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Evaluation of Antibacterial Activity of Root Canal Sealers Used in Primary Teeth Against *E.faecalis*

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Abstract

Aim: The aim of this study was to assess the antibacterial activity of seven different root canal sealers used in primary teeth against Enterococcus faecalis.
Material and Method: The study was conducted using the agar diffusion inhibitory test. The sealers used were Sure-paste, Metapex, Tg Pex, Calcicure, Endoflas FS, Sealapex, $Ca(OH)_2$ mixed with %2 Chlorhexidine Gluconate solution and one control group material (Klorhex Oral Gel). The injectable form sealers and the freshly-mixed sealers were placed into the prepared wells on the agar plates which were inoculated with the Enterococcus faecalis. After incubation periods of 24 hours, 48 hours, and 72 hours, the zones of the growth inhibition were observed and measured. The average of 8 measurements for each material was taken. The data were compared using one-way analysis of variance (ANOVA) ($p<0.05$).
Results: Even though Sure-paste, Calcicure and $Ca(OH)_2$ powder mixed with Klorhex irrigation solution have shown antibacterial activity against E.faecalis, Metapex, Tgpex, Endoflas FS and Sealapex have not. Calcicure and $Ca(OH)_2$ powder mixed with Klorhex irrigation solution was more effective than Sure-paste ($p<0.05$). The largest zone of inhibition was found with Klorhex oral gel (control group). But, there was no significant differences between Klorhex oral gel, Calcicure and $Ca(OH)_2$ powder mixed with Klorhex irrigation solution. There were changes in the antibacterial activity of Sealapex from the 24-h period through the 48-h period and 24-h period through the 72-h period.
Conclusion: Based on the results of this study it can be concluded that $Ca(OH)_2$/Chlorhexidine gluconate and injectable $Ca(OH)_2$ pastes more effective than $Ca(OH)_2$, iodoform and zinc oxide eugenol mixtures.

Key Words: Agar diffusion, antibacterial, root canal sealer, primary teeth, Enterococcus faecalis.

Özet
Amaç: Süt dişlerinde kullanılan 7 farklı kök kanal dolgu materyalinin Enterococcus faecalis'e karşı antibakteriyel etkinliklerini in vitro ortamda değerlendirilmesi amaçlanmıştır.
Gereç ve Yöntem: Agar difuzyon yöntemi kullanılarak yaptığımız bu çalışmada Sure-paste, Matepex, Tg Pex, Calcicure, Endoflas FS, Sealapex, %2 lik Klorheksidin Glukonat solüsyonu ile $Ca(OH)_2$ (Kalsiyum hidroksit) tozundan hazırlanan pat ve kontrol grubu materyali (Klorhex Oral Gel) kullanılmıştır. Enjektable formda bulunan ve karıştırılarak hazırlanan patlar Enterococcus feacalis mikroorganizmasının inokule edildiği agar petrilerinde hazırlanan çukurlara yerleştirildi. 24, 48 ve 72. saat inhibisyon süresinden sonra, oluşan inhibisyon zonları gözlendi ve ölçüldü. Her bir materyal için elde edilen 8 ölçümün ortalamaları alındı. Veriler tek yönlü varyans analizi (ANOVA) ile karşılaştırıldı.($p <0.05$)
Bulgular: Çalışmada kullanılan materyallerden Sure-paste, Calcicure ve $Ca(OH)_2$ tozu/Klorhex irrigasyon solüsyonu karışımı E.faecalis suşları üzerinde antibakteriyel etkinlik göstermiş, Metapex, Tgpex, Endoflas FS ve Sealapex ise herhangi bir aktivite göstermemiştir. Calcicure ve $Ca(OH)_2$ tozu/Klorhex irrigasyon solüsyonu karışımı Sure-paste'den daha etkili bulunmuştur.($p<0.05$) Fakat, en geniş inhibisyon zonu Klorhex oral gel (kontrol grubu) ile bulundu. Fakat, Klorhex oral gel, Calcicure ve $Ca(OH)_2$ tozu/Klorhex irrigasyon solüsyonu karışımı arasında anlamlı bir fark yoktur. 24-saat süresinden 48-saat süresine kadar ve 24-saat süresinden 72-saat süresine kadar geçen zamanda Sealapex'in antibakteriyel etkinliğinde değişiklik olmuştur.
Sonuç: $Ca(OH)_2$ tozu/Klorhex irrigasyon solüsyonu karışımı ve enjektabl $Ca(OH)_2$ patlarının, $Ca(OH)_2$, iyodoform and çinko oksit ojenol karışımlardan ve $Ca(OH)_2$ bazlı bir kök kanal dolgu materyali olan Sealapex'den daha fazla antimikrobiyal etkinlikleri vardır.

Anahtar Kelimeler: Kök kanal dolgu patı, kalsiyum hidroksit, klorheksidin glukonat, süt dişi, enterococcus faecalis.

Introduction

Maintenance of the integrity of the primary dentition until the normal exfoliation period is vital for proper development and maturation of the child, maintaining arch length, arch symmetry, normal occlusion, function and esthetics[1-3]. Nonvital primary teeth with and without periapical pathosis should be retained until their normal exfoliation for preventing undesirable aberrations in the future dentition. So, endodontic treatment of these teeth has been a successful method. The success of endodontic treatment depends on the clinical and radiological state of the treated teeth, achieved through chemo-mechanical preparation of root canals elimination of infecting microorganisms and obturation of the root canal space with a good root canal sealer[1,4-6]. However, even after careful

cleaning and shaping of the root canal system, microorganisms might still remain in the tubules or lateral canals[7]. Therefore, to avoid the growth of these microorganisms, endodontic filling materials should have an antibacterial effect.

Root canals provide an ideal culture media for bacterial growth which harbor different types of microorganisms[1]. Tomie-Karovie and Jelinek[8], Edward and Nord[9] demonstrated that root canal infections of primary teeth are usually polymicrobial in nature. It is known that the predominant pathogens in the root canal are especially anaerobes[10,11]. E.faecalis was the most common facultative anaerobic bacteria isolated from the root canals with pulpal and periapical infections[12-14]. In addition to this, E.faecalis was used in various studies which evaluate antibacterial activity of root canal sealers[1,4,6,13,14].

The major goal of root canal treatment is the inhibition of microorganisms from root canal system and prevention of subsequent reinfection[15]. For this reason, to avoid the growth of the microorganisms, root canal sealers should have antibacterial effect.

The purpose of this study was to compare the antibacterial effect of different root canal sealers used in primary teeth .

Materials and Methods

The sealers and one control group material (Klorhex Oral Gel) tested for comparing antibacterial activity against E.faecalis in this study were; Sure-paste (Sure-endo, Sure Dent Co., Korea), Metapex (Meta Biomed Co., Korea), Tg Pex (Technical & General Ltd., London), Calcicure (Voco, Germany), Endoflas FS (Sanlor Laboratories, USA), Sealapex (Kerr, USA), $Ca(OH)_2$ (Calcium Hydroxide U.S.P, Sultan Healthcare Inc., USA) mixed with Klorhex irrigation solution (%2 Chlorhexidine Gluconate, Drogsan, Ankara, Turkey). The compositions of the sealers are shown in Table 1. Non-injectable materials as Sealapex and Endoflas FS were mixed according to manufacturers' instructions. $Ca(OH)_2$ powder and %2 chlorhexidine mixed the specified proportions (2gr calcium hydroxide powder per ml of %2 chlorhexidine) on the sterilized glass with spatula. After the pastes were prepared with toothpaste consistence, each freshly mixed dental material was back loaded into sterile 2ml syringes and kept ready.

Antibacterial activities of the sealers were evaluated against the E.faecalis (ATCC 29212) for Agar Diffusion Test. The strain was obtained from the Department of Microbiology, Faculty of Medicine of Kirikkale University. 8 petri dishes previously sterilized were prepared containing Mueller-Hinton agar, in the necessary volume to obtain 4 mm of thickness. Inocula from a 24 hour growth of the test organisms were added in sterile saline, incubated at 37 ºC, and allowed to grow to obtain a turbidity equivalent to the 0.5 McFarland Standard. Using a sterile swab the entire surface of the agar plate was swabbed 3 times to ensure even distribution of the inoculum. 8 wells (6mm diameter, 4mm depth) were made by removal of agar at equidistant points using sterile pipette tip, and these were filled immediately with sealers. Klorhex Oral Gel was used as a positive control. After the sealers setting, the agar plates were incubated at 37 ºC for 24 hours to allow the microorganisms to grow and reagents to diffuse through the culture medium. After incubation, the inhibition zones (mm) formed around the wells containing sealers were measured with a millimeter ruler and recorded at 24 hour, 48 hour and 72 hour intervals. The test was performed in 8 replicas, and the average reading with a standard deviation was calculated for each sealer tested (Table 2). The data were compared using one-way analysis of variance (ANOVA).

Table 1: Materials used in this study

MATERIAL NAME	MANUFACTURERS	COMPOSITIONS
Klorhex Oral Gel	Drogsan, Ankara, Turkey	%1 Chlorhexidine Gluconat
Sure-paste	Sure Dent Co., Korea	Calcium hydroxide, Barium sulfate, Thickening agent, Glycol derivatives, Purified water
Metapex	Meta Biomed Co., Korea	Iodoform, Calcium hydroxide, Silicone oil
Tg Pex	Technical & General Ltd., London	Ioodoform, Calcium hydroxide
Calcicure	Voco, Germany	Calcium hydroxide, Water, Cellulose derivatives, Barium sulfate
Endoflas FS	Sanlor Laboratories, USA	Barium sulfate, Zinc oxide, Eugenol, İodoform, Calcium hydroxide, Zinc acetate
Sealapex	Kerr, USA	Calsium oxide, Bismuth trioxide, Zinc oxide, Sub-Micron silica, Titanium dioxide, Zinc stearate , Tricalcium Phosphate, Ethyl toluene sulfonamide, Methylene methyl salicylate, İzobutil salicylate, Pigment
Klorhex irrigation solution	Drogsan, Ankara, Türkiye	%2 Chlorhexidine Gluconat
Calcium Hydroxide U.S.P,	Sultan Healthcare Inc., USA	$Ca(OH)_2$ powder

Wound edges were closed with primary interrupted sutures (Fig 3.c,3.d). Histopathological examination revealed cystic lesion with an squamous epithelial lining with prominent granular layer and cyst content of lamellar keratin (Fig.4).

Results

The mean diameter of inhibition haloes (mm) around the agar wells and standard deviations of the antibacterial activity for freshly mixed, 24 hour old, 48 hour old and 72 hour old endodontic sealers used in primary teeth are shown in Table 2. However, wells filled with Klorhex oral gel called positive control group showed zone of growth inhibition in all intervals. Even though Sure-paste, Calcicure and $Ca(OH)_2$ powder mixed with Klorhex irrigation solution have shown antibacterial activity against E.faecalis, Metapex, Tgpex, Endoflas FS and Sealapex have not. Calcicure and $Ca(OH)_2$ powder mixed with Klorhex irrigation solution was more effective than Sure-paste ($p<0.05$).

	Duration of incubation; mean diameter (SD) of inhibition zone (mm)[a]		
Root canal sealers	**24 h**	**48 h**	**72 h**
Sure-paste	0	7,625 (3,77)	8 (4,14)
Metapex	0	0	0
Tg pex	0	0	0
Calcicure	12,375 (0,51)	12,375 (0,51)	12,375 (0,51)
Endoflas FS	0	0	0
Sealapex	0	0	0
$Ca(OH)_2$ powder / Klorhex irrigation solution	12,75 (0,46)	12,75 (0,46)	12,75 (0,46)
Control group (Klorhex Oral Gel)	12,75 (0,88)	12,75 (0,88)	12,5 (1,06)

Table 2. Mean diameter of zones of growth inhibition with 7 root canal sealers and control group (Klorhex oral gel) [a]Each value is the mean of 8 measurement; SD = standart deviation

The largest zone of inhibition was found with Klorhex oral gel (control group). But one-way ANOVA indicated no significant differences between Klorhex oral gel, Calcicure and $Ca(OH)_2$ powder mixed with Klorhex irrigation solution.
There was no change in the antibacterial activity of all of the sealers except that Sealapex from the 24-h period through the 48-h period and 24-h period through the 72-h period. From the 48-h period through the 72-h period, there was no change in the antibacterial activity of all of the sealers.

Discussion

However, even after instrumentation, irrigation and intracanal dressing procedures in endodontic treatment, total elimination of microorganisms from the root canal system is not possible because of the complexity of the root canal system. Therefore, both intracanal medication and root canal filling materials should have an antibacterial activity[16-18].

Generally, three in vitro techniques have been used for evaluating antibacteriel activities of materials which get infection under control: the diluation method, which yields a quantitive result for the amount of antimicrobial agent that is required; the agar diffusion method, which gives an inhibition zone around the wells containing the agent: and the direct exposure method, which relies on direct and close contact between the test microorganism and the tested material. All of these methods have advantages and disadvantages. The disadvantage of the agar diffusion test is that the result of this method depens on the solubility and diffusibility of the tested material. For example, a material which diffuses more easily than others, may have larger zones of microbial growth inhibition. The agar diffusion test used in this study is one of the most frequently used methods for evaluating the antimicrobial activity of root canal sealers and it permits to compare the root canal sealers against the test microorganisms directly[17,19-21].

The contrubition of microorganisms to indication and development of pulpal and periapical infections are mixed and polimicrobial, with predominance of obligate anaerobes, facultative anaerobes and rarely aerobes[22,23]. E.faecalis, a facultative anaerobic microorganism, has been used in various studies of antibacterial properties as used in this study, because of it is the most resistant species to eliminate from root canal and can survive in filled root canals. It is frequently isolated failure of the endodontic treatments. E.faecalis has been reported to survive in an alkaline environment and has been resistant to antibacterial agents. In addition to all these features, E.faecalis isolation

and identification is relatively easy, so it was used in various antibacterial efficiency studies[21,22,24-27].

Calcium hydroxide has also been used as a root filling material for primary teeth alone[28,29] or mixed with iodoform ; it is commercially available as Vitapex[30], Metapex[31] and TgPex. Calcium hydroxide is frequently used intracanal medication because of its consistent antibacterial activity[32,33]. The antibacteriel effect of $Ca(OH)_2$ is related to its ionic dissociation into calcium and hydroxil ions and their toxic effects on bacteria, acts by inhibiting cytoplasmic membrane enzymes[34]. However, specific microbes, such as E. faecalis are resistant to $Ca(OH)_2$[12]. But, there is respectable variation in the release of hydroxyl ions and therefore in the antibacterial activity for different composition of these materials[35].

Chlorhexidine (CHX) is a broad-spectrum antimicrobial agent that has been reported to be an effective medicament in endodontic therapy[35]. In addition, it is also effective against strains resistant to $Ca(OH)_2$[37]. Depending on this information, Basrani et all.[38] and Gomes et all.[39] demonstrated that CHX was effective against E.faecalis, but $Ca(OH)_2$ alone had no effect. Other studies showed that the antibacterial effect of $Ca(OH)_2$ increased significantly when mixed with chlorhexidine[40,41]. CHX gel or $Ca(OH)_2$ powder mixed with CHX gel may have promise as an intracanal medicament in teeth with infected canals and may have use of a root canal cement, but further research is required[42].

In 1928 Walkoff [1] introduced iodoform as a root canal filling material in primary teeth and proved to be a bactericidal, non-irritant radiopaque material. On the other hand, $Ca(OH)_2$ + Iodoform exhibited no antibacterial activity against most pure cultures in agar diffusion tests[43]. Nurko and Garcia[30] reported that Vitapex (calcium hydroxide/iodoform paste) used as a root canal filling material for infected primary teeth, found clinically and radiographically successful. In the present study, the Vitapex and showed the worst antimicrobial results against E.faecalis as the previous studies[1,44]. The weak activity may be partially explained by the fact that $Ca(OH)_2$ an ingredient of these sealers has been demonstrated to interfere with the antiseptic capacity of dyadic combinations of endodontic medicaments (45). Endoflas is a resorbable paste which contains similar components as Vitapex, with the addition of zinc-oxide and eugenol[46]. In 2007, Reddy and Ramakrishna[1] found weak inhibitory effect against Gram-positive bacteries as E.faecalis for ZOE. In this study, ZOE containing Endoflas showed no antibacterial effect.

Sealapex, is an calcium hydroxide-based sealer, used in root canal treatment for primary teeth. It showed the high success rate, the absence of any adverse response and the resorption of the extruded material[47]. Sealapex, releases calcium hydroxide in the fresh state. A study that evaluate the antibacterial activity of five different endodontic sealers, using both Direct Contact Test (DCT) and Agar Diffusion Test (ADT), According to DCT results, Sealapex inhibit bacterial growth when freshly mixed but lose their activity over time[48]. Depending to this data, absence of the antibacterial effect for Sealapex may be explained.

Based on the results of this in vitro study, it was concluded that $Ca(OH)_2$/Chlorhexidine gluconate mixture and injectable $Ca(OH)_2$ pastes have more antimicrobial efficiency than $Ca(OH)_2$, iodoform and zinc oxide eugenol mixtures and a $Ca(OH)_2$ based root canal sealer called Sealapex.

References

1. Reddy S, Ramakrishna Y. Evaluation of antimicrobial efficacy of various root canal filling materials used in primary teeth: a microbiological study. J Clin Pediatr Dent 2007;31:193-8.
2. Cox ST Jr, Hembree JH Jr, McKnight JP. The bactericidal potential of various endodontic materials for primary teeth. Oral Surg Oral Med Oral Pathol 1978;45:947-54.
3. Goerig AC, Camp JH. Root canal treatment in primary teeth: a review. Pediatr Dent 1983;5:33-7.
4. Wright KJ, Barbosa SV, Araki K, Spångberg LS. In vitro antimicrobial and cytotoxic effects of Kri 1 paste and zinc oxide-eugenol used in primary tooth pulpectomies. Pediatr Dent 1994;16:102-6.
5. Camp JH. Pulp therapy for primary and young permanent teeth. Dent Clin North Am 1984;28:651-68.
6. Bodrumlu E, Avşar A, Eğilmez T. Üç farklı rezin esaslı kök kanal patının antimikrobiyal etkinliğinin değerlendirilmesi. GÜ Diş Hek Fak Derg 2008;25:19-22.
7. Oguntebi BR. Dentine tubule infection and endodontic therapy implications. Int Endod J 1994;27:218-22.
8. Tomie-Karovie K, Jelinek E. Comparative study of the bacterial flora in the surroundings, the root canals and sockets of deciduous molars. Int Dent J 1971;21:375-88.
9. Edwards S, Nord CE. Identification and characterization of micro-organisms isolated from infected primary teeth. J Int Assoc Dent Child 1972;3:15-8.
10. Brauner AW, Conrads G.Studies into the microbial spectrum of apical periodontitis. Int Endod J 1995;28:244-8.
11. Le Goff A, Bunetel L, Mouton C, Bonnaure-Mallet M. Evaluation of root canal bacteria and their antimicrobial susceptibility in teeth with necrotic pulp. Oral Microbiol Immunol 1997;12:318-22.
12. Dahlén G, Samuelsson W, Molander A, Reit C. Identification and antimicrobial susceptibility of enterococci isolated from the root canal. Oral Microbiol Immunol 2000;15:309-12.
13. Kayaoglu G, Erten H, Alaçam T, Ørstavik D. Short-term antibacterial activity of root canal

sealers towards Enterococcus faecalis. Int Endod J 2005;38:483-8.
14. Mickel AK, Nguyen TH, Chogle S. Antimicrobial activity of endodontic sealers on Enterococcus faecalis. J Endod 2003;29:257-8.
15. Grossman L. Antimicrobial effect of root canal cements. J Endod. 1980;6:594-7.
16. Lai CC, Huang FM, Yang HW, Chan Y, Huang MS, Chou MY, Chang YC. Antimicrobial activity of four root canal sealers against endodontic pathogens. Clin Oral Investig. 2001;5:236-9.
17. Gomes BP, Pedroso JA, Jacinto RC, Vianna ME, Ferraz CC, Zaia AA, de Souza-Filho FJ. In vitro evaluation of the antimicrobial activity of five root canal sealers. Braz Dent J. 2004;15:30-5.
18. Pizzo G, Giammanco GM, Cumbo E, Nicolosi G, Gallina G. In vitro antibacterial activity of endodontic sealers. J Dent. 2006;34:35-40.
19. Estrela C, Bammann LL, Pimenta FC, Pécora JD. Control of microorganisms in vitro by calcium hydroxide pastes. Int Endod J. 2001;34:341-5.
20. al-Khatib ZZ, Baum RH, Morse DR, Yesilsoy C, Bhambhani S, Furst ML. The antimicrobial effect of various endodontic sealers. Oral Surg 1990;70:784-90.
21. Bodrumlu E, Semiz M. Antibacterial activity of a new endodontic sealer against Enterococcus faecalis. J Can Dent Assoc. 2006;72:637.
22. Sundqvist G, Figdor D, Persson S, Sjögren U.Microbiologic analysis of teeth with failed endodontic treatment and the outcome of conservative re-treatment. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 1998;85:86-93.
23. Peciuliene V, Balciuniene I, Eriksen HM, Haapasalo M. Isolation of Enterococcus faecalis in previously root-filled canals in a Lithuanian population. J Endod. 2000;26:593-5.
24. Fuss Z, Weiss EI, Shalhav M. Antibacterial activity of calcium hydroxide-containing endodontic sealers on Enterococcus faecalis in vitro. Int Endod J. 1997;30:397-402.
25. Lin YH, Mickel AK, Chogle S. Effectiveness of selected materials against Enterococcus faecalis: part 3. The antibacterial effect of calcium hydroxide and chlorhexidine on Enterococcus faecalis. J Endod. 2003;29:565-6.
26. Shabahang S, Torabinejad M. Effect of MTAD on Enterococcus faecalis-contaminated root canals of extracted human teeth. J Endod. 2003;29:576-9.
27. Orstavik D, Haapasalo M. Disinfection by endodontic irrigants and dressings of experimentally infected dentinal tubules. Endod Dent Traumatol. 1990;6:142-9.
28. Nadkarni U, Damle SG. Comparative evaluation of calcium hydroxide and zinc oxide eugenol as root canal filling materials for primary molars: a clinical and radiographic study. J Indian Soc Pedod Prev Dent. 2000;18:1-10.
29. Rosendahl R, Weinert-Grodd A. Root canal treatment of primary molars with infected pulps using calcium hydroxide as a root canal filling. J Clin Pediatr Dent. 1995;19:255-8.
30. Nurko C, Ranly DM, García-Godoy F, Lakshmyya KN. Resorption of a calcium hydroxide/iodoform paste (Vitapex) in root canal therapy for primary teeth: a case report. Pediatr Dent. 2000;22:517-20.
31. http://www.metadentalco.com. (Access date: 13.09.2008)
32. Siqueira JF Jr, Lopes HP. Mechanisms of antimicrobial activity of calcium hydroxide: a critical review. Int Endod J. 1999;32:361-9.
33. Fava LR, Saunders WP. Calcium hydroxide pastes: classification and clinical indications. Int Endod J. 1999;32:257-82.
34. El Karim I, Kennedy J, Hussey D. The antimicrobial effects of root canal irrigation and medication. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2007;103:560-9.
35. Morrier JJ, Benay G, Hartmann C, Barsotti O. Antimicrobial activity of Ca(OH)2 dental cements: an in vitro study. J Endod. 2003;29:51-4.
36. Delany GM, Patterson SS, Miller CH, Newton CW. The effect of chlorhexidine gluconate irrigation on the root canal flora of freshly extracted necrotic teeth. Oral Surg Oral Med Oral Pathol. 1982;53:518-23.
37. Siqueira JF Jr, Batista MM, Fraga RC, de Uzeda M. Antibacterial effects of endodontic irrigants on black-pigmented gram-negative anaerobes and facultative bacteria. J Endod. 1998;24:414-6.
38. Basrani B, Tjäderhane L, Santos JM, Pascon E, Grad H, Lawrence HP, Friedman S. Efficacy of chlorhexidine- and calcium hydroxide-containing medicaments against Enterococcus faecalis in vitro. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2003;96:618-24.
39. Gomes BP, Souza SF, Ferraz CC, Teixeira FB, Zaia AA, Valdrighi L, Souza-Filho FJ. Effectiveness of 2% chlorhexidine gel and calcium hydroxide against Enterococcus faecalis in bovine root dentine in vitro. Int Endod J. 2003;36:267-75.
40. Ercan E, Dalli M, Dülgergil CT. In vitro assessment of the effectiveness of chlorhexidine gel and calcium hydroxide paste with chlorhexidine against Enterococcus faecalis and Candida albicans. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2006;102:27-31.
41. Haenni S, Schmidlin PR, Mueller B, Sener B, Zehnder M. Chemical and antimicrobial properties of calcium hydroxide mixed with irrigating solutions. Int Endod J. 2003;36:100-5.
42. Athanassiadis B, Abbott PV, Walsh LJ.The use of calcium hydroxide, antibiotics and biocides as antimicrobial medicaments in endodontics. Aust Dent J. 2007; 52:64-82.
43. Tchaou W.S., Turng B.F., Minah G.E. and Coll J.A. Inhibition of pure cultures of oral bacteria by root canal filling materials. Pediat Dent. 1996;18:444-9.
44. Amorim Lde F, Toledo OA, Estrela CR, Decurcio Dde A, Estrela C. Antimicrobial analysis of different root canal filling pastes used in pediatric dentistry by two experimental methods. Braz Dent J. 2006;17:317-22.
45. Seow W.K. The effects of dyadic combinations of endodontic medicaments on microbial growth inhibition. Pediat Dent. 1990;12:292-7.
46. Fuks AB, Eidelman E, Pauker N. Root fillings with Endoflas in primary teeth: a retrospective study. J Clin Pediatr Dent. 2002;27:41-5.
47. Sari S, Okte Z. Success rate of Sealapex in root canal treatment for primary teeth: 3-year follow-up.

Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2008;105:93-6.

48. Cobankara FK, Altinöz HC, Ergani O, Kav K, Belli S. In vitro antibacterial activities of root-canal sealers by using two different methods. J Endod. 2004;30:57-60.

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