

# Primary Tooth Enamel Surface Topography With In Vitro Argon Laser Irradiation Alone and Combined Fluoride and Argon Laser Treatment: Scanning Electron Microscopic Study

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#### Abstract

**Purpose:** The purpose of this descriptive scanning electron microscopic study was to characterize surface alterations in primary tooth enamel after in vitro argon laser irradiation alone and combined with topical fluoride treatment either before or after argon laser irradiation.

**Methods:** Twenty extracted or exfoliated primary teeth underwent soft tissue debridement and a fluoride-free prophylaxis. Buccal and lingual surfaces were determined to be caries-free by macroscopic examination (stereo-zoom binocular microscope,  $\times$ 16). Treatment groups were: (1) no-treatment control; (2) argon laser irradiation (ALI; 11.5 J/ cm<sup>2</sup>); (3) 1.23% acidulated phosphate fluoride (APF before ALI); and (4) ALI before APF. Both buccal and lingual surfaces were evaluated following standard scanning electron microscopic preparation techniques.

**Results:** With controls, enamel surfaces were relatively smooth with occasional enamel prism ends present on their surfaces. There were no areas with cavitations or surface defects. With ALI, the lased surfaces were roughened mildly to moderately irregular without cavitation of the enamel or exposure of enamel prism ends. The surfaces possessed adherent granules to globules, with most being  $<3 \,\mu\text{m}$  in greatest dimension. Only occasional fine cracks and porosities in the surface coatings were noted and these were typically less than  $1\mu\text{m}$  in width or diameter. With APF before ALI, the surfaces possessed an irregular contour, with numerous granules to globules varying in size from 1 to  $3 \,\mu\text{m}$  in greatest dimension. With ALI before APF, a homogenous confluent surface was present that masked typical enamel surface markings. The previously noted adherent granules and globules with argon laser treatment alone or APF before ALI were not seen. The argon laser effects on the enamel surfaces were masked by the uniformity of these surface coatings.

**Conclusions:** Argon laser irradiation and combined APF and argon laser treatment of primary tooth enamel created surfaces that may provide a protective barrier against a cariogenic attack. The surface coatings associated with combined APF and argon laser treatment may contain fluoride-rich calcium and phosphate mineral phases that could act as reservoirs for fluoride, calcium, and phosphate and provide a certain degree of protection from a caries lesion challenge. (*Pediatr Dent.* 2003;25:491-496)

Keywords: primary tooth enamel, argon laser, fluoride, scanning electron microscopy

Received December 19, 2003 Revision Accepted May 8, 2003

The primary dentition is at a much greater risk for caries development than the permanent dentition.<sup>1-17</sup> There are many factors that participate in this decreased resistance to dental caries development.<sup>4,8-12,16,18</sup> The composition of primary enamel is considerably different than that for permanent teeth, with a higher organic content and a lower mineral content. The time from initial demineralization of the enamel surface to the development of clinically detectable white spot lesions and frank cavitation is reduced due to the thin layer of enamel overlying the dentin of the primary tooth compared with a permanent tooth.

This may help to account for the fact that two thirds of caries in primary teeth occur on smooth surfaces, whereas only about 10% to 15% of caries lesions develop in the smooth surfaces of permanent teeth.<sup>1,2,7,13,15-17</sup> In contrast, pit and fissure caries account for 85% to 90% of lesions in the permanent dentition, while only approximately one third of caries in the primary dentition are ascribed to pits and fissures.

Relatively frequent hypomineralized and hypocalcified areas on smooth surfaces of primary teeth may in part account for increased caries susceptibility.<sup>3-7,12,16,18</sup> In addition, the rapid amelogenesis that must occur during the in utero, perinatal, and early infancy periods may also result in a lessened degree of mineralization and less caries-resistant primary tooth enamel. Feeding habits such as the use of cariogenic fluids in baby bottles, illness in early infancy with possible disruption and disturbance in amelogenesis and enamel maturation, inadequate oral hygiene measures, lack of knowledge regarding the value of the primary dentition by parents, and unavailability of dental personnel trained in infant oral health, may all contribute in some way to the increased prevalence of caries in the primary dentition.<sup>1-12,15,16</sup>

A recent laboratory study<sup>19</sup> has demonstrated improved caries lesion resistance with primary tooth enamel that was exposed to low-fluence (energy) argon laser irradiation (11.5 J/cm<sup>2</sup>, 231 mW) for a very short time period (10 seconds). Utilizing caries-like and artificial caries models, low-fluence argon laser irradiation of sound primary tooth



Figure 1. Surface morphology of primary tooth enamel. Sound primary enamel surfaces (A) are characterized by relatively frequent prism ends (arrows) that are represented by shallow surface depressions and fine porosities within these depressions.

enamel lessened the degree of enamel caries development by 40%.<sup>19</sup>

When topical fluoride treatment was performed before or after argon laser irradiation, reductions of 50% to almost 60% occurred in enamel lesion depths.<sup>19</sup> These encouraging results<sup>19</sup> prompted a study to investigate the effects of low-fluence argon laser alone and combined argon laser and acidulated phosphate fluoride on the surface morphology of primary tooth enamel.

The purpose of this descriptive scanning electron microscopic (SEM) study was to characterize surface alterations in primary tooth enamel after in vitro low-fluence argon laser irradiation alone and combined topical fluoride treatment either before or after low-fluence argon laser irradiation.

### Methods

Twenty extracted or exfoliated primary teeth underwent soft tissue debridement and a fluoride-free prophylaxis. Buccal and lingual enamel surfaces were determined to be caries free by macroscopic examination using a stereo-zoom binocular microscope at  $\times 16$  magnification. The primary teeth were then assigned to one of the following treatment groups:

- 1. no-treatment control (N=5 teeth, 10 surfaces);
- argon laser irradiation (231 mW, 10 seconds, 11.5 J/ cm<sup>2</sup>; HGM Model 5, HGM Medical Laser Systems, Salt Lake City, Utah; N=5 teeth, 10 surfaces);
- 1.23% APF treatment for 4 minutes (Oral-B Minute Gel, Oral-B Products, South Boston, Mass; N=5 teeth, 10 surfaces) before ALI;
- 4. argon laser irradiation before APF treatment (N=5 teeth, 10 surfaces).

The argon laser parameters (11.5 J/cm<sup>2</sup>, 231 mW, nonpulsed continuous 10 seconds exposure) are based on a prior in vitro study that showed a statistically significant caries inhibitory effects with low-fluence argon laser irradiation of primary teeth.<sup>19</sup> The primary tooth enamel surfaces were critical-point dried, coated with platinum and palladium under vacuum, and evaluated qualitatively for intactness, surface morphology, surface changes, and porosities in a blinded manner by scanning electron microscopy (JEOL JSM6100, JEOL USA, Inc, Peabody, Mass) at 10 kV. Ten buccal and lingual surfaces per treatment group were available for SEM examination.

# Results

The sound enamel surfaces from the no-treatment control groups were relatively smooth with frequent enamel prism ends present on their surfaces (Figure 1). There were no areas with cavitations or surface defects. The enamel surfaces were intact without surface deposits or porosities. With argon laser irradiation (Figure 2), the lased primary tooth enamel possessed irregular roughened surfaces with occasional areas of fine surface cracking and discontinuities of less than 1  $\mu$ m in width or diameter. The surfaces were



Figure 2. Treatment effect of ALI alone. ALI alone (B) leads to the formation of an irregular, mildly undulating surface with occasional surface porosities (p) and fine fissures (f) of less than 1  $\mu$ m in diameter and width. ALI alone (B) also creates fine granular-to-globular material (arrows) that lines the enamel surface, and these are <3  $\mu$ m in diameter.



Figure 4. When ALI is followed by APF treatment (D), the enamel surface has a relatively homogenous architecture with infrequent fine porosities (p) of less than 1  $\mu$ m in diameter and vague to indistinct granular-to-globular material (arrows) that are less than 3  $\mu$ m in diameter (SEM, space bars=5  $\mu$ m).

without cavitation or cratering of the enamel. Following argon laser irradiation alone (Figure 2), the surfaces possessed a granular-to-globular architecture without evidence of the enamel prism ends noted within the no-treatment control group (Figure 1). The argon lased surfaces possessed granular-to-globular irregularities that protruded above the underlying enamel surface, with most of these protrusions being <3  $\mu$ m in greatest dimension. Only occasional fine cracks, discontinuities, and porosities in the enamel surfaces were noted, and these were typically less than 1  $\mu$ m in width or diameter.

When acidulated phosphate fluoride (APF) treatment occurred before argon laser irradiation (ALI; Figure 3), the primary teeth possessed what appeared to be surface coatings that masked the underlying enamel surfaces. These surface coatings had somewhat irregular contours with numerous granular-to-globular irregularities protruding from the surface coatings. These protrusions varied in size from 1  $\mu$ m to 3  $\mu$ m in greatest dimension



Figure 3. APF treatment followed by ALI results in an enamel surface that varies markedly from no-treatment sound surface (A) and argon laser only irradiated surface (B). With APF followed by ALI (C), there appears to be a surface with fine porosities (p) of less than 1  $\mu$ m in diameter and occasional fine fissuring (f). There are also granular-to-globular deposits (arrows) on the surface that range in size from 1 to 3  $\mu$ m.

and extended above the bases of the surface coatings. The surface coatings were quite porous and had a prominent fracturing pattern with fine cracking of the surface coatings. The prominent fracturing pattern could be due to the argon laser treatment after APF exposure, but was most likely secondary to the desiccation necessary during specimen preparation for SEM examination. There was no evidence of exposure of the underlying primary tooth enamel surface.

With ALI followed by APF treatment (Figure 4), the primary tooth surfaces had what appeared to be relatively homogenous and confluent surface coatings that masked the underlying enamel surface typically seen with the no-treatment control group (Figure 1). Only infrequent, loosely adherent surface granules and globules were present (Figure 4) in contrast to the frequent granules and globules protruding above the surface coatings seen with ALI alone (Figure 2) and combined APF treatment followed by ALI (Figure 3). There were also infrequent small isolated porosities without fracturing of the surface coatings (Figure 4).

#### Discussion

The findings in the present study provide interesting insights into the effects of low-fluence ALI alone and in combination with APF treatment. The enamel surfaces of the primary teeth were markedly altered by the experimental treatments. Of particular interest was the lack of crazing, cratering, and exfoliation (sloughing of layers of surface enamel) that are typically found with other types of lasers that utilize high fluences (energies), such as with  $CO_2$  lasers.<sup>20-22</sup> Also, the tissue loss or cutting potential of Ho:YAG lasers did not occur.<sup>20-22</sup> The surface morphology changes with low-fluence ALI more closely mimic those reported with the argon fluoride excimer laser, which produces quite fine surface porosities with irregularly packed reprecipitation of enamel crystalline material.<sup>20-22</sup> The argon lased enamel surfaces possessed

granular-to-globular adherent material of relatively small size (1-3  $\mu$ m).

Previously, similar structures have been noted with lowfluence argon-lased permanent enamel; and based upon morphologic features, these granular-to-globular structures have been considered to represent redeposited mineral phases due to the mobilization of calcium, phosphate, and fluoride from the lased enamel.<sup>20</sup> In fact, some of the globules share some of the morphologic features typically ascribed to calcium fluoride.<sup>20,21,24-26</sup>

The presence of adherent calcium, phosphate, and fluoride-rich phases<sup>20,23-25</sup> on low-fluence argon lased tooth surfaces would help to explain the effects of ALI on in vitro caries lesion formation in permanent enamel, primary enamel, and root surfaces.<sup>26-33</sup> Over the past decade, ALI alone results in a 25% to 35% reduction in in vitro lesion depths, compared with primary and permanent enamel and root surfaces that have not received the benefits of argon laser exposure.<sup>26-33</sup> Recent in vivo pilot studies<sup>34-36</sup> have confirmed the in vitro findings of substantial reductions in lesion depths and areas when enamel surfaces have been exposed to an argon laser for as little as 10 seconds at low fluence (250 mW). In one clinical trial,<sup>35</sup> caries lesions developed in only 12% of the teeth that had been lased, compared with 100% of control teeth that had not received ALI. This beneficial, caries-resistance effect occurs while maintaining an intact surface, as previously noted by polarized light and SEM.<sup>26-33</sup>

The addition of APF treatment prior to argon lasing of primary tooth surfaces produced enamel surfaces that appeared different from those that were exposed to APF after ALI. Both masked the enamel surfaces of the primary teeth. With fluoride application prior to argon lasing, the resulting enamel surfaces had a relatively high degree of porosities and globular-to-granular material embedded in an amorphous background. The globular component composing the surfaces appeared similar to the globular precipitates seen with ALI alone. In contrast, the surface coatings with fluoride treatment after argon lasing provided a confluent surface with only a mildly irregular morphology. There were hints of globular deposits embedded within and covered by a more homogenous surface coating-like material. The creation of the surface coatings and globular and granular adherent material with APF treatment in conjunction with low-fluence ALI is most likely due to the acidic nature of APF and its effect on enamel solubility.<sup>21-25</sup>

During APF treatment, a thin layer of the surface enamel may become solubilized and the resultant mobilized mineral phases undergo reprecipitation on the enamel surface as fluoride-rich calcium and phosphate mineral phases (fluoridated dicalcium phosphate dihydrate, fluoridated octacalcium phosphate, fluoridated hydroxyapatite).<sup>21-25</sup> The predominant precipitate with APF is calcium fluoride, which may be maintained in the imbrication lines of enamel surfaces for several weeks to months and allow for prolonged release of fluoride to the tooth surface.<sup>21-25</sup> In previous laboratory studies, <sup>29,31,32,37</sup> the combination of ALI and APF treatment consistently reduced lesion depth by 40% to 60% in both in vitro enamel and root surfaces caries models, while maintaining intact surfaces as noted by polarized light and SEM. Similar results have been reported with primary tooth enamel as well.<sup>19</sup> Whether fluoride treatment precedes or follows argon laser exposure has not provided any difference in caries resistance.<sup>19,29,30,32,37</sup> It would appear that both types of surface coatings noted with the 2 combinations of fluoride and argon laser treatment in the present study have been shown previously to provide equal resistance for the treated enamel surfaces from a cariogenic attack. Similar findings have now been documented with a clinical trial.<sup>34</sup>

A greater than 60% reduction in in vivo lesion depth was achieved when intraoral fluoride (neutral sodium fluoride) treatment occurred before ALI, compared with no-treatment, control-matched teeth in the same patients. In this clinical trial,<sup>34</sup> there was also a significant increase in resistance (32% reduction in lesion depth) to caries development with combined fluoride treatment and argon laser exposure compared with ALI alone.

Although ALI and its relationship with caries reduction both in the laboratory and in clinical pilot studies have been shown, the mechanism for increasing caries resistance has not been determined. It has been suggested that the laser treatment creates fine microporosities within the tooth substance.<sup>20-22,28,32,38</sup> A microsieve network is formed that would effectively trap and precipitate mineral phases mobilized from the subsurface enamel and root surface during a cariogenic attack. Interestingly, quantitative polarized light microscopic techniques have shown that such a microsieve network exists in lased enamel.<sup>38</sup>

The combination of fluoride and laser treatment affects the critical pH at which enamel undergoes dissolution. The critical pH of sound enamel (pH 5.5) is reduced by 5-fold following laser irradiation (pH=4.8) and by a further 6-fold when fluoride is used in combination with lasing (pH=4.3).<sup>39-41</sup> Although high fluence was not employed in the present study, it is possible to completely inhibit lesion formation in the absence of fluoride when a laser fluence of 170 J/cm<sup>2</sup> is used.<sup>21,22,41</sup> In the presence of low levels of fluoride (0.2 ppm), complete inhibition of caries formation occurs with a laser fluence of 85 J/cm<sup>2</sup>. The benefit of combining fluoride treatment and argon laser exposure is readily apparent.

#### Conclusions

Argon laser irradiation and combined APF and argon laser treatment of primary tooth enamel create enamel surfaces that may provide a protective barrier against a cariogenic attack. The altered primary tooth enamel surfaces associated with combined APF and argon laser treatment may contain fluoride-rich calcium and phosphate mineral phases that could act as a reservoir for fluoride, calcium, and phosphate and provide a certain degree of protection from a caries challenge.

# Acknowledgement

This study was supported by an American Academy of Pediatric Dentistry Foundation Research Grant.

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#### Abstract of the Scientific Literature

# GLASS IONOMERS AS SEALANT AND CARIES PREVENTIVE MATERIALS

This study evaluated the retention and caries-preventive effectiveness of a resin-modified glass ionomer cement (Vitremer) and a conventional glass ionomer cement (Ketac Bond) as fissure sealants and the association between 2 caries risk variables and caries incidence after 36 months. The sample consisted of 208 children (6-8 years old) with 4 intact and unsealed permanent molars. Caries prevalence was assessed at baseline, and children were randomly divided into experimental (100-400 teeth) and control (108-432 teeth) groups. The experimental group received a professional sealant application in a split-mouth design. The control group were observed only. Oral hygiene instructions and plaque control were provided at each recall examination after 6, 12, 24, and 36 months. Very low sealant retention was observed after 3 years of application, with a statistically significant difference for retention rates between Vitremer and Ketac Bond. No statistically significant correlation was found between caries incidence and previous caries experience for the experimental groups. Children in the control group with caries experience during the baseline examination showed a probability in developing caries in the first molars 4.2 times higher compared to those without caries experience. In the experimental and control groups, presence of incipient caries was statistically associated with caries incidence in the first molars. Although glass ionomer cements showed low retention, prevention of dental caries in the sealed first molars was observed. The authors concluded that Vitremer was effective to prevent occlusal caries lesions.

**Comments:** This well-designed protocol provided an excellent tool for caries-risk assessment facilitating the clinical decision of sealant application. **MG** 

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