Marginal Microleakage of Newly Synthesized Nanostructured Biomaterials Based on Active Calcium Silicate Systems and Hydroxyapatite

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SUMMARY
Introduction Calcium silicate cements can be successfully used for the treatment of root perforations due to their exceptional biological and sealing properties. The aim of this study was to test, using dye penetration method, marginal microleakage of newly synthesized nanostructured biomaterials based on calcium silicate system and hydroxyapatite after their application in interradicular perforation of extracted teeth.

Material and Methods The study included 34 extracted human molars. Newly synthesized nanostructured materials: one based on calcium silicate system (CS), and the other one based on hydroxyapatite and active calcium silicate system (HA-CS) were tested. Mineral trioxide aggregate (MTA; Angelus, Londrina, Brazil) was used as control. Marginal microleakage was evaluated using dye penetration test 6 months after the application of materials in experimentally prepared inter-radicular perforations in extracted human molars. Dye penetration was analyzed using light microscope at 30X magnification, a method of quantifying visual information in computer software for image processing (Adobe Photoshop CS5 Extended, version 12.0 x 32). The values were expressed in millimeters, and the results statistically analyzed using one-way ANOVA and Tukey post-hoc test (α=0.05).

Results The shortest dye penetration was measured for calcium silicate system (0.44 mm), while slightly higher values were found for MTA (0.54 mm). Dye penetration for hydroxyapatite and active calcium silicate system (2.00 mm) was longer than for the other two materials (p<0.05).

Conclusion The lowest marginal microleakage was observed in CS and was comparable to MTA. Microleakage in HA-CS was significantly higher than CS and MTA.

Keywords: marginal microleakage; calcium silicate cements; hydroxyapatite

INTRODUCTION
Marginal seal or adequate marginal adaptation of material along the cavity walls should be able to prevent leakage of tissue fluid and consequently bacterial microleakage. Therefore, it is considered as significant factor for long-term success of endodontic treatment [1].

Calcium silicate cements were introduced in endodontic practice in the mid 90-ies of the last century in order to be used as materials for closing root perforations and root apical closure after apical surgery. Studies have demonstrated superior marginal seal achieved by calcium silicate cements compared to amalgam and cements based on zinc-oxide-eugenol (ZOE; Super EBA and IRM cement), which have been commonly used in these indications [1]. It has also been demonstrated that calcium silicate cements achieve good marginal seal in cavities contaminated with blood [1, 2], they are biocompatible [1], bioactive [3] and induce regeneration of hard dental [4, 5] and periodontal tissue [6]. Consequently, their indications have expanded significantly [7].

The main issue for clinical use of calcium silicate cements is their long setting time of several hours increases the risk of material dissolution and leaching from the site of application. Moisture is needed for hydration and setting of these materials, which is why additional external moisture is needed during initial bonding. That inevitably delays completion of endodontic treatment [9]. In previous years, numerous studies have been conducted in order to overcome this problem [10-13].

Modern research has focused on synthesis and evaluation of new nanostructured biomaterials in similar endodontic indications [14]. Nanomaterials are characterized among others, by pronounced activity of particles and consequently, faster setting in comparison with conventional microstructural materials [15]. The Institute of Nuclear Sciences in Vinča, according to the recipe of Jokanović et al. [16], synthesized two new biomaterials: one based on calcium silicate system (CS) and the other one based on hydroxyapatite and active calcium silicate system (HA-CS). These materials showed initial setting of 10 minutes (CS) and 15 minutes (HA-CS), respectively. Previous studies have shown that these materials have less genotoxic and cytotoxic effects, but higher osteogenic potential than commercial calcium silicate cements [17, 18].

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The aim of this study was to test, using dye penetration method, marginal microleakage of newly synthesized nanostructured biomaterials based on calcium silicate system and hydroxyapatite (HA-CS) after their application in interradicular perforation of extracted teeth.

MATERIAL AND METHODS

Three materials in total were tested in this study: new material based on calcium silicate system (CS), new material based on mixtures of hydroxyapatite and calcium silicate system (HA-CS) and control material MTA (MTA Angelus®, Londrina, Brazil). Experiments were performed on 34 human, extracted, maxillary and mandibular molars with fully developed and non-fused roots. Access cavities were prepared with high speed handpiece with a bur size #4 inter-radicular perforations were prepared. The size of perforations was the same as diameter of round bur while the depth depended on the thickness of the pulp chamber. After washing perforations with distilled water and air-drying, the teeth were randomly assigned into three experimental groups according to the materials used: CS, HA-CS and MTA (n=10). Positive controls were teeth with perforations that were not sealed and negative controls were two teeth without perforations.

Prior to the application of the test material, the teeth were placed in a sponge soaked with artificial tissue fluid and placed in the prepared cavities using condensers. On the top of the material a moist cotton pellet was placed and the teeth were incubated at 37°C for 24 h. After complete setting of materials access cavities were permanently closed using composite. Teeth were incubated in sponges at 37°C for the next 6 months. After 6 months marginal microleakage was evaluated using passive dye penetration method. The teeth were coated with two coats of varnish, except in the area of the material in the furcation (about 1 mm around material) and placed in 50% solution of silver nitrate (AgNO₃) for 2 h. After quick rinsing teeth were immersed in photo developer for 6 h. Teeth were cut longitudinally in the region of perforation using diamond disc of 0.7 mm thickness and linear speed saw with water cooling (Isomet testers 4000, Buehler, Lake Bluff, IL, USA). Dye penetration between the material and cavity walls was analyzed using light microscopy and photographed at 30X magnification. The depth of dye penetration was measured using the method of quantifying of visual information in computer software for image processing (Adobe Photoshop CS5 Extended, version 12.0 x 32). The obtained values were expressed in millimeters. The results were statistically analyzed using one-way ANOVA with Tukey post-hoc test. The level of significance was set at α=0.05

RESULTS

The shortest dye penetration was found for CS (0.44±0.54 mm) (Figure 1). Similar values were measured for MTA (0.54±0.76 mm) (Figure 2). The longest dye penetration was recorded for HA-CS (2.00±0.70 mm) (Graph 1, Figure 3). There was no statistically significant difference in marginal dye penetration between CS and MTA. HA-CS showed dye penetration that was significantly longer than for CS and MTA (p<0.05).

In addition to color penetration between restorations and dentin, staining of tested materials was also noticed (Figures 4, 5 and 6).

DISCUSSION

Dye or bacteria penetration or fluid filtration methods are most commonly used for the evaluation of marginal microleakage of different materials in vitro [19–22]. In the current study dye penetration method was used due to simplicity as described in numerous studies [19, 20, 23, 24, 25]. One of the main objections to dye penetration method is related to the size of dye molecules that are commonly used which are actually smaller than the size of bacteria [19]. Therefore, for materials that show penetration of small dye molecules, it can be expected that leakage of larger molecules (bacteria and their products) will be absent [1].

When choosing dye for the assessment of marginal leakage chemical nature of the tested material should be taken into account. Wu et al. [26] found that methylene blue is unstable in the presence of alkaline substances resulting in its decolorization. As calcium hydroxide is major ingredient of MTA, the use of methylene blue for
Figure 1. Inter-radicular perforation filled with calcium silicate material. There is no dye marginal leakage (×30).

Slika 1. Interradiklna perforacija ispunjena kalcijumsilikatnim materijalom. Ne uočava se marginalni prodor boje (×30).

Figure 2. Inter-radicular perforation filled with MTA. There is no dye marginal leakage (×30).

Slika 2. Interradiklna perforacija ispunjena materijalom MTA. Ne uočava se marginalni prodor boje (×30).

Figure 3. Inter-radicular perforation filled with hydroxyapatite-calcium silicate material. There is dye marginal leakage and absorption with material color change (×30).

Slika 3. Interradiklna perforacija ispunjena materijalom hidroksijapatitom i kalcijumsilikatnim cementom. Uočavaju se prodor i apsorpcija boje sa delimičnim prebojavanjem materijala (×30).

Figure 4. Inter-radicular perforation filled with calcium silicate material. There is dye marginal leakage and absorption with material color change (×30).

Slika 4. Interradiklna perforacija ispunjena kalcijumsilikatnim materijalom. Uočava se prodor i apsorpcija boje sa delimičnim prebojavanjem materijala (×30).

Figure 5. Inter-radicular perforation filled with hydroxyapatite-calcium silicate material. There is dye absorption with most of material having changed color (×30).

Slika 5. Interradiklna perforacija ispunjena materijalom hidroksijapatitom i kalcijumsilikatnim cementom. Uočava se apsorpcija boje sa prebojavanjem većeg dela materijala (×30).

Figure 6. Inter-radicular perforation filled with MTA. There is dye absorption and color change throughout the whole thickness of material (×30).

Slika 6. Interradiklna perforacija ispunjena materijalom MTA. Uočavaju se prodor i apsorpcija boje celom debljinom materijala (×30).
the evaluation of MTA and other materials of similar composition would cause its discoloration and eventually unreliable results. A solution of silver nitrate was chosen as dye due to its stability in the presence of the high pH and high molecular weight of silver particles [25].

Marginal microleakage was assessed after the application of the test material in experimentally prepared, inter-radicular perforations of extracted teeth. During the experimental period the teeth were stored in an incubator, with a sponge soaked in artificial tissue fluid to simulate clinical conditions [21]. In the current study, the deepest dye penetration was measured after the teeth were longitudinally cut. Therefore, it was not possible to determine precisely in which part of the cavity dye penetrated the most. Dye penetration method done in vitro cannot be directly related to the complex in vivo microleakage but it can be possible indicator of microleakage in clinical conditions [20].

The lowest dye penetration in the current study was observed for CS material. The values of dye penetration for MTA were slightly higher but with no statistically significant difference compared to CS. Given that CS and MTA have similar composition and physical properties are likely similar. Good marginal seal of calcium silicate cements is associated with mild expansion of cement after setting [27, 28].

However, different microleakage evaluating methods have shown that MTA and similar calcium silicate cements do not have the ability of absolute hermetic sealing [21, 22, 25, 30], which is consistent with the results of the current study. In the group of samples with CS, the measured values of dye penetration were lower compared to the MTA samples, however, larger number of samples in the CS group showed microleakage than in the MTA group. The measured values of dye penetration in the MTA group were very different, and the results are consistent with the findings of De Deus et al. [30]. They evaluated the marginal microleakage of different calcium silicate cements (PC, MTA Angelus and MTA Bio) and reported very variable results within groups. They also detected microleakage in all samples but with no statistically significant difference between different calcium silicate cements [30]. In contrast, Hashem et al. [19] observed differences in the permeability of various commercial calcium silicates, considering that different chemical composition and different setting time may play a role in the material attachment to the cavity walls. Also, studies that evaluated permeability of calcium silicate cements using bacteria penetration method have reported different results. Montelano et al. [2] recorded the most pronounced microleakage in the first days of application of materials, while Parirokh et al. [21] detected significantly delayed bacterial penetration (between 39 and 73 days of experiment) highlighting the positive impact of phosphate solution on the adhesion of material.

Literature reviews describe inconsistency in results for the permeability of calcium silicate cements using different testing methods. Many factors influence microleakage of MTA: dye, pH of dye, storage conditions of samples before experiment, and setting time of material prior to immersion of samples in dye solution [1]. Also, different results may be due to the difference in diameter and depth of perforations, number of samples, or duration of observational period [30].

For adequate understanding of results obtained in the current study, it is important to explain the way of dye penetration. Samples that showed dye penetration did not confirm strictly marginal microleakage. In fact, besides the dye detected at the junction of materials and dentin, staining of materials was noted likely due to dye absorption (Figures 4, 5 and 6). Similar results were reported by Tobón-Arroyave et al. [20]. They found that dye penetration in MTA was different in nature than in conventional cements (IRM and Super EBA). When IRM and Super EBA cements were used as apical plug, dye penetration was noted as a circle around the material, between material and dentin. In contrast, in all MTA samples color change was noted in the entire thickness of the material [20]. The reason for these results could be found in MTA structure made of numerous pores and capillaries that could cause more pronounced permeability. It is important however to note that MTA samples in that study had setting time of just 30 minutes before their immersion in dye, therefore results could reflect incomplete setting of material. In the current study, samples were allowed 24 hours setting time before incubation and then after incubation in humid environment for 6 months. This protocol could allow more complete setting and consequently lower dye penetration and absorption.

HA-CS showed the deepest dye penetration and in the majority of samples material disintegration was observed. Since hydroxyapatite is well known for its porous structure and inadequate mechanical properties, the results were somewhat expected. During synthesis HA-CS, calcium silicate was added to hydroxyapatite in order to improve its mechanical properties. Based on the current results it can be noted that additional quantity of calcium silicate (HA:CS=2:1) under the experimental conditions of this study, did not prevent dissolution and leaching of material from the site of application. Literature findings also suggest that calcium phosphate cements do not provide adequate sealing due to their porosity and solubility and in that sense they are inferior to calcium silicate cements [24, 25].

It is worth noting that in some MTA and CS samples surface disintegration or dissolution of the outer layer material was noted but still less than in HA-CS samples. In contact with water or phosphate solution, cement releases calcium ions with partial decalcification of calcium silicate hydrate, whereas the expansion of cement after contact with liquid may result in microfractures. All these may constitute an obstacle to long-term stability of the material [13]. However, when interpreting results of an in vitro study, it must be noted that tissue response was absent. Namely, one of the characteristics of the tested bioactive materials in this study is formation of hydroxyapatite on their surface in contact with tissue fluids [3, 29], as well as the formation of calcified tissue via calcium silicate or calcium phosphate cements. Therefore, one might expect that in clinical setting material disintegration could be
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CONCLUSION

The lowest marginal microleakage or the best marginal seal was observed in CS and MTA. Microleakage of HA-CS was significantly higher compared to the other two tested materials.

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Ispitivanje marginalne mikropropustljivosti novosintetisanih nanostrukturalnih biomaterijala na bazi aktivnih kalcijumsilikatnih sistema i hidroksiapatita

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KRATAK SADRŽAJ

Uvod Primena cemenata od kalcijumsilikata u lečenju perforacija korena zuba asziva se na njihovim izuzetnim biološkim svojstvima i odgovarajućem rubnom zaptivanju. Cilj ovog rada bio je da se testom probara boje ispituje marginalna mikropropustljivost novosintetisanih nanostrukturalnih biomaterijala na bazi kalcijumsilikatnih sistema i hidroksiapatita nakon primene materijala u interradiksnom perforaciji ekstrahovanih zuba.

Materijal i metode rada Ispitivanje je realizovano na 34 ekstrahovana humana molara. Testirani su novosintetisani nanostrukturalni materijal: materijal na bazi aktivnih kalcijumsilikatnih sistema (CS) i materijal na bazi hidroksiapatita i aktivnih kalcijumsilikatnih sistema (HA-CS). Kao kontrolni materijal korišćen je komercijalni kalcijumsilikatni cement (MTA Angelus, Londrina, Brazil). Materalna propustljivost je ispitavana testom probara boje šest meseci nakon primene materijala u eksperimentalno preprisane interradiksnhe perforacije na ekstrahovanim humanim molarima. Probara boje je analiziran svetlosnim mikroskopom na uveličanju od 30 puta metodom kvantifikovanja vizuelnih informacija kompjuterskim programom za obradu slike (Adobe Photoshop CS5 Extended, verzija 12.0x32). Dobijene vrednosti su izražene u milimetrima, a dobijeni rezultati statistički su obrađeni primenom jednofaktorskog testa ANOVA sa Tukeyim (Tukey) post-hoc testom (α=0,05).

Rezultati Najmanji probara boje izmereni je kod materijala CS (0,44 mm), dok su nešto veće vrednosti zabeležene kod MTA (0,54 mm). Probara boje kod materijala HA-CS (2,00 mm) bio je značajno veći u odnosu na materijale CS i MTA (p<0,005).

Zaključak Najmanja marginalna mikropropustljivost zabeležena je kod materijala CS i bila je komparabilna sa MTA. Mikropropustljivost materijala HA-CS bila je značajno veća u odnosu na materijal CS i kontrolni materijal MTA.

Ključne reči: marginalna mikropropustljivost; kalcijumsilikatni cement; hidroksiapatit

UVOD

Kvalitetno rubno zaptivanje, odnosno odgovarajuća marginalna adaptacija materijala uz zidove kaviteta, treba da onemogući protok tkivnih tečnosti pod poslagom i posle bakterijskog mikrocurenja, zbog čega se smatra značajnim faktorom za dugoročan uspeh endodontskog lečenja [1].

Cementi od kalcijumsilikata uvedeni su u endodontski praksu sredinom devedesetih godina prošlog veka, najpre kao materijali namenjeni lečenju korenih perforacija i zatvarajući vrha korena zuba nakon apskne hirurgije. Istraživanja su ukazala na znatno bolje rubno zaptivanje kalcijumsilikata u odnosu na amalgamu i komercijalne cemente na bazi ZOE (Super EBA i IRM cement), koji su obično korišćeni u navedenim indikacijama [1]. Kasnije je utvrđeno da kalcijumsilikatni cementi ostvaruju kvalitetno rubno zaptivanje i u kvitetima koji su kontaminirani krvlju [1, 2], da su biokompatibilni [1], bioaktivni [3] i da indukuju regeneraciju čvrstih zubnih tkiva [4, 5] i periodontijuma [6], zbog čega su njihove indikacije danas značajno proširene [7].

Glavni problem koji otežava kliničku primenu kalcijumsilikata je dugo vreme vezivanja [8]. Inicijalno vezivanje od nekoliko sati nosi rizik od rastvaranja i ispiranja materijala s mesta aplikacije. Za hidrataciju i ocvršćivanje materijala neophodna je vlaga, pa se preporučuje da se nakon primene (u periodu inicijalnog vezivanja) materijalu obezbedi eksterna vlaga, što neminovno odlaze završetak endodontskog lečenja [9]. Ranijih godina uđena su brojna istraživanja radi prevazilaženja ovog problema [10-13].

Treminje istraživanja je fokusirana na sintezu i ispitivanja moguće primene nanostrukturalnih biomaterijala u sličnim endodontskim indikacijama [14]. Nanomaterijale, između ostalog, odlučuje izraženija aktivnost čestica, te posleđico brže vezivanje u odnosu na konvencionalne mikrostrukturne materijale [15]. U Institutu za nuklearne nauke u Vinči, prema recepturi Jokanovića i saradnika [16], sintetisasu su dva nova biomaterijala: materijal na bazi aktivnih kalcijumsilikatnih sistema (CS) i materijal na bazi hidroksiapatita i aktivnih kalcijumsilikatnih sistema (HA-CS). Nanotechnologijom dobijeni su materijali s inicijalnim vezivanjem od 10 minuta (CS), odnosno 15 minuta (HA-CS). Dosadašnja istraživanja su pokazala da dobijeni materijali imaju slabija genotoksicksa i citotoksicksa dejstva, kao i veći osteogeni potencijal u odnosu na komercijalne kalcijumsilikatne cemente [17, 18].

Cilj ovog istraživanja bio je da se testom probara boje ispituje marginalna mikropropustljivost materijala CS i HA-CS nakon primene materijala u interradiksnim perforacijama ekstrahovanih humanih zuba.

MATERIJAL I METODE RADA

U ovom istraživanju testirani su materijal na bazi kalcijumsilikatnih sistema (CS), materijal na bazi mešavine hidroksiapatita i kalcijumsilikatnih sistema (HA-CS) i kontrolni materijal MTA (MTA Angelus, Londrina, Brazil). Istraživanje je urađeno na 34 humana ekstrahovana maksilarne i mandibularne molare s potpuno razvijenim i nefuzionsanim korenovima. Pristupni kaviteti su preparisani visokotaktnošnom bušilicom, nakon čega je ručnim K-turpijama (K-files, VDW GmbH, Nemačka) utvrđena radna dužina kanala korena zuba, na 1 mm kraće od anatomskog foramen. Kanali korena su preparisani tehnikom
crown-down, serijom mašinskih endodontskih instrumenta Bio Race (FKG Dentaire, Swiss Dental Products, Švajcarska). Tokom instrumentacije kanali su ispirani sa NaOCl u koncentraciji od 0,5%. Za finalno ispiranje je korišćeno 5 ml desetoprocentne limunske kiseline (tokom jednog minuta) i 5 ml 0,5% NaOCl. Nakon sušenja kanali su opturisani monokonom tehnikom uz siler Acrosal (Septodont, Francuska).

Po vezivanju silera, u centru poda pulpne komore, kolenjakom i okruglim borenem veličine #4, prepiran su interadikanske perforacije tako da je širina perforacija odgovarala promjeru borenog stava, dok je dubina bareda iznijela od debljine podne pulpne komore. Posle ispiranja prepiranih katetiva destilovanim vodom i sušenja, zubi su metodom slučajnog izbora podeljeni u tri eksperimentalne grupe, a perforacije zatvorene test materijala CS, HA-CS i MTA (n=10). Pozitivnu kontrolnu grupu činila su dva zuba kod kojih perforacije nisu zatvorene materijalima, a negativnu dva zuba bez perforacija.

Pre primene ispitanih materijala zubi su do nivoa gledno-cementne granice postavljeni u sunder natopljeni veštačkom tkinom tečnošću – Henkovic uravnoteženim slanim rastvorom (engl. Hank's balanced salt solution – HBSS). Materijali su zamešani sa destilovanim vodom u odnosu 3:1 i u katetive kondenzovani nabijajući. Preko materijala je postavljena vlažna vatica, a zatim su zubi inkubirani na 37°C tokom 24 časa. Po vezivanju materijala pristupni katetiti su konačno zatvoreni kompozitom, a zubi u natopljenom sunderu inkubirani su na 37°C narednih šest meseci.

Posle šest meseci marginalna mikropropustljivost je ispitivana metodom pasivnog prorađa boje. Zubi su premazani sa dva sloja laka, osim u predelu materijala u furkaciji (1 mm oko materijala) i potopljeni u pedesetoprocentni rastvor srebra-nitra (AgNO₃) tokom dva časa. Nakon kratkog ispiranja zubi su ostavljani u fotografskom razvoju narednih šest sati. Zatim su longitudinalno sečeni u predelu interadiksnih perforacija dijamantskim diskom debljine 0,7 mm, linearnom preciznom testerom sa vodenim hladnjem (Isomet tester 4000, Buehler, Lake Bluff, IL, SAD). Prodor boje između materijala i zidova katetiva je analiziran svetlosnim mikroskopom i fotografisan pri uveličanju od 30 puta. Dubina prodora boje je merena metodom kvantifikovanja vizuelnih informacija kompjuterskim programom za obradu slike (Adobe Photoshop CS5 Extended, verzija 12,0x32). Dobijene vrednosti su izražene u milimetrima, a rezultati statistički obradjeni primenom jednofaktorskog testa ANOVA sa Tukjevim (Tukey) post-hoc testom. Nivo značajnosti utvrđen je na α=0,05.

REZULTATI

Najmanji prodor boje izmeren je kod materijala CS (0,44±0,54 mm) (Slika 1). Slične vrednosti izmerene su za materijal MTA (0,54±0,76 mm) (Slika 2). Najveći prodor boje zabeležen je kod materijala HA-CS (2,00±0,70 mm) (Grafikon 1, Slika 3). Nisu uočene statistički značajne razlike u marginalnom prodoru boje između materijala CS i MTA. Kod materijala HA-CS izmerene vrednosti prodora boje bile su statistički značajno veće u odnosu na prodor boje kod materijala CS i MTA (p<0,05).

Osim prodora boje, na spinu materijala i zubnih struktura uočeni je prebojavanje testiranog materijala primjenjenog u veštacki formirane interadiksne perforacije (Slike 4, 5 i 6).

DISKUSIJA

Za procenu marginalne mikropropustljivosti materijala u studijama in vitro obično se koriste metode prodora boje [19, 20], prodora bakterija [21] i filtracije tečnosti [22]. U ovom istraživanju za procenu mikropropustljivosti je primenjena metoda prodora boje, koja je zbog svoje jednostavnosti korišćena u velikom broju istraživanja [19, 20, 23, 24, 25]. Glavni nedostatak ove metode je, međutim, u tome što je veličina molekula boje koje se obično koriste manja od veličine bakterija [19]. Kod materijala kod kojih se beleži prodor malih molekula boje može se očekivati izostanak mikrocurenja većih molekula, poput bakterija i njihovih proizvoda [1].

Prilikom odabira boje za procenu marginalne propustljivosti uzeta je u obzir hemijska priroda materijala koji se ispituju. Naime, Vl (Vl) i saradnici [26] su ustanovili da je metilen plavo nestabilno u prisustvu alkaličkih supstanaca, što dovodi do njegovog obezbojanja. Kako je kalcijum-hidroksid glavna hemijska smesa koja se oslobađa iz MTA, upotreba metilen plavog prilikom ispitivanja MTA i drugih materijala sličnog hemijskog sastava bi, usled diskoloracije boje, mogla dovesti do nepouzdanih rezultata. Rastvor srebro-nitra je stabilan u prisustvu materijala visoke vrednosti pH, a čestice srebra imaju veliku molekularnu masu, što su i bili razlozi za njegovu primenu u ovom istraživanju [25].

Marginalna mikropropustljivost je ispitivana nakon primene testiranih materijala u eksperimentalno prepiranu interadiksnu perforaciju na ekstrahovanim zubima. Tokom eksperimentalnog perioda zubi su čuvani u inkubatoru, u sunder natopljenom veštačkom tkinom tečnošću, kako bi se što bliže simulirali klinički uslovi [21]. U ovom istraživanju merena je najdublja tačka prodora boje uz napomenu da longitudinalnim sečenjem zub, koje se obično primenjuje, nije moguće precizno utvrditi u kojem delu katetiva je boja najdublje prodirla. Zbog toga se prodor boje pri određivanju mikropropustljivosti u uslovljima in vitro ne može direktno uporediti s kompleksnim mikrocurenjem u uslovima in vivo, ali se može smatrati pokazateljem moguće propustljivosti materijala u kliničkim uslovima [20].

Najmanji prodor boje u ovom istraživanju uočen je kod materijala CS. Vrednosti prodora boje kod MTA bile su nespoznate, ali bez statistički značajne razlike u odnosu na CS. S obzirom na to da su CS i MTA materijali sličnog hemijskog sastava, i njo­hove fizičke osobine mogu biti slične. Dobro rubno zaptivanje kalcijumsilikatnih cemenata se povezuje sa blagom ekspanzijom cementa prilikom vezivanja [27, 28], odnosno bioaktivnom pri­romod i stvaranjem hidroksiapatita na površini materijala u kontaktu s fosfatima iz tkivnih tečnosti [29].

Ipak, različitim metodama ispitivanja mikropropustljivosti uočeno je da MTA i slični kalcijumsilikatni cementi nemaju sposobnost apsolutnog hermetičkog zaptivanja [21, 22, 25, 30], što je u skladu s rezultatima ovog istraživanja. U grupi uzoraka ispunjenih materijalom CS izmerene su nespoznate vrednosti prodora boje u odnosu na uzorke ispunjene sa MTA, ali je prodor boje uočen u većem broju uzoraka nego u MTA grupi. Izmerene ne vrednosti prodora boje u MTA grupi bile su vrlo različite, a dobijeni rezultati su u skladu s nalazima De Deus (De Deus) i saradnika [30], koji su, ispitujući marginalnu mikropropustljivost različitih kalcijumsilikatnih cemenata (PC, MTA Angelus i MTA Bio), pisali o vrlo variabilnim rezultatima unutar grupa. Isti autori su mikropropustljivost otkrili u svim uzorcima, ali bez
statistički značajnih razlika u pogledu propustljivosti različitih kalcijumsilikatnih cemenata [30]. Suprotno tome, Hašem (Hašem) i saradnici [19] su uočili razlike u propustljivosti različitih komercijalnih kalcijumsilikatnih cemenata, smatrajući da različit hemijski sastav i različita brzina vezivanja cemenata može imati ulogu u adaptaciji materijala za zidove kavite. I studije u kojima je propustljivost kalcijumsilikatnih cemenata ispitana metodom prodora bakterija takođe su ukazale na različite rezultate. Tako su Montelano (Montelano) i saradnici [2] najizraženije mikrocurenje zabeležili u prvom danima primene materijala, dok su Pariroh (Pariroh) i saradnici [21] otkrili prodor bakterija značajno kasnije (između 39 i 73 dana eksperimenta), ističući pozitivan uticaj fosfatnog rastvora na pranjanje materijala.

Uopšteno posmatrajući, pregledom literature uočava se ne-konzistentnost rezultata u pogledu dobijenih vrednosti propustljivosti kalcijumsilikatnih cemenata u okviru različitih metoda ispitivanja. Iz dosadašnjih istraživanja može se zaključiti da brojni faktori utiču na mikropropustljivost MTA: od vrste boje, pH vrednosti boje, uslova čuvanja uzoraka pre eksperimenta, odnosno vremena vezivanja uzoraka materijala pre njihovog potapanja u boju [1]. Takođe, različiti rezultati mogu biti posledica razlika u prečniku i dubini perforacije, broja uzoraka, odnosno razlika u trajanju opservacionog perioda [30].

Zaključak

Zaključak

Najmanja marginalna mikropropustljivost, odnosno najbolje rubno zaptivanje zabeleženo je kod materijala CS i bilo je slično sa MTA. Mikropropustljivost materijala HA-CS bila je značajno veća u odnosu na materijal CS i kontrolni materijal MTA.