Morphological changes in hard dental tissues prepared by Er:YAGLaser, Carisolv and rotary instruments

A scanning electron microscopy evaluation

Introduction

In recent years, prevention and early caries detection, as well as changes in the understanding of chemical and biological basis of demineralisation process in hard dental tissue and possibilities carious lesion to undergo remineralisation supersede the classical operative approach of caries treatment postulated by G. V. Black and promoted minimally invasive preparation (MIP). The main categories of MIP techniques include rotary handpieces and burs, chemo-mechanical cleaning with Carisolv gel, air abrasion and dental lasers. 1,2 These trends for the replacement of the conventional method of preparation led to focus the attention of researchers on the impact of alternative techniques for MIP on hard dental tissues and underlying dental pulp. MIP techniques claim to be able to achieve controlled removal of infected and softened dentin while preserving healthy the hard dental tissues and do it with minimal discomfort for the patient. However, currently available data provide contradictory evidence for the impact of alternative techniques of MIP on hard dental tissues compared to conventional preparation. Possible reasons for this are the variety of experimental studies and difficulties to standardise the results of clinical researches. It is worth noting that the researchers giving the most positive evaluation of the alternative methods of preparation (Carisolv, laser) use mainly clinical criteria for evaluation (perception and tolerance of the patient, noise, atraumatic work, colour and texture of the dentine when probing, etc) which are all rather subjective. New improved versions of alternative systems for preparation have been made available on the market claiming to be highly clinically efficient, but there is still little information about them (the modified Carisolv colourless gel, multi-frequency high-energy lasers, air-abrasion). This makes it necessary that research in this rapidly developing, promising field of dentistry should be periodically updated.

The objective of the present in vitro study was to evaluate by SEM the ultrastructural changes in hard dental tissues treated with several alternative systems for caries removal and preparation: Er: YAG laser (LiteTouch), chemomechanical preparation with Carisolv gel, conventional preparation with diamond burs/air turbine and steel burs/micromotor.

Fig 1a



Fig 1b



Fig 1c

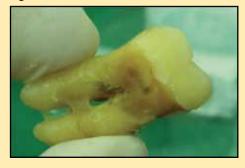


Figure 1 a, b, c: Extracted teeth with carious lesions

Materials and methods

Experimental design: the study used 20 human teeth freshly extracted because of advanced periodontal disease. The preparations involved natural carious lesions on tooth surface (Fig. 1a, b, c).

According to the preparation technique the teeth were divided into 4 groups of 5 teeth (n = 5):

- Group 1. Laser preparation by Er: YAG laser (LiteTouch, Syneron, Israel) (Fig. 2a, b, c)
- Group 2. Chemomechanical preparation with Carisolv colourless gel (MediTeam AB, Savedalen, Sweden) (Fig. 3a, b, c)
- Group 3. Mechanical rotary preparation by diamond burs/air turbine
- Group 4. Mechanical rotary preparation by steel burs/micromotor

Preparations are made strictly according to manufacturer's instructions for service.

The removal of caries is proved by clinical methods – observation and probing. After preparation the teeth are immersed for 1 hour in 4% buffered fixative solution of glutaraldehyde (0.075 M, pH 7.3). Then they are rinsed in distilled water and placed for 90 min in cold buffer solution of sodium cacodylate (0.02M, pH 7.2, 660 mOsm) for fixation of organic matter. Subsequent dehydration is carried out in ethanol in ascending series of 30%, 50%, 70%, 80%, 95% and 100% for one hour in each series, the drying of the teeth based on CPD (Critical Point Drier) method in a desiccator. The dried specimens are mounted on a metal stand and gold-coated (200-250 nm) by cathode atomisation under vacuum. Scanning microscopy is performed with the electron microscope of Philips (Holland) 515 model SEM with accelerating voltage of 25 kV in secondary emission mode. For each specimen we made five photographs with the same magnification (x2000) of randomly chosen areas and different numbers of photos at a different degree of magnification.

On the SEM photomicrographs we evaluated, described and compared the morphological findings and differences in the enamel and dentin tissues after treating the teeth using alternative methods for caries removal and cavity preparation.

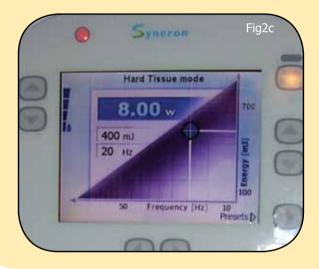
Results

When analyzing the SEM photomicrographs of the specimens examined, it is found that the conventional method of cavity preparation with steel burs and micromotors at low speed without water cooling (group 4) results in contaminated surface covered with smear layer of dentin debris without visible dentinal tubules orifices (Figs. 4a, 4b). Thick smear layer covers all treated surfaces. The walls of the cavities are smooth and rounded and the border between enamel and dentin is hardly noticeable.





Figure 2 a, b, c: Laser preparation with Er: YAG laser LiteTouch (Syneron, Israel) "Hard tissue mode" (400mJ/20Hz; 8.00W)



In group 3 (preparation with diamond burs, air turbine and water cooling) a thin, smooth and in some places missing smear layer was observed (Fig. 5a). In the area of water turbulence there were patent dentinal tubules orifices, but without having a clear outline of both tubules lumens and peri- and inter-tubular dentin (Fig. 5b). The boundary between enamel and dentin is unclear and the cavity forms have smooth contours.

The dental surface topography after chemomechanical preparation with Carisolv gel (group 2) was clearly rougher compared with that of groups 1 and 2, the dentinal tubules orifices are visible and there is almost no smear layer (Fig. 6a). Preparing the organic matrix using chemomechanical preparation with Carisolv and protecting mineralised dental tissue at the same time results in a rough appearance of the treated surfaces and considerable microretention development (Figs. 6b, 6c). Denatured collagen fibers and surface contaminations occur in some places, blocking the

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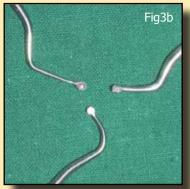




Figure 3 a, b, c: Chemomechanical preparation with Carisolv colourless gel and hand excavators

dentinal tubules orifices (Fig. 6d). Cavity forms in Carisolv group follow the initial caries lesions forms without going beyond their boundaries.

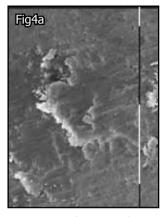
Cavity forms prepared with Er:YAG laser (Group 1) are characterised by a lack of definite and precise geometric configuration and outlined cavity elements (Fig. 7a). There is rough and irregular surface without no smear layer (Fig. 7b). Dentinal tubules are clearly exposed. Intertubular dentin is ablated more than peritubular dentin and that made dentinal tubules appearance more prominent (Fig. 7c). In the enamel the typical architectonics of enamel prisms grouped in bundles is observed. Laser ablation of part of the enamel makes the surfaces highly strong retentive (Figs. 7d, 7e).

Discussion

The philosophy of minimally invasive cavity preparation approach is based on several principles – to remove only irreversibly damaged dental tissues and to avoid macroretention preparation in healthy tissues. Additionally, these techniques should protect the underlying pulp and leave the treated surface suitable for adhesive bonding. Antibacterial effects of the alternative preparation techniques must not be lower than those of standard necrotomy with rotary instruments and even to excel them.

Nowadays the laser devices available for clinical use are capable of effective, controlled ablation of hard dental tissues.² Some clinical trials suggested Carisolv gel to be highly efficient in caries removal, leaving clean and retentive dentinal surfaces.² However, not all researchers agree with these conclusions. Therefore, such studies should be periodically updated due to constant introduction of new technologies.

The experimental results of the present study revealed significant differences in the surface morphology of the studied samples which would affect the ability to perform effective adhesive bonding. These morphological



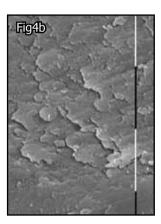
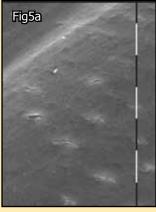
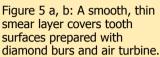


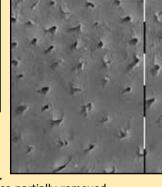
Figure 4a,b: SEM photomicrographs of tooth surfaces prepared with steel burs. The surface is covered with a layer of debris, dentinal tubules orifices are not visible (x500, 2000)

differences are highly dependent on the mechanism of action of the specific preparation systems.

Laser devices use a variety of physical media as sources for generating different wavelength that is absorbed and interact with specific molecules in human tissues. The explanation for the hard tissue ablation is the water content that evaporates when exposed to laser irradiation creating high internal pressure and subsequent microexplosions. In this interaction of the laser irradiation with tissue if there is inadequate water cooling that will lead to undesirable thermal effects.3 Depending on parameters such as pulse energy and frequency carbon dioxide lasers, Nd: YAG and Er: YAG lasers cause changes in enamel and dentin as roughing, craters, cracking, slicing, carbonification, melting and recrystallisation described in many previous studies. 4-6 These changes depend on the laser type, mode of operation, system for water cooling and proper operation.3 Additionally, the abilities to ablate carious dentin and enamel strongly vary according to different experimental studies.4-6 For argon-fluoride laser







Fiq5b

In the area of water turbulence partially removed contaminants and single dentinal tubules lumens were observed (x500, 2000)

and the exciter laser there are data on their ability to remove dental caries which is not of sufficient efficiency.⁵ Krypton fluoride exciter laser emitting in ultraviolet range has been shown to remove dentin, but enamel resists the attempt for ablation.⁵

The high-power and high-frequency Er: YAG laser (LiteTouch, Israel), which was used in the present study, has an advanced hydrokinetic system that is said to be capable of effective and safe ablation of hard dental tissues. LiteTouch laser incorporates unique software which allows for the broadest range of energy and frequency settings. The unique LiteTouch hand piece prevents loss of energy and along with the precision control over pulse duration, pulse energy and repetition rate optimise, allows for a wide range of hard tissues procedures. LiteTouch is the first laser in the world to use a novel mechanism that controls energy output, offering optimal control of treatment parameters. Easy adjustable water spray flow, frequency and energy levels on a touch screen following special software. LiteTouch is also innovative in respect of its optical system incorporated in the ergonomic hand piece working with sapphire tips. The proposed mechanism of action of this system is the photons radiation that laser source delivers in targeted air - water jet, resulting in water droplets microexplosions. It is believed that this process is the mechanism of ablating particles from dental tissues without overheating, and without smear layer formation.⁷ Another characteristic of this laser is the wavelength (2940 nm) which is absorbed predominantly by water and also the sapphire tips, showing stability in providing focused energy of laser irradiation.8 This combination allows precise microinvasive cavity preparation with minimal heating and optimal rate of radiation absorption by the hydroxylapatite incorporated water.7 The program "hard tissue mode" removes enamel, dentin and dental caries effectively and without visible carbonisation or disturbance of the specimen microstructure. Evaluated under electron microscope the dental tissues treated with Er: YAG laser showed rough and

irregular surface without presence of a smear layer and with patent dentinal tubules. Intertubular dentin is ablated more than peritubular giving a characteristic appearance of the dentinal surface with mild prominent dentinal tubules. Enamel shows preserved prismatic structure, but also strong retentions due to microexplosions on its surface. Overall, the cavity form is irregular, devoid of strict geometry and dotted with microretentions, but without presence of contaminants or smear layer. The observed changes correspond to changes in hard dental tissues reported by other authors in previous studies on Er: YAG lasers^{9,10}, but without thermally affected surfaces, areas of extensive recrystallisation, melted surfaces or cracks in the dentin, as described in some in vitro studies.³⁻⁵

It has also been reported that there are better abilities for adhesive bonding¹¹, faster ablation of enamel and dentin compared with rotating burs¹² and an increase in dentinal microhardness after treatment with Er: YAG pulsed lasers¹³. The latter statement is not confirmed by other studies. The marked surface irregularities and lack of smear layer observed in the recent study, noted



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Fig6a

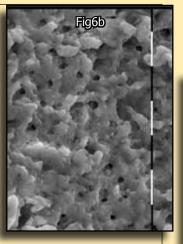
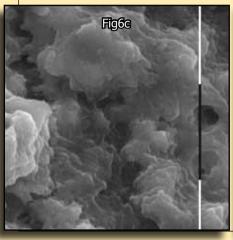
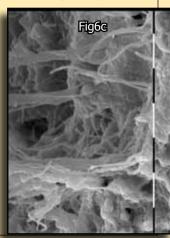


Figure 6 a, b: Dentin surfaces treated with colourless Carisolv gel - clean and highly retentive, with great part of exposed open dentinal tubules (x500, 2000)

Figure 6c, d: Dentin surfaces treated with colourless Carisolv gel there is a rough, granular surface which is highly retentive. In some sections single collagen fibrils are evident (x 3000)





also in other studies^{14,15} provide solid evidence for the physical mechanism of bonding with composite materials after laser treatment¹¹. This fact is not yet fully explored as a possible opportunity to eliminate acid etching of hard dental tissues and its related adverse effects on the underlying dentin and pulp.

Carisolv is a chemo-mechanical, minimally invasive method for selective softening of caries in dentin and its subsequent removal with hand excavators. 16 The system consists of gel containing three amino acids (glutamine, lysine and leucine) and transparent liquid (0.5% NaOCI), which are mixed immediately before application. The obtained chlorinated amino acids selectively tear the damaged collagen fibers in carious dentin without damaging the underlying demineralised but not denaturated collagen. The macerated infected dentin is removed manually with the help of excavators. Carisolv gel used in this study is colourless and concentration of the amino acids in it is twice as small, while the sodium hypochlorite concentration is increased twofold.

The suggested mechanism of action of Carisolv gel is based primarily on the proteolytic effect of NaOCI, which dissolves the denaturated collagen in the caries lesion. 16 It is assumed that the three amino acids enhance the effect of NaOCI on the collagen and also reduce the involvement of healthy dental tissues. Carisolv chemical effects on the underlying pulp have been assessed as safe and alkaline pH (~ 11) of the gel neutralises acids and has a bactericidal effect on cariogenic flora. 1,16 The presence of NaOCI in Carisolv is problematic, however, because of the danger of NaOCI inhibiting the bonding agents polymerisation. Another clinical problem is the inability of Carisolv to affect the enamel and that requires a combination with rotary instruments in the process of caries excavation. 16 Additionally, the results as reported by studies on Carisolv capacity to remove the smear layer vary. According to some authors Carisolv almost completely removes the smear layer remaining visible and patent dentinal tubules. 15,17 According to other authors Carisolv is unable to eliminate smear layers and there are no patent dentinal tubules. 18 The latter study was conducted on non-carious dentin surface the researchers observing an irregular smear layer over enamel and dentin, and all dentinal tubules orifices filled with debris.

A third group of researchers take an intermediate position – according to them Carisolv does not eliminate completely the smear layers, and there are partially patent dentinal tubules and residues of contaminant smear layer covering the dentinal surface.²

The dentin surfaces treated with Carisolv and observed by SEM in the present study are clean, smear layer-free with some remnants of denatured collagen fibers. Conventional rotating burs form smear layer on dental surface while Carisolv increase the surface roughness, leaving a relatively clean area. The dentin topography after Carisolv application is granular and rough compared to preparation with rotating instruments and possesses comparable roughness similar to that observed after laser preparation. The marked structural changes of the dental tissues and surface roughness observed in our study may play a crucial role in adhesion to composite materials, possibly without using etching agents.

However, literature data on structural changes after Carisolv preparation vary considerably and we can conclude that this system for chemo – mechanical removal of dental caries is likely sensitive to the technique of application, mineralisation and other dentin characteristics.^{2,19}

The results of some contemporary studies showed that despite differences between individual authors, generally the amount of smear layer after treatment with Er: YAG laser and Carisolv in all cases is less than that after

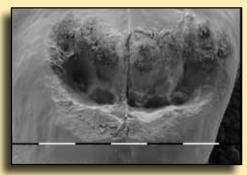


Fig7a

Figure 7a: A cavity prepared with Er: YAG laser - unclear cavity outlines, craters shading into one another are observed. There are no precise outlined cavity elements (x20)



Fig7e

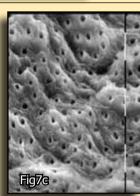
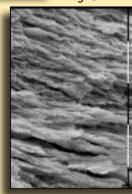


Figure 7b, c: Laser treated dentin. The surface is clean and free from debris, all dentinal tubules are open. The surface is irregular, rough, which creates high retentiveness. At greater magnification the more effective removal of intertubular dentin is seen and that makes dentinal tubules orifices appear convex (x500, 2000)

Fig7d & Fig7e: Figure 7d, 7e: Enamel surfaces treated with Er: YAG laser revealed characteristic architectonics of bundles of enamel prisms with different orientation. The surface is highly retentive and free from contaminants and smear layer (x2000, 500)



preparation with conventional rotating instruments, and surface changes are characterised by markedly rugged topography.^{2,3,12,15}

The morphological features of hard dental tissues observed in our study lead us to the general conclusion that cavity preparation with Er: YAG laser and Carisolv is consistent with the principles of minimally invasive preparation, leaving clean surfaces and strong microretentions suitable for adhesive restorations. These assumptions about the benefits of alternative techniques for minimally invasive preparation of dental tissues for adhesive restorations should be confirmed in future clinical studies.

Conclusions

Fig7b

SEM analysis of hard dental tissues treated with steel and diamond burs showed surfaces covered with a thick layer of debris, which could compromise the adhesion of filling materials. Dental tubules orifices are obturated with

debris, with exception the areas under water turbulence where the debris is partially removed.

Carisoly gel does not affect the enamel or healthy dentine. The surface topography of the dentine remaining after complete caries removal with Carisolv is rougher than that after conventional preparation with rotating burs. No typical smear layer is observed but thin patches of contaminants, much less prominent than after drilling are visible.

All laser-treated samples showed no evidence of thermal damage or signs of carbonification and melting. The SEM examination revealed characteristic microirregularities of the laser prepared dentin surface without any smear layers, and open dentinal tubules. Intertubular dentin is ablated more than peritubular dentin and that made the dentinal tubules appearance better exposed. Er:YAG laser ablated enamel effectively leaving well exposed enamel prisms without debris. The surfaces are very retentive. DA

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