aluminate blue spinel *Adhesive*: Phosphoric acid acrylate, 2-hydroxyethyl metacrylate, bisphenol A-glycidyl dimethacrylate, alcohol, di-methacrylates, silicon dioxide, initiators, stabilizers | Etch enamel lesions for 30 s. Rinse for 30 s and air-dry for 30 s. Apply adhesive resin for 30 s and remove resin surplus using a cotton roll and light-cure for 30 s (600 mW/cm$^2$). Repeat adhesive resin application, apply a glycerine gel and light-cure for 60 s. |
| Resin infiltration (resin infiltrant, RI) | Icon smooth surface | *Icon-Etch*: 15% hydrochloric acid *Icon-Dry*: 100% ethyl alcohol *Icon Infiltrant*: Methacrylate-based resin matrix, initiators, additives | Etch enamel lesions for 120 s, rinse for 30 s and air-dry for 30 s. Apply ethanol for 30 s, and air-dry for 30 s. Infiltrate resin for 180 s with rubbing, remove resin surplus using a cotton roll and light-cure for 60 s (600 mW/cm$^2$). Repeat resin infiltrant application for 60 s, apply a glycerine gel and light-cure 60 s. |
| Microabrasion without polishing (MA) | Opalustre | *Microabrasion slurry*: 6.6% hydrochloric acid and silicon carbide microparticles (20–160 μm) | Apply slurry to enamel lesions using rubber cups with the aid of a low-speed handpiece for a period of 60 s at intervals of 30 s. |
| Microabrasion with polishing (MAP) | Opalustre Diamond Excel | *Microabrasion slurry*: 6.6% hydrochloric acid and silicon carbide microparticles (20–160 μm) *Polishing paste*: Diamond micronized granulation 2–4 μm base lubricant, thickener, emulsifier | Apply slurry to enamel lesions using rubber cups with the aid of a low-speed handpiece for a period of 60 s at intervals of 30 s. Apply diamond paste using felt discs with the aid of a low-speed handpiece for 30 s. |
| Control (CON) | Distilled water | Distilled water | Specimens were soaked in distilled water. |

**Table 2.** Microhardness values (VHN) and intergroup comparisons in each evaluation period.

| | Groups with median and interquartile ranges | | | | | | |
|---|---|---|---|---|---|---|---|
| Evaluation stage | AR | RI | MA | MAP | CON | $p$* | Significant pairs** |
| Sound enamel | 330.7 (0.6) | 330.8 (0.6) | 330.7 (0.5) | 330.9 (0.3) | 330.9 (0.6) | .511 | – |
| Initial demineralization | 132.0 (0.3) | 131.7 (0.6) | 131.8 (0.5) | 131.7 (0.7) | 131.7 (0.5) | .279 | – |
| Microinvasive treatment | 211.6 (0.2) | 261.9 (0.9) | 304.5 (0.3) | 305.6 (0.5) | 131.7 (0.5) | .000 | AR&RI, AR&MA, AR&MAP, AR&CON, RI&CON, MA&CON, MAP&CON |
| Further demineralization | 173.5 (0.4) | 212.6 (0.8) | 272.5 (0.3) | 270.9 (0.5) | 85.9 (0.7) | .000 | AR&RI, AR&MA, AR&MAP, AR&CON, RI&CON, MA&CON, MAP&CON |

*Kruskal–Wallis test.
**Bonferroni post hoc test ($p < .05$).
AR: adhesive resin; RI: resin infiltrant; MA: microabrasion without polishing; MAP: microabrasion with polishing; CON: control

**Table 3.** $p$ Values of microhardness for intragroup comparisons (between evaluation periods).

| | | | Groups | | | | |
|---|---|---|---|---|---|---|---|
| Evaluation stage | | | AR | RI | MA | MAP | CON |
| Sound enamel | – | Initial demineralization | .000 | .000 | .000 | .000 | .006 |
| Initial demineralization | – | Microinvasive treatment | .000 | .000 | .000 | .000 | 1.000 |
| Sound enamel | – | Microinvasive treatment | .000 | .171 | .171 | .171 | .000 |
| Microinvasive treatment | – | Further demineralization | .171 | .171 | .171 | .171 | .006 |

Underlined $p$ values show a significant difference (Friedman's and Bonferroni post hoc test, $p < .05$).
AR: adhesive resin: RI: resin infiltrant; MA: microabrasion without polishing; MAP: microabrasion with polishing; CON: control

and without polishing demonstrated similar hardness values, which were significantly higher than the hardness values of specimens treated with Excite F (Kruskal–Wallis, $p < .05$). Polishing did not change the microhardness values of microabraded enamel surfaces (Kruskal–Wallis, $p > .05$). Further demineralization did not significantly affect the microhardness values of the specimens treated with resin infiltration and microabrasion (Friedman's test, $p > .05$).

### *Surface roughness*

The median and interquartile ranges of surface roughness values (Ra) are presented for each group and evaluation period in Tables 4 and 5. The roughness values were significantly lower for both infiltrated and microabraded enamel surfaces in comparison with the values for demineralized enamel surfaces but did not reach the values of sound enamel (Friedman's test, $p < .05$). Specimens treated with Excite F showed the lowest roughness results, followed by Icon, microabrasion with polishing, microabrasion without polishing, and control (Kruskal–Wallis, $p < .05$). Polishing led to a decrease in roughness values of microabraded lesion surfaces, but statistically significant differences between the two groups did not remain following further demineralization (Kruskal–Wallis, $p < .05$). The infiltrated and microabraded enamel surfaces had lower roughness values compared with the specimens subjected to further demineralization in all of the groups (Friedman's test, $p < .05$).

### *Surface morphology*

Observable alterations were seen in the SEM analysis of enamel surface morphology in the four evaluation periods. Figure 1(a) shows sound and intact enamel surfaces. The surface structure changed with voids and many micropores after initial demineralization (Figure 1(b)). After Icon and Excite F applications, enamel porosities were sealed following infiltration of resinous materials, and the specimen surfaces showed no visible areas of unprotected enamel (Figure 2(a,c)). After microabrasion, the surface structure with dissolution of interprismatic enamel was similar to the enamel conditioning pattern (Figure 2(e,g)). Polishing did not induce visible changes in the microabraded surface morphology (Figure 2(g)). Further demineralization after microinvasive treatment did not clearly affect enamel surface morphology in any of the groups (Figure 2(b,d,f,h)).

## Discussion

A non-invasive evaluation method used for white spot lesions in many *in vivo* and *in vitro* studies is the laser fluorescence method, based on the red end of the electromagnetic spectrum with light wavelength of 655 nm (Diagnodent or Diagnodent pen, KaVo, Biberach, Germany). The difference between the fluorescing capacity of the sound tooth and the carious tooth tissue can be recorded by the device. The difference in the properties of reflection, transmission and color absorption between demineralized and healthy teeth affects the laser fluorescence reading, which helps detect caries. One theory regarding the exact mechanism of the method is that the red light stimulates fluorescent light of a different wavelength due to a change in tooth tissue, such as porosity due to demineralization. The second theory is that fluorescence depends on emission from the metabolites, such as porphyrins produced by cariogenic bacteria. The ability to detect artificial caries lesions induced without bacteria by the laser fluorescence device has also been reported [21,22]. In this study, there were no statistically significant differences in the values obtained with the laser fluorescence device among the groups for the initial demineralization period. This shows that the demineralization solution produced uniform artificial carious lesions in the study. The values obtained with the laser fluorescence device after demineralization were significantly different from baseline values. It could be related to an

**Table 4.** Roughness values (Ra) and intergroup comparisons in each evaluation period.

| | Groups with median and interquartile ranges | | | | | | |
|---|---|---|---|---|---|---|---|
| Evaluation stage | AR | RI | MA | MAP | CON | $p$* | Significant pairs** |
| Sound enamel | 0.020 (0.000) | 0.020 (0.000) | 0.020 (0.000) | 0.020 (0.000) | 0.020 (0.000) | .406 | – |
| Initial demineralization | 0.171 (0.001) | 0.171 (0.000) | 0.171 (0.000) | 0.171 (0.000) | 0.171 (0.000) | .173 | – |
| Microinvasive treatment | 0.029 (0.001) | 0.042 (0.001) | 0.081 (0.001) | 0.061 (0.001) | 0.171 (0.000) | .000 | AR&RI, AR&MA, AR&MAP, AR&CON, RI&MA, RI&MAP, RI&CON, MA&MAP, MA&CON, MAP&CON |
| Further demineralization | 0.061 (0.001) | 0.067 (0.001) | 0.111 (0.000) | 0.094 (0.000) | 0.246 (0.001) | .000 | AR&MA, AR&MAP, AR&CON, RI&MA, RI&MAP, RI&CON, MA&CON, MAP&CON |

*Kruskal–Wallis test.
**Bonferroni post hoc test ($p < .05$).
AR: adhesive resin; RI: resin infiltrant; MA: microabrasion without polishing; MAP: microabrasion with polishing; CON: control

**Table 5.** $p$ Values of roughness for intragroup comparisons (between evaluation periods).

| Evaluation stage | | | Groups | | | | |
|---|---|---|---|---|---|---|---|
| | | | AR | RI | MA | MAP | CON |
| Sound enamel | – | Initial demineralization | .000 | .000 | .000 | .000 | .006 |
| Initial demineralization | – | Microinvasive treatment | .000 | .000 | .000 | .000 | 1.000 |
| Sound enamel | – | Microinvasive treatment | .006 | .006 | .006 | .006 | .006 |
| Microinvasive treatment | – | Further demineralization | .006 | .006 | .006 | .006 | .006 |

Underlined $p$ values show a significant difference (Friedman's and Bonferroni post hoc test, $p < .05$).
AR: adhesive resin; RI: resin infiltrant; MA: microabrasion without polishing; MAP: microabrasion with polishing; CON: control

increase in the porosity of enamel and adequate detection of changes in mineral content by the device.

White spot lesions are characterized by tiny pores within the lesion body of initial enamel caries, whereas the surface of the lesion remains relatively intact (pseudointact surface layer). Because it could hamper the resin from penetrating into the lesion, the pseudointact surface layer is removed by acid etching with hydrochloric acid using a resin infiltration technique [2,4,23]. Hydrochloric acid in similar concentrations is also accepted in esthetic dentistry to remove superficial discolorations and as a proposed treatment approach for the management of white spot lesions using enamel microabrasion [10–12]. However, more superficial enamel surface removal has been achieved with the resin infiltration technique compared with enamel microabrasion [2,5]. The primary aim of the resin infiltration technique is not complete removal of the surface layer, but rather, to increase the penetrability of low-viscosity light curing resins into the pores of incipient lesions, thereby protecting the lesion from further acid attacks. Moreover, after curing the resin material, lesion progression might be arrested and a mechanical support for the enamel lesion structure might be achieved [2,24,25].

Mechanical properties of enamel submitted to different treatments have been evaluated by surface or cross-sectional hardness evaluation [6]. This study found no differences in microhardness between the lesions treated with resin infiltration using an infiltrant (Icon) and microabrasion (with or without polishing), except for the infiltration using adhesive resin (Excite F). Thus, the first null hypothesis, that there is no difference regarding surface microhardness (1a) of the lesions between the two microinvasive treatment techniques, was not rejected.

The mechanical properties of the resin infiltration with low viscosity resin might be influenced by the degree of demineralization, penetration of the resin, and monomer and solvent compositions of infiltrants [6,23]. The previous approach to infiltrate the artificial and natural enamel lesions using infiltrants with differing penetration coefficients showed that the resin infiltrants with high penetration coefficients were able to penetrate more deeply into subsurface lesions [2,26–28]. In this study, higher VHN values were found for Icon infiltrated enamel lesions than for Excite F infiltrated enamel lesions, indicating better penetration ability of the resin infiltrant to fill the spaces between the remaining crystals of the porous lesion. The results are in accordance with the literature [6,29].

When microabrasion techniques were employed, in combination with polishing or not, microhardness was significantly increased compared with untreated lesions and reached the values of sound enamel, meaning that these microinvasive caries treatment methods are effective for management of incipient caries lesions (Tables 2 and 3). Although limited data are available in the literature regarding the effect of microabrasion on enamel hardness, the technique previously revealed an increased microhardness of enamel after MAP [16,17]. Improvement in hardness based on the claim that the acid compound may change enamel prismatic structure, inducing a compacting effect on enamel and thus resulting in higher enamel hardness [16,17], may explain the increased VHN at the enamel surface measured in this study.

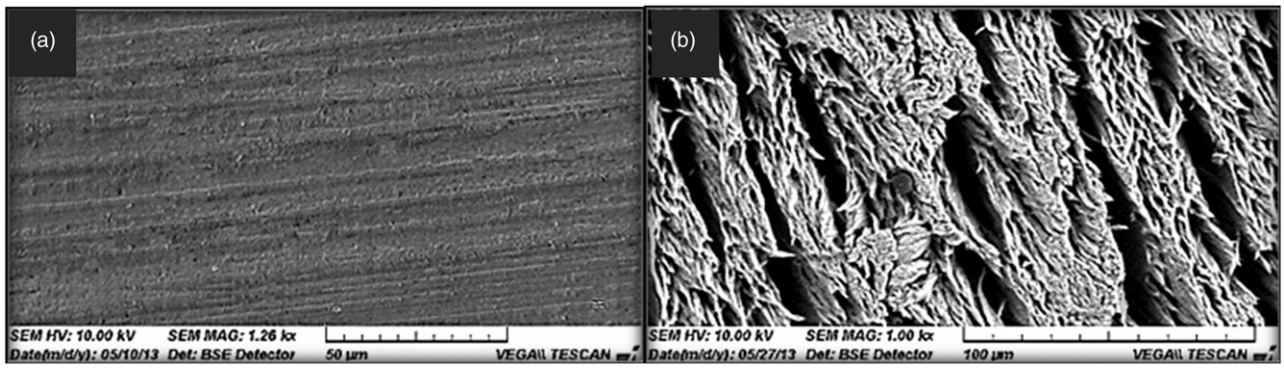


**Figure 1.** Scanning electron microscopy images for the sound and intact enamel surfaces (a) and for the enamel surface subjected to demineralization solution (b). Note the pattern at the enamel surface representing the smear layer obtained with abrasive paper in (a). Many voids and micropores can be observed at the enamel surface after initial demineralization (b).

Mineral loss of untreated lesions in the control group increased significantly after new acid attack ($p$=.006, Table 3). On the other hand, the infiltrated and microabraded lesions tended to present lower hardness after further demineralization, and this reduction was not significant in the Bonferroni *post hoc* test ($p$=.171, Table 3). Therefore, the second null hypothesis that new acid attacks do not alter the surface microhardness (2a) of incipient lesions after microinvasive treatment was not rejected.

The results of this study are, however, not in agreement with those of other authors [29]. Lower surface microhardness was found for the resin infiltrated group after the new acid challenge. By contrast, one study demonstrated reduced lesion progression compared with untreated artificial lesions after further demineralization and increased resistance against demineralization using repeated resin application [6]. Possibly, two applications of infiltrants might compensate for polymerization shrinkage and fill porosities within the lesion body and may thus improve hardness and reduce mineral loss when submitted to second demineralization [6]. This could also explain the hardness values obtained after further demineralization in this study.

The microabrasion technique used with and without polishing of enamel did not provide the same degree of improvement in surface roughness as did the resin infiltration technique used following an adhesive resin and an infiltrant (Excite F and Icon; $p$=.000). Hence, the first null hypothesis that there is no difference regarding the surface roughness (1b) of the lesions between the two microinvasive treatment techniques was rejected.

With the aim of evaluating surface characteristics, Icon-treated surface features of incipient caries lesions were compared with adhesive resin infiltrated lesion surfaces. Because Excite F was demonstrated to have better penetration behavior than other commercially available bonding agents [27,30], it was used to completely fill the artificial enamel lesions in this study. Excite F showed the best roughness results with lower Ra values than those of the resin infiltrant (Table 4). These results might be related to the small amount of resin left on the enamel surface or in some irregular regions of the etched lesion surface, as determined previously by correlating the height of infiltrated lesion surfaces with non-infiltrated lesion enamel surfaces [31]. Our results are in agreement with previous studies [31,32]. They also used Excite and Icon, and Excite was found to create considerably thicker resin coats on lesion surfaces than the other materials. The authors reported that the thicker resin layer on the surface may be explained by the low solvent content and a certain amount of nanofil particles [31]. In contrast, Icon can easily penetrate into the enamel subsurface due to its low viscosity and small contact angle, which could thus also explain the thinner resin layer on lesion surfaces measured by 3D topography images in studies [31,32].

Infiltration of incipient lesions with resin infiltrant resulted in significantly lower Ra values when compared to the surface roughness after initial demineralization. However, the roughness of infiltrated lesions was still significantly increased if compared to the roughness of sound enamel. In recent studies, the effect of resin infiltration on Sa surface roughness of subsurface lesions has been found to be minimal, and improvement of baseline surface roughness by resin infiltration has not been achieved [32,33]. Our results are in agreement with those reported in the studies. Areal roughness measurements were performed by 3D scanning microscopy in these studies, and the authors suggested that Ra values are 2D profile line parameters with limited information value, in contrast to Sa values, which are considered area parameters measuring the texture of the complete biomaterial surface [33]. Although stylus profilometry has been reported for the precise determination of the surface roughness profile [34], it has also been mentioned in recent studies that more reliable and repeatable data can be represented with Focus Variation 3D Scanning Microscope [31–33].

The effects of finishing procedures on the surface roughness of infiltrated enamel lesions were also evaluated by three-dimensional topography images of specimen surfaces. The use of finishing stripes after infiltration has not been found to be advantageous regarding surface roughness [32]. The authors suggested, based on these findings, that although resin infiltrants are capable of penetrating deeply into the porous enamel lesion, they cannot form a smooth coat on the lesion surfaces. Therefore, in clinical situations, microbial colonization of infiltrated but rough subsurface lesions will result in further enamel demineralization, caries lesion progression, and increased roughness [33]. Recently, to improve the surface properties of infiltrated lesions,

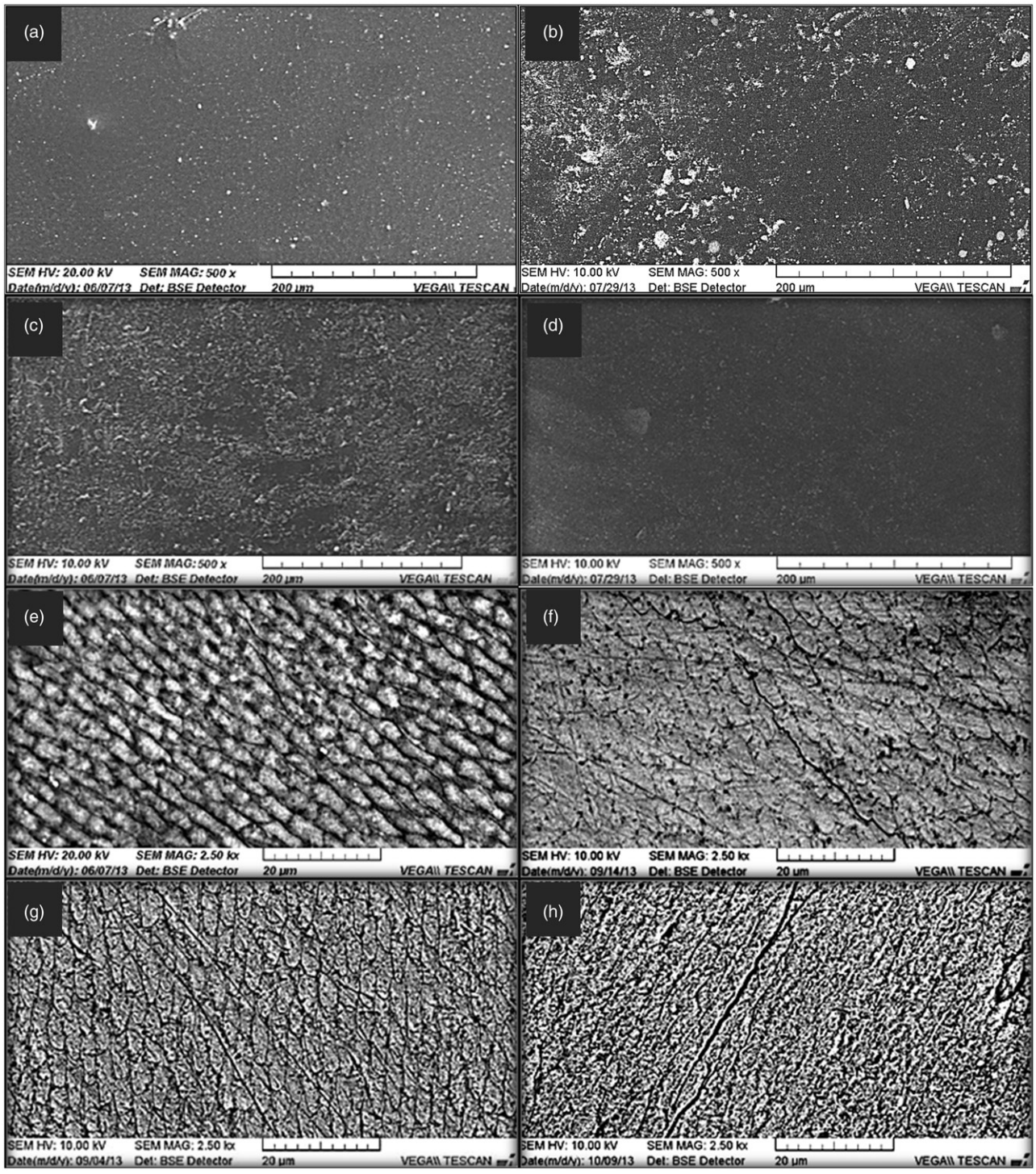


**Figure 2.** Scanning electron microscopy images, representing left (a,c,e,g) demineralized enamel specimens following infiltration or microabrasion treatment and right (b,d,f,h) respective enamel surface following further demineralization. (a,c) Show Excite F and Icon infiltrated enamel lesions, respectively. The enamel surfaces are completely hidden by a uniform resin layer. (e,g) Show microabraded enamel surfaces with a discernible enamel conditioning pattern. Polishing does not induce visible changes in the microabraded enamel surfaces (g). Further demineralization (b,d,f,h) did not clearly affect the morphology of enamel surfaces in all groups.

a two-step treatment approach using a supplemental layer of a flowable composite as a resinous top coat has been advocated [25,32,33]. Further research is needed for assessment of incipient lesions treated with the infiltration treatment approach in the long term.

In this study, the microabrasion technique decreased the roughness of the enamel surfaces that were subjected to demineralization, but the difference in sound versus microabraded teeth (with or without polishing) was statistically significant ($p$=.006). Unfortunately, no studies considering the effect of the microabrasion technique on the characteristics of incipient carious lesions by roughness analysis have been performed to date. However, compared with sound enamel surfaces, a greater amount of superficial enamel roughness was previously reported after microabrasion treatment of bovine incisors [16,17,35,36]. The authors

suggested that the chemical features of enamel microabrasion by the erosive action of the acids were responsible for the roughness effects [12,35]. Many studies have examined the potential erosive and abrasive effects of several parameters on the remaining enamel surface for microabrasion [12,37]. Regarding different compounds (combination of acids and abrasives) used for the technique, a tendency towards reduction in roughness has been reported when a final polish was performed using abrasive materials (diamond or aluminium oxide particles) to minimize the surface roughness of the enamel [16,17,36–38]. In this study, as presented in Table 4, a smoother enamel surface was obtained for the MAP group polished with diamond paste compared with that of the MA group without polishing, as reported in many other studies [16,17,36–38]. The reestablishment of the roughness of microabraded enamel surfaces followed by polishing was also confirmed by morphological analysis. The enamel surfaces polished with diamond paste were reported to have a similar morphology to that of normal enamel [16,17,39], as was not the case when the teeth with incipient caries lesions were used in this study. In spite of the polishing procedure, the morphology of enamel surfaces obtained in this study still presented an acid conditioning pattern, as they were already demineralized using a demineralization solution (Figure 2(g)).

The roughness of both infiltrated and microabraded lesions increased following further demineralization in this study ($p = .006$, Table 5). Thus, the second null hypothesis was rejected: significant alterations of surface roughness (2b) of incipient lesions were observed after new acid attacks. Despite this increase in surface roughness, both infiltration and microabrasion significantly reduced lesion progression compared with untreated control lesions ($p = .000$, Table 4). SEM images showed that both resin materials had been continuously coated on the lesion surface and did not even seem incomplete following further demineralization (Figure 2(a,c)). Infiltration with Excite F did not result in better surface roughness or demineralization resistance compared with Icon.

Statistical differences between the two microabrasion groups (MA vs. MAP) did not remain following further demineralization (Table 4), indicating that polishing did not significantly influence roughness of the lesion surfaces, which was confirmed by SEM images (Figure 2(e–h)). It has been stated that an increase in enamel roughness after microabrasion treatment can be reversed by saliva exposure [35,39]. More studies are needed to confirm the effectiveness of the microabrasion technique in clinical situations.

Considering the limitations of this *in vitro* study, it can be concluded that the Icon infiltration system and microabrasion groups resulted in similar and better microhardness than did adhesive resin, Excite F and Icon, which, although they could be considered more protective against further demineralization than Excite F, presented better surface roughness than microabrasion with and without polishing.

## Disclosure statement

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