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ORIGINAL ARTICLE
ORIGINALNI RAD

DOI: 10.1515/sdj-2015-0003
UDC: 615.46:616.314-7 : 616.31-002-085

Effect of Carisolv Gel on Sound, Demineralized and Carious Dentin: In Vitro Study

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SUMMARY

Intoduction Pain and discomfort during dental interventions caused by high-speed dental burs are the most frequent reasons for avoiding dental visits. Numerous possibilities have been offered in the last decade aiming to replace burs for caries removal, one of them is chemo-mechanical method of caries elimination. The aim of this study was to analyze micromorphological changes caused by Carisolv on sound, demineralized and carious (softened) dentin using SEM.
Material and Methods Study included 30 teeth (20 intact and 10 with carious lesions) extracted for orthodontic reasons. Carisolv™ system (MediTeam, Sweden) that contains Carisolv gel and specific nickel-titanium hand instruments was used for chemo-mechanical caries elimination. In our study only Carisolv gel was applied on sound, demineralized and carious dentin for 20 min. Dentin surface was analyzed using scanning electronic microscope (SEM).
Results SEM showed that Carisolv gel affected soft carious dentin only with no changes on sound dentin regardless of demineralization status.
Conclusion Carisolv did not affect sound and demineralized dentin while it selectively dissolved carious dentin.
Keywords: Carisolv; sound dentin; carious dentin; demineralization; SEM

INTRODUCTION

In line with development of contemporary technologies, cavity preparation techniques and restorative materials have been progressively upgraded and improved. Nevertheless, the preparation technique and caries elimination that have been the most popular for years is by using high-speed burs. The invasiveness of this preparation method is the reason for many adverse effects on dental tissues. High-speed burs can brake enamel prisms, not only on the edges of cavities, but deeper in the intact enamel. This cracks caused by broken enamel prisms are areas were bacteria and their toxins can penetrate further in dentin and cause carious lesions [1]. Also, burs are not selective and in addition to carious removal a part of sound dental tissue is removed. Thus, the tooth walls become weak and possibility for breaking tooth becomes greater [2].

Taking into consideration negative consequences that may occur after using high-speed burs, sound dental tissues should be preserved while preparation techniques and caries elimination should be minimally invasive [3-8]. Atraumatic restorative treatment has been proposed by the World Health Organization [9]. In the last years the most favorable preparation techniques and methods of caries elimination have become those with minimal pain and unpleasant sensations, increased efficiency and comfortable treatment for patients.

The efficacy of chemo-mechanical method of caries elimination with the use of Carisolv, as an alternative to burs has been shown in the literature. It has also been shown that patients described treatment more pleasant and less painful than the classical method of caries elimination [10]. Carisolv has been demonstrated highly selective for carious dentin however its effect on sound and demineralized dentin has not been studied yet.

The aim of this study was to analyze using SEM micromorphological changes of sound, demineralized and carious (soft) dentin caused by Carisolv.

MATERIAL AND METHODS

Carisolv™ system (MediTeam, Sweden; Carisolv gel and specific nickel-titanium hand instruments) was used for chemo-mechanical caries removal. Carisolv gel is a mixture of two gels. One contains 3 amino acids (leucin, lysine and glutamic acid), NaCl, NaOH, erythrosine and distilled water. The other gel is 0.5% NaOCl. When both are mixed an isotonic alkaline gel with pH 11 is created. The action of active substance is limited to 20–30 minutes.

Hand instruments are designed in a way that both sides have working tips with diameter of 0.3 to 2 mm. They are made of titanium and can be found in different sizes. Instruments are used to apply gel in carious lesion and

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to remove soft carious dentin while sound tissue should be retained. Working part of instrument has 2-8 cutting edges, which enables activity in several directions. Sharp edge of the instrument removes caries at an angle of 90° providing better in-depth control.

For the SEM analyses, 20 intact and 10 teeth with carious lesions with no pulp involvement and extracted due to orthodontic or prosthetic reasons were used. After extraction teeth were cleaned and assigned into the three groups. All teeth were placed on an evaporation device and enamel was removed in intact teeth by breaking. In 10 out of 20 specimens Carisolv gel was applied on sound dentin, with no prior demineralization. In the remaining 10, after demineralization with phosphorous acid (25-30 seconds) Carisolv gel was applied on demineralized dentin. In the third group of 10 carious teeth Carisolv gel was applied directly on carious lesions. The specimens were exposed to Carisolv for 20 minutes without the use of mechanical excavation.

After 20 min changes on dental surfaces were analyzed using SEM. The specimens were attached to the abutments, placed in the evaporator and coated with gold for SEM analysis (Scanning electronic microscope, JEOL JSM-5300, scientific-research center, Faculty of Dentistry in Nis, Serbia).

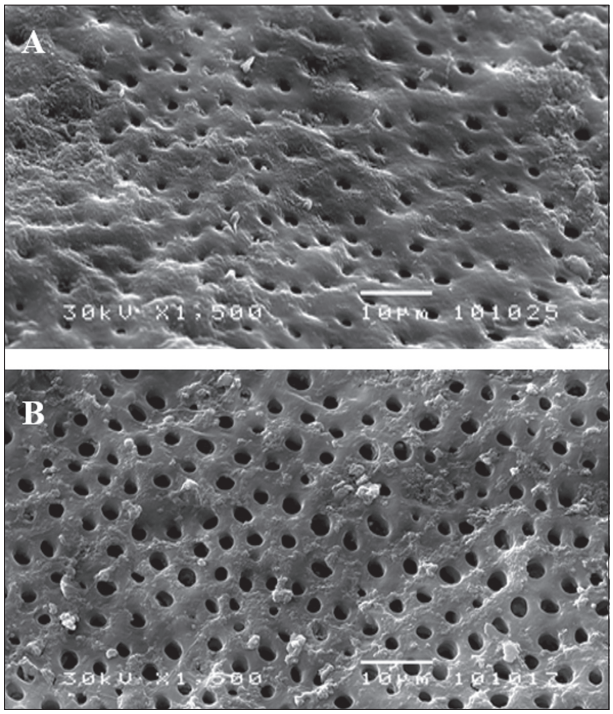

Figure 1. A) Sound dentin- non treated (1,500×); B) Sound dentin after 20-minute exposure to Carisolv (1,500×)
Slika 1. A) Zdrav, netretiran dentin (1500×); B) Zdrav dentin nakon 20 minuta izlaganja gelu *Carisolv* (1500×)

RESULTS

Table 1 shows specimen distribution per groups (sound, carious and demineralized dentin). SEM showed no significant difference in the structure and dentin surface between teeth where Carisolv gel was applied on sound dentin and those with no treatment (Figures 1a and 1b).

In the second group where teeth were treated with phosphorous acid, demineralization of dentin surface leaving 3-5 μm firm layer on compactly set collagen fibers was noticed (Figure 2a). After 20-minute exposure of demineralized dentin to Carisolv gel neither micromorphological changes nor changes in the firmness of this layer were noticed (Figure 2b).

In the third group of teeth, after the application of Carisolv gel on carious dentin, SEM micrographs showed that

Table 1. Distribution of specimens used for SEM analysis
Tabela 1. Raspodela uzoraka korišćenih za SEM analizu

Tooth type Grupa zuba	Sound teeth Zdrav dentin *Carisolv*		Sound teeth Zdrav dentin *H_3PO_4 + Carisolv*		Carious teeth Karijesni dentin *Carisolv*	
	N	%	N	%	N	%
Incisors Sekutići	1	10.0	/	/	2	20.0
Canines Očnjaci	3	30.0	1	10.0	/	/
Premolars Pretkutnjaci	4	40.0	5	50.0	3	30.0
Molars Kutnjaci	2	20.0	4	40.0	5	50.0
Total Ukupno	10	100.0	10	100.0	10	100.0

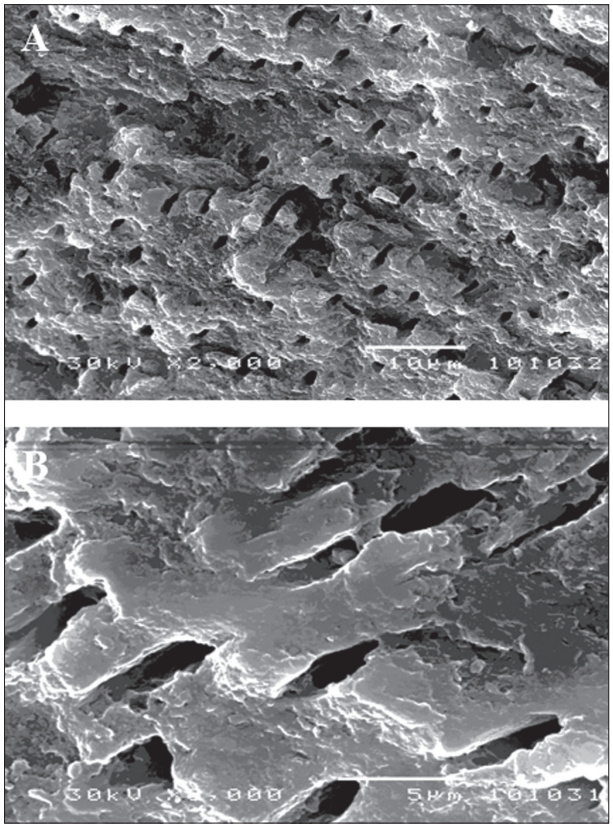

Figure 2. A) Dentin surface after 30 sec exposure to H_3PO_4. Impaired collagen structure due to demineralization is noticeable (2,000×); B) Demineralized dentin after 20 minutes exposure to Carisolv (5,000×)
Slika 2. A) Površina dentina nakon 30 sekundi izlaganja fosfornoj kiselini. Oštećena struktura kolagena je vidljiva (2000×); B) Demineralizovani dentin nakon 20 minuta izlaganja gelu *Carisolv* (5000×).

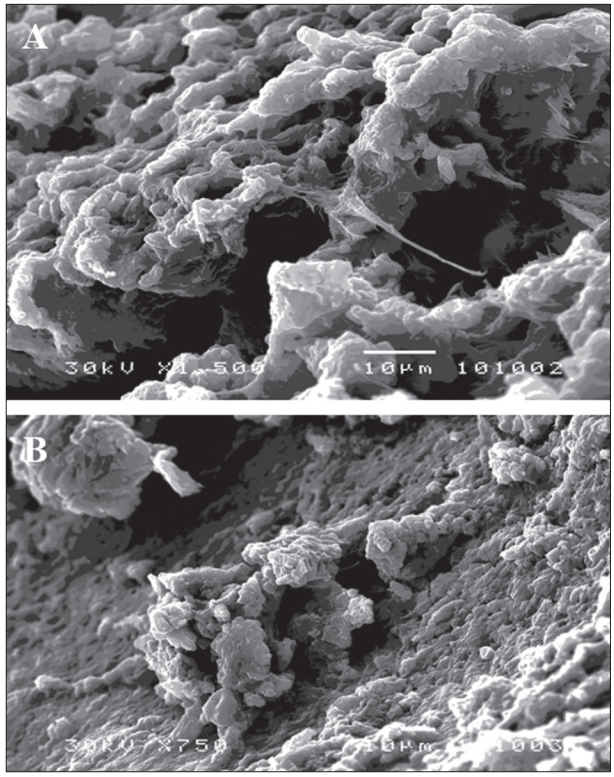

Figure 3. A) Carious dentin, denatured, collagen fibers (1,500×); B) After 20 minutes exposure to Carisolv, denatured dentin was partially dissolved and separated from sound dentin (750×).
Slika 3. A) Karijesno izmenjeni dentin, denaturisana kolagena vlakna (1500×); B) Nakon 20 minuta izlaganja gelu *Carisolv*, denaturisani dentin je delimično rastvoren i odvojen od zdravog dentina (750×).

Carisolv gel detached only carious dentin and enabled its easier removal with hand instruments (Figure 3a). In addition, SEM micrographs showed that dental surface under demarked carious dentin was unchanged indicating that Carisolv gel did not change sound dentin (Figure 3b).

DISCUSSION

The development of systems for chemo-mechanical caries removal most likely originates from patients' desire for painless caries removal, without the use of burs and with less unpleasant sensations during work [11]. These are also perhaps the most frequent reasons for avoiding visits to dentist and regular check-ups.

In 1998, Carisolv system consisting of Carisolv gel and hand instruments with nickel-titanium working part was introduced to the market. Carisolv gel works by direct action of NaOCl and three natural amino acids on carious dentin [7]. Painless caries removal without previous application of local anesthesia is great advantage of the chemo-mechanical method. Selective action of Carisolv gel on carious dentin, lack of thermal stimulations, pressure and vibrations created during mechanical preparation are great advantages of this system. Application of isotonic Carisolv gel on dentin does not cause thermal and osmotic stimulations of pulp since it has body temperature [5]. Sharp edges of hand instruments specific for carious dentin only further contribute to painless treatment [12]. However, it has been noted that for the treatment of initial carious lesions chemo-mechanical method of caries elimination was not suitable [13]. On the other hand due to its selective action on carious dentin this method reduces the risk of iatrogenic pulp perforations and pushing carious dentin in pulp. Dammaschke et al. [14, 15] showed that gel does not penetrate through the sound dentin and therefore there is no risk of damaging pulp tissue. It is important to carefully select cases for the treatment using Carisolv system.

The main purpose of our research was to find out whether Carisolv gel affects selectively soft and carious dentin, without damaging surrounding sound dentin. SEM is frequently used method for studying dentin morphology [4]. Pseudo three-dimensional image for topography and content of observed surface is also available [16]. Our study analyzed Carisolv gel effects on fractured dentin surface because changes caused by chemical agents are less visible if dentin surface is covered with smear layer. After 20–minute exposure of sound dentin to Carisolv gel no significant micromorphological changes were noticed. Exposure time was limited to 20 minutes due to manufacturer's recomendation.

Wennerberg et al. [17] presented in their study that Carisolv gel did not have any influence on surface topography of sound dentin, or if there were any changes, they were insignificant which is in accordance with our study. However, in their study it was not clearly explained if dentin surface was covered with smear layer or not. Other studies confirmed selective action of Carisolv on soft carious dentin without any effect on sound dentin and enamel [6, 18]. Nevertheless, surface topography of sound dentin after caries removal showed no smear layer, as opposed to dentin surface that had smear layer after caries elimination using burs.

It has been shown that deeper layers of carious lesion are insignificantly demineralized and contain intact organic matrix with unchanged collagen meshwork and potential for remineralization [19, 20, 21]. Also, after dentin conditioning with 35% phosphorous acid, its structure becomes poorer in minerals, richer in proteins with exposed collagen fibers, and has increased permeability [22]. Our results showed no changes in topography of demineralized dentin after 20-minute exposure to Carisolv gel. Hannig et al. [23] also reported no notable ultra structural changes in already demineralized dentin structure after application of Carisolv gel. There is an assumption that Carisolv gel most likely protects intact collagen matrix of dentin that may be remineralized.

However, it should be emphasized that SEM can analyze only surface topography of dentin. Jepsen et al. [24] performed biochemical analyses of residual dentin after chemo-mechanical caries elimination and found that major part of collagen (organic dental structure) was denatured with Carisolv. Tonami et al. [25] also described selective action of Carisolv on carious dentin only with collagen fibers denatured or fully dissolved but without the damage of sound dentin.

The cost of material and the length of procedure are considered to be main disadvantages of the chemo-mechanical method. On the other hand, chemo-mechanical method avoids usage of local anesthetics, which makes it suitable in pediatric dentistry and for patients with anxiety and medical handicap [12, 26].

CONCLUSION

There was no visible change of sound and demineralized dentin prior to and after the application of Carisolv gel whereas carious dentin was selectively dissolved and separated from sound dentin. The evaluation of dentin ultrastructure after complete chemical and mechanical caries removal may be the purpose of some future research.

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Received: 29/12/2014 • Accepted: 05/02/2015

Uticaj gela *Carisolv* na zdrav, demineralizovan i karijesno izmenjen dentin – studija *in vitro*

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KRATAK SADRŽAJ

Uvod Bol i nelagodnost tokom stomatoloških intervencija prouzrokovani svrdlima visokoturažnog kolenjaka najčešći su razlozi za izbegavanje posete stomatologu. Poslednjih godina pojavile su se brojne mogućnosti koje bi izostavile svrdlo kao način uklanjanja karijesa, a jedan od njih je hemijsko-mehanička metoda uklanjanja karijesa. Cilj ovog istraživanja bio je da se analiziraju mikromorfološke promene nastale nakon primene gela *Carisolv* na zdrav, demineralizovan i karijesno izmenjen dentin pomoću skening-elektronske mikroskopije (SEM).

Materijal i metode rada U istraživanju je korišćeno 30 zuba ekstrahovanih iz ortodontskih razloga (20 intaktnih i 10 s karijesnim lezijama). Korišćen je sistem Carisolv™ (*MediTeam*, Švedska), materijal za hemijsko-mehaničko uklanjanje karijesa koji je sastoji od gela Carisolv i specifičnih nikl-titanijumskih ručnih instrumenata. U našoj studiji korišćen je samo gel Carisolv, koji je aplikovan na zdrav, demineralizovan i karijesno izmenjen dentin tokom 20 minuta. Skening-elektronski mikroskop je primenjen za analizu površine dentina.

Rezultati SEM analiza je pokazala da ovaj gel utiče samo na karijesni dentin, dok na zdrav dentin nije imao nikakvog uticaja bez obzira na stanje demineralizacije.

Zaključak Carisolv nije pokazao nikakve promene na zdravom i demineralizovanom dentinu, ali je uticao na selektivno rastvaranje i uklanjanje dentina izmenjenog karijesom.

Ključne reči: gel Carisolv; zdrav dentin; karijesno promenjeni dentin; demineralizacija; SEM

UVOD

U skladu s razvojem savremenih tehnologija, tako su i preparacija kaviteta i materijali za restauraciju zuba doživeli znatna unapređenja i poboljšanja. Ipak, najpopularnija tehnika uklanjanja karijesno promenjenog dentina je pomoću visokoturažnog kolenjaka i svrdla. Ova metoda je invazivna za zubna tkiva i može izazvati različite negativne efekte. Upotreba visokoturažnog kolenjaka i svrdala dovodi do oštećenja gleđnih prizmi, i to ne samo na rubovima kaviteta, već i u dubljim delovima gleđi. Ove pukotine predstavljaju mesta za ulaz bakterija koje mogu izazvati karijes [1]. Takođe, svrdla ne deluju selektivno samo na karijesno izmenjen dentin, već uklanjaju i deo zdravog zubnog tkiva. To dovodi do slabljenja zuba i moguće frakture krunice zuba [2].

Imajući u vidu sve loše posledice koje mogu nastati nakon upotrebe visokoturažnog kolenjaka i svrdla, potrebno je pronaći tehniku za eliminaciju karijesa koja bi bila minimalno invazivna [3-8]. Svetska zdravstvena organizacija je već dala predlog atraumatskog restorativnog lečenja [9]. Poslednjih godina su razvijane tehnike i metode eliminacije karijesa koje uključuju minimalnu nelagodnost i bol, bolju efikasnost i prihvatljivije su za pacijenta.

Sistem *Carisolv* je hemijsko-mehanička metoda uklanjanja karijesa i opisana je u literaturi kao alternativa svrdlima. Ova metoda je ugodna i manje bolna nego klasična metoda eliminacije karijesa [10]. Takođe se pokazala kao vrlo selektivna samo za karijesni dentin; ipak, njen efekat na zdrav i demineralizovan dentin još nije proučen.

Cilj ovog istraživanja bio je da se pomoću skening-elektronske mikroskopije (SEM) analiziraju mikromorfološke promene zdravog, demineralizovanog i dentina izmenjenog karijesom nakon upotrebe gela Carisolv.

MATERIJAL I METODE RADA

Sistem Carisolv™ (*MediTeam*, Švedska), koji čine gel Carisolv i posebni nikl-titanijumski ručni instrumenti, korišćen je za hemijsko-mehaničko uklanjanje karijesa. Gel Carisolv sadrži dve komponente. Jedna se sastoji od tri amino-kiseline (leucin, lizin i glutaminska kiselina), NaCl, NaOH, eritrozin i destilovane vode, dok drugu čini 0,5% NaOCl. Kada se pomešaju ove komponente, dobije se izotonični gel sa baznom pH vrednošću (pH=11). Delovanje aktivne supstance je ograničeno na 20–30 minuta.

Ručni instrumenti su dizajnirani tako da imaju radni deo na obe strane, a veličina radnog dela je od 0,3 do 2 mm. Oni su napravljeni od nikl-titanijuma i mogu se naći u različitim veličinama. Instrumenti se koriste za unošenje gela u karijesnu leziju i uklanjanje mekanog i karijesnog dentina. Radni deo instrumenta ima 2–8 oštrica koje omogućuju delovanje u nekoliko pravaca. Oštra ivica instrumenta uklanja karijes pod uglom od 90 stepeni i pruža dobru dubinsku kontrolu.

Za SEM analizu je korišćeno 20 intaktnih i 10 zuba koji su imali karijesne lezije, ali bez invazije pulpe, koji su bili izvađeni iz ortodontskih ili protetičkih razloga. Nakon ekstrakcije zubi su očišćeni i svrstani u tri grupe. Svi su zubi najpre postavljeni u uređaj za isparavanje. Gleđ zuba je lomljenjem odvojena od ostalog zubnog tkiva. Gel Carisolv je potom postavljen na dentinsku površinu 10 zdravih zuba. Dentin drugih 10 zuba je najpre demineralizovan fosfornom kiselinom (25–30 sekundi), a potom je aplikovan gel Carisolv. Na 10 karijesnih zuba gel je postavljen direktno u karijesne lezije. Svi uzorci su bili izloženi gelu Carisolv 20 minuta bez bilo kakvog mehaničkog efekta. Nakon 20 minuta zubi su pripremljeni za SEM analizu. Uzorci su postavljeni na postolje, držani u isparivaču i obloženi zlatom za skening-elektronski mikroskop (JEOL JSM-5300, Naučno-istraživački centar, Stomatološki fakultet u Nišu, Srbija).

REZULTATI

Tabela 1 prikazuje raspored uzoraka po grupama (zdrav, karijesni i demineralizovani dentin). SEM analizom nisu uočene značajne razlike u strukturi dentina između zuba koji nisu bili tretirani i zuba gde je gel Carisolv bio primenjen na zdrav dentin (Slike 1a i 1b).

U drugoj grupi, u kojoj su zubi bili tretirani fosfornom kiselinom, uočena je demineralizacija površine dentina sa čvrstim slojem kompaktno postavljenih kolagenih vlakana debljine 3–5 μm (Slika 2a). Nakon dvadesetominutnog izlaganja demineralizovane površine dentina gelu Carisolv, nisu uočene mikromorfološke promene, niti promene u čvrstoći ovoga sloja (Slika 2b).

U trećoj grupi zuba, nakon primene gela Carisolv na karijesni dentin, SEM analiza je pokazala da je gel doveo do odvajanja karijesno izmenjenog dentina od zdravog dentina, što je omogućilo njegovo lakše uklanjanje (Slika 3a). Osim promena na karijesnom dentinu, drugih promena (na zdravom dentinu) nije bilo (Slika 3b).

DISKUSIJA

Razvoj sistema za hemijsko-mehaničko uklanjanje karijesa najverovatnije potiče od želje pacijenata za bezbolnim uklanjanjem karijesa, bez korišćenja svrdla i neugodnog osećaja [11]. To su možda najčešći razlozi za izbegavanje poseta stomatologu i redovne preglede.

Sistem Carisolv, koji se sastoji od gela Carisolv i ručnih instrumenata s nikl-titanijumskim radnim delom, uveden je na tržište 1998. godine. Gel deluje pomoću NaOCl i tri prirodne amino-kiseline [7]. Bezbolno uklanjanje karijesno promenjenog tkiva bez prethodne upotrebe lokalne anestezije velika je prednost hemijsko-mehaničke metode. Selektivno delovanje gela na karijesno izmenjeni dentin, usaglašenost temperature i izostanak pritiska i vibracija koji postoje tokom uklanjanja karijesa pomoću svrdala velika je prednost ovog sistema. Primena izotoničnog gela Carisolv na dentin ne dovodi do temperaturne i osmotske razlike u pulpi zuba zato što je gel iste temperature kao i telesna temperatura [5]. Oštri rubovi ručnih instrumenata specifičnih za uklanjanje karijesnog dentina samo dodatno doprinose bezbolnom lečenju [12]. Ipak, ova metoda i nije baš najpogodnija za uklanjanje početnih karijesnih lezija [13]. S druge strane, zbog selektivnog delovanja samo na karijesni dentin, ona smanjuje rizik od jatrogenog otvaranja pulpe i potiskivanja karijesno promenjenog dentina u pulpu. Damaške (*Dammaschke*) i saradnici [14, 15] su pokazali da gel ne prodire kroz zdrav dentin, pa ne postoji opasnost od oštećenja pulpe. Ipak, važno je dati pravilnu indikaciju za upotrebu gela Carisolv.

Glavni cilj ovog istraživanja bio je da se utvrdi da li gel Carisolv deluje selektivno samo na meki i karijesno promenjeni dentin bez oštećenja okolnog dentina. SEM je metoda koja se često koristi za proučavanje morfologije dentina [4]. Takođe je moguće dobiti i pseudotrodimenzionalnu sliku za analizu posmatrane površine [16]. Ova studija je analizirala efekat gela Carisolv na oštećenu površinu dentina zato što su promene manje vidljive ako je površina prekrivena razmaznim slojem. Nakon dvadesetominutnog izlaganja zdravog dentina gelu Carisolv nije uočena značajna promena površine. Vreme ekspozicije je bilo ograničeno na 20 minuta prema preporuci proizvođača.

Venerberg (*Wennerberg*) i saradnici [17] su u svojoj studiji pokazali da gel Carisolv nije imao nikakav uticaj na površinsku topografiju zdravog dentina ili su promene bile minimalne, što je u skladu s našim rezultatima. Međutim, oni ne objašnjavaju da li je površina dentina bila pokrivena razmaznim slojem ili ne. Druge studije su potvrdile selektivno delovanje gela Carisolv samo na mekom karijesnom dentinu, bez uticaja na zdrav dentin i gleđ [6, 18]. Takođe, nakon uklanjanja karijesa ovim gelom ne ostaje razmazni sloj na površini, što nije slučaj nakon eliminacije karijesa pomoću svrdala.

Analizom karijesne lezije pokazano je da su dublji slojevi dentina neznatno demineralizovani i sadrže intaktnu organsku matricu nepromenjenih kolagenskih vlakana s potencijalom za remineralizaciju [19, 20, 21]. Takođe, nakon kondicioniranja dentina fosfornom kiselinom u koncentraciji od 35%, njegova struktura postaje siromašnija mineralima a bogatija proteinima s izloženim kolagenskim vlaknima i povećanom propustljivošću [22]. Ovi rezultati su pokazali da ne postoje nikakve promene u topografiji demineralizovanog dentina nakon dvadesetominutnog izlaganja gelu Carisolv. Hanig (*Hannig*) i saradnici [23] su takođe zapazili da nema značajnih ultrastrukturnih promena kod demineralizovanog dentina nakon primene gela Carisolv. Pretpostavlja se da ovaj gel verovatno štiti netaknutu kolagensku matricu dentina koji može biti remineralizovan.

Međutim, treba naglasiti da SEM može samo analizirati topografiju površine dentina. Jepsen (*Jepsen*) i saradnici su izvršili biohemijsku analizu preostalog dentina nakon hemijsko-mehaničkog uklanjanja karijesa i utvrdili da je najveći deo kolagena bio denaturisan reakcijom gela Carisolv [24]. Tonami (*Tonami*) i saradnici [25] su takođe opisali selektivno delovanje ovoga gela na karijesni dentin, gde su kolagena vlakna bila denaturisana ili potpuno rastvorena, ali bez oštećenja zdravog dentina.

Troškovi materijala i dužina postupka su glavni nedostaci ove hemijsko-mehaničke metode. S druge strane, ne postoji potreba za lokalnim anestetikom, što ga čini prikladnim u dečjoj stomatologiji i kod bolesnika koji imaju strah od stomatološke intervencije, odnosno kod onih s medicinskim hendikepom [12, 26].

ZAKLJUČAK

Nije bilo vidljivih promena na zdravom i demineralizovanom dentinu pre i nakon primene gela Carisolv, dok je karijesni dentin selektivno rastvoren i uklonjen. Analiza ultrastrukture dentina nakon potpune hemijske i mehaničke eliminacije karijesa može biti cilj budućih istraživanja.