
Penetration of propylene glycol into dentine

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Abstract

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Aim This study aimed to evaluate penetration of propylene glycol into root dentine.

Methodology Safranin O in propylene glycol and in distilled water were introduced into root canals with and without artificial smear layer. Dye diffusion through dentinal tubules was determined spectrophotometrically. The time required for dye to exit through the apical foramen using propylene glycol and distilled water as vehicles was also determined. The extent and areas of dye

penetration on the split surfaces of roots were assessed using Adobe Photoshop and NIH Image Software.

Results Propylene glycol allowed dye to exit faster through the apical foramen. The area and depth of dye penetration with propylene glycol was significantly greater than with distilled water ($P < 0.0001$). Smear layer significantly delayed the penetration of dye.

Conclusion Propylene glycol delivered dye through the root canal system rapidly and more effectively indicating its potential use in delivering intracanal medicaments.

Keywords: dentine, diffusion, intracanal medicaments, penetration, propylene glycol.

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Introduction

Previous studies have shown that bacteria in infected root canals and periradicular tissues are capable of invading and residing deeply within dentine and in cementum around the periapex (Ando & Hoshino 1990, Kiryu *et al.* 1994, Peters *et al.* 2001). Furthermore, it has been demonstrated that although bacteria in artificial smear layers and prepared reservoir channels in deeper layers of root dentine could be eliminated by procedures such as ultrasonic irrigation with NaOCl (Huque *et al.* 1998), microorganisms within fins and isthmuses could still remain viable (Sato *et al.* 1996). Such microorganisms may cause root canal treatment to fail. Thus, the placement of medicaments between appointments may be necessary to disinfect root canals and to reduce periapical pathosis, thus preventing bacteraemia and other local or systemic immunological reactions (Debelian *et al.* 1994, Walton & Rivera 1996, Murray & Saunders 2000).

Studies have been conducted on the efficacy of a mixture of antibiotics against various forms of oral infections, including those of endodontic origin (Hoshino *et al.* 1992, Sato *et al.* 1993, Hoshino *et al.* 1996, Sato *et al.* 1996). These were based on the concept of Lesion Sterilization and Tissue Repair (LSTR) therapy. Results of these studies proved to be highly promising as the drug mixture, consisting of metronidazole, ciprofloxacin and minocycline, known as 3Mix, was found to be effective against various oral bacteria from different sources. However, a suitable vehicle to deliver the drug mixture into infected root canals would be helpful. In choosing the appropriate vehicle, one factor that needs to be considered is its ability to facilitate better diffusion of medicaments through root dentine and possibly cementum even in the presence of anatomical aberrations such as fins, isthmuses and blocked canals. Diffusion into the surrounding periradicular tissues may also be an advantage.

Propylene glycol (1,2-propanediol), a dihydric alcohol, is a vehicle that has potential for use in root canal treatment. Its chemical formula is $\text{CH}_3\text{CH}(\text{OH})\text{CH}_2\text{OH}$ and it has a molecular weight of 76.09 (United States Pharmacopeia 1989). Seidenfeld & Hanzlik (1932) described propylene glycol and conducted studies on its use as a

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vehicle and pharmaceutical solvent for preparations in medicine. It has been reported to be a widely used vehicle for various pharmaceutical and commercial products such as drugs, cosmetics and foods (Morshed *et al.* 1988). In addition, it has also been used extensively for caries detection as a constituent of Caries Detector® (Fusayama 1988). In endodontics, it had been used as a vehicle for calcium hydroxide (Saijo 1957, Laws 1962, Laws 1971, Simon *et al.* 1995). Unfortunately, no studies have been conducted to determine the extent of penetration of propylene glycol in dentinal tubules or the time required for propylene glycol to diffuse through the root canal system.

The purpose of this study was therefore to determine the efficiency of propylene glycol to diffuse into dentine and through the root canal system using a dye diffusion protocol.

Materials and methods

Experimental procedure 1: Movement and amount of dye in propylene glycol or distilled water across open dentinal tubules with and without smear layer

Preparation of specimens

Ten extracted maxillary central incisors that were not root filled, were free of caries and cracks and stored in 70% alcohol, were used in this study. The histories of the teeth were not known. The crowns of the teeth were removed at the level of the cemento-enamel junction with a high speed bur under water coolant spray. Cementum covering the coronal one-third of roots was removed parallel to the root canal. To ensure an even removal of cementum and to make parallel the remaining root dentine, a 1.5-mm orthodontic wire attached to the head of an air turbine handpiece and parallel to the diamond bur (SF-13 Dia-Burs, Mani Inc, Tochigi, Japan) was inserted into the root canal. This served as a pivot which the bur followed during cementum removal. The root canals were enlarged using a no. 3 Peeso reamer to a depth of 7 mm.

Removal of smear layer

To remove smear layer, dentinal debris and soft tissue adherent to the cementum surface of roots, teeth were placed in a Sono Cleaner 50Z ultrasonic bath (Kaijo, Tokyo, Japan) with 5% NaOCl (Wako Pure Chemical, Osaka, Japan). The root canals were further irrigated ultrasonically with 5% NaOCl using an ultrasonic unit (Solfy, Morita, Osaka, Japan).

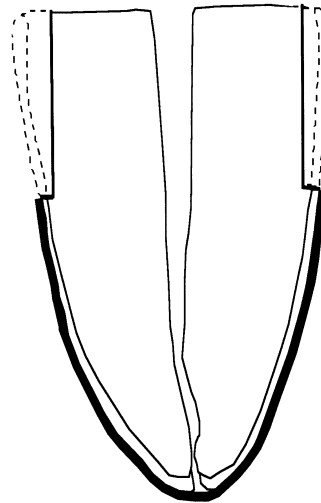


Figure 1 Schematic illustration of specimen preparation for experimental procedure 1. Dotted lines represent area of the root removed and exposed, whilst the heavy line surrounding the lower two-thirds represents the area covered with inlay wax.

Preparation and application of smear layer

In order to standardize the presence of smear layer experimentally, decalcified and pulverized dentine with dental plaque was closely adapted to the root canal walls using a finger spreader, in accordance with the method developed previously (Huque *et al.* 1998).

Application of dye

The specimens were dried and the remaining apical two-thirds of each root were covered with inlay wax (Fig. 1). Ten microliters of 0.1 mol L⁻¹ safranin O (Schmid GmbH, Köngen, Germany) was introduced into the root canals and sealed with inlay wax. The prepared root samples were placed in individual vials, each containing 2 mL of propylene glycol (Wako Pure Chemical, Osaka, Japan) for the propylene glycol group or distilled water for the distilled water groups.

Measurement of dye released through the dentinal tubules

The amount of dye released through the dentinal tubules from the root canals was measured using a U-3200 spectrophotometer (Hitachi, Tokyo, Japan) at different times. The procedure was repeated four times on each of the 10 samples after changing the conditions of the root canals, i.e. with or without smear layer, as well as the vehicle used for safranin O, i.e. distilled water or propylene glycol. Thus, these conditions were propylene glycol without

smear layers, distilled water without smear layer, propylene glycol with smear layer and distilled water with smear layer.

Forty-eight-hour readings from the samples were compared using ANOVA.

Experimental procedure 2: Time and depth of dye penetration through the root canal system and beneath fin areas

Sixty extracted human teeth, consisting mostly of maxillary premolars and mandibular incisors, were used. After crown removal, pulp tissue and debris from the root canals were removed. Canal patency was established by inserting a smooth broach into the root canals 1 mm short of the apical foramen. The presence of fins in root canals were confirmed with the aid of a SMZ-10 stereomicroscope (Nikon, Tokyo, Japan). The orifices of canals were enlarged to a diameter of 2 mm. The root canals were irrigated ultrasonically with 5% NaOCl to remove smear layer and pulp debris. After ultrasonic irrigation, outer root surfaces were covered with inlay wax leaving the apical foramina open. This was confirmed by inserting a smooth broach through the root canal. The specimens were separated randomly into equal groups of 30 roots each: group I for application of safranin O in propylene glycol, and group II for application of safranin O in distilled water. After introducing 10 μL of 0.1 mol L⁻¹ safranin O solution into each canal, the orifices of roots were sealed with inlay wax. To avoid contamination by dye exiting through the apical foramen, coronal ends of roots were suspended from the cover of a container. This container also served to provide a moist environment by the placement of wet tissue papers. All samples were maintained at 37°C, and the time for the exit of dye through the apical foramen was noted with the appearance of dye solution at the periapex. After 24 h, the roots were split into two using a pair of straight crown scissors and the depth of dye penetration through the dentinal tubules was measured by directly scanning the samples with the use of Canon IX-4025 scanner (Canon, Tokyo, Japan). The scanned images were viewed using Adobe Photoshop 4.0J (Adobe Systems, California, USA). These were subsequently treated to delineate the split surfaces of the root from the root canal wall and its background. RGB images of the specimens were subsequently produced for analysis. Image analysis was performed to determine the area of dye penetration using a MacIntosh G3 computer and the public domain NIH Image program (developed at the U. S. National Institutes of Health and available from the Internet by anonymous

FTP from zippy.nimh.nih.gov or on floppy disk from the National Technical Information Service, Springfield, Virginia, part number PB95-500195GEI) as shown in Figure 2.

An additional 24 maxillary premolar roots were used to observe the extent of dye penetration through the root canal system. These specimens were separated randomly into two groups and treated in a similar manner as described above, except that the roots were not covered with inlay wax.

Readings from the samples were compared using the paired *t*-test.

Results

Experimental procedure 1

Figure 3 shows the mean amount (in micrograms) of dye released by the teeth under different conditions after 1 h and 48 h. Although dye release was noted in three groups, i.e. propylene glycol without smear layer (PGNS), propylene glycol with smear layer (PGS) and distilled water without smear layer (DWNS) during the first hour, the greatest amount of release was noted in the specimens where safranin O in propylene glycol was applied in the absence of smear layers. The presence of smear layer affected dye diffusion through the tubules greatly during the first hour, when distilled water was used as the vehicle. Statistical analysis using ANOVA revealed that there are significant differences in the amount of dye released after a 48-hour period amongst the four conditions ($P < 0.0001$).

Experimental procedure 2

Figure 4 shows the distribution of the time taken for dye to exit the apical foramen of the specimens. More than half of the specimens that were treated with safranin O in propylene glycol allowed some dye to be released in less than 1 min. In contrast, 53% of the specimens treated with safranin in distilled water showed no dye leakage through the apical foramen in a 24-hour period. The overall median time for some dye to be released was 0.60 min for the propylene glycol group ($n = 28$) and 2.19 min for the distilled water group ($n = 14$).

Propylene glycol allowed for a greater surface area to be penetrated with dye as compared with distilled water ($P < 0.0001$) (Figs 2, 5a). The second group of specimens that were not covered with inlay wax revealed similar statistically significant findings (Fig. 5b). In addition, it

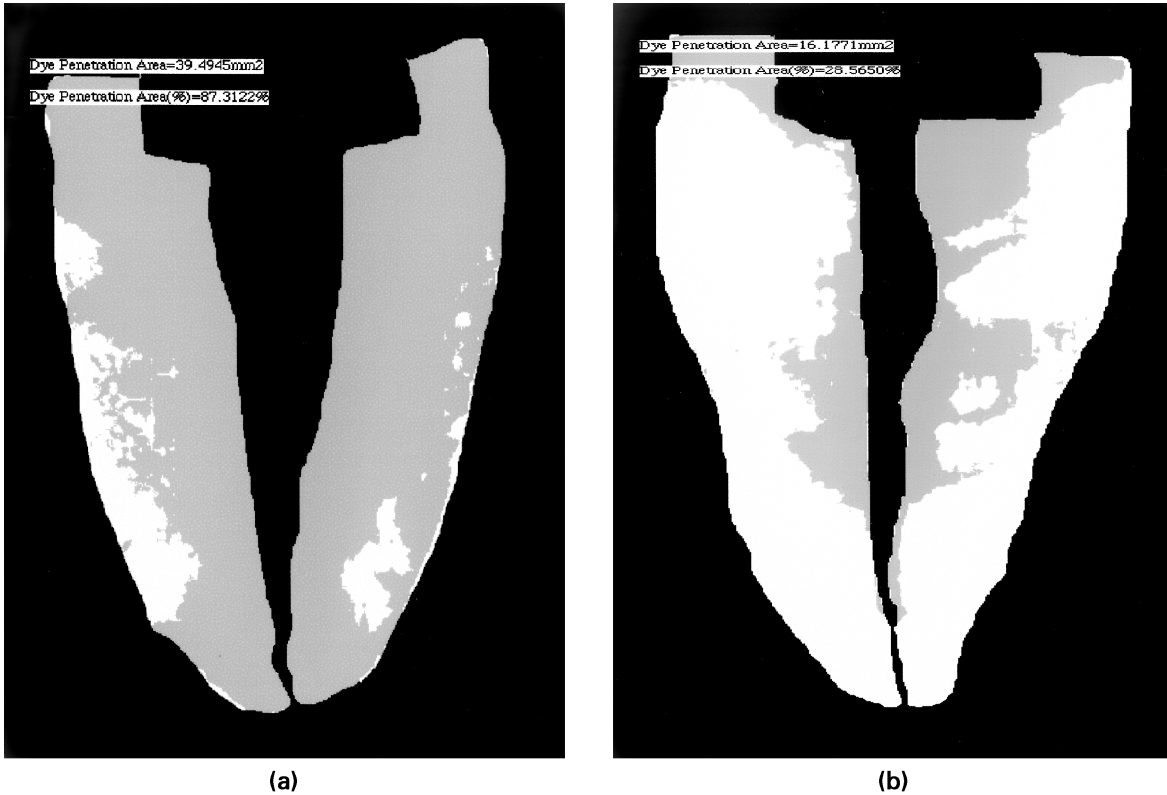


Figure 2 NIH Image analysis of specimen showing percentage of surface area penetrated by dye (indicated by dark gray areas) when propylene glycol (a) and distilled water (b) were used as vehicles.

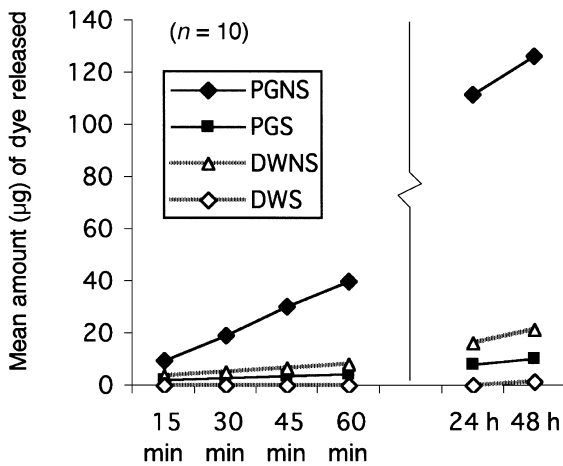


Figure 3 Mean amount of dye released through open dentinal tubules within 1 h and 48-hour periods under different conditions. Significant difference exists amongst the four conditions ($P < 0.0001$). (PGNS – propylene glycol without smear layer; PGS – propylene glycol with smear layer; DWNS – distilled water without smear layer; DWS – distilled water with smear layer)

was observed that propylene glycol actually diffused through cementum in eight of 12 specimens. This was not observed in any of the samples treated with safranin O in distilled water.

Discussion

Propylene glycol is a colourless liquid with a mildly acrid smell and somewhat sweet taste. It has been reported to offer all the advantages of ethylene glycol, with low toxicity and no demonstrable cumulative effects in experimental animals (Seidenfeld & Hanzlik 1932). Its antimicrobial activity for general use has also been documented. Olitzky (1965) has reported that concentrated solutions of propylene glycol have a marked germicidal efficiency and that its use as a vehicle may provide a potential for preventing or treating microbial infections. It is considered by the Council on Pharmacy and Chemistry of the American Medical Association as a harmless constituent for pharmaceutical products, particularly when administered for a limited period and within acceptable daily intake as prescribed by the World Health

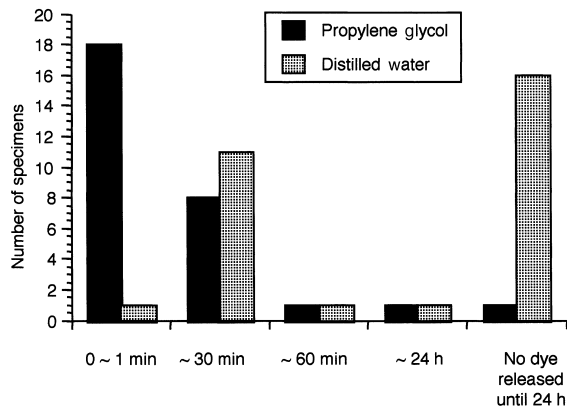


Figure 4 Time of dye exit from the apical foramen.

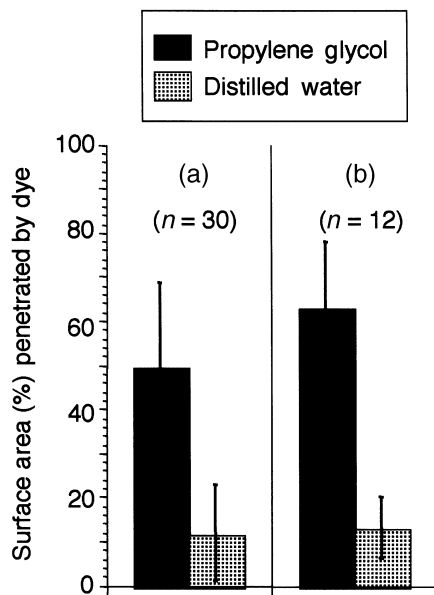


Figure 5 Mean area penetrated (in percentage) by dye on the split surfaces when root surfaces were covered with (a) or without (b) inlay wax. Significant difference exists in both conditions ($P < 0.0001$).

Organization, that is, 25 mg kg⁻¹ body weight (Kollöffel *et al.* 1996). Glover & Reed (1996) reported adverse effects associated with propylene glycol, but it is interesting to note that this occurred following excessive ingestion of a propylene glycol-containing product. Based on chronic toxicity data, the use of propylene glycol when given in small amounts does not produce any deleterious effects (Ruddick 1972). Aside from the fact that it is a well-recognized vehicle for drugs, propylene glycol has also been found to be less cytotoxic than other commonly

used vehicles for intracanal medicaments and possesses antibacterial properties that are highly beneficial in endodontic treatment (Bhat & Walkevar 1975, Thomas *et al.* 1980). It possesses hygroscopic properties that allow absorption of water, resulting in a sustained release of intracanal medicaments for prolonged periods (Fava & Saunders 1999).

Distilled water is one of the commonest vehicles used in delivering medicaments into the root canal. It is an aqueous substance having a high viscosity and surface tension (Ho 1983) that causes a high degree of solubility when the paste that it forms with the medicament comes in direct contact with tissue and tissue fluids, resulting in its rapid solubilization and resorption by tissue macrophages (Fava & Saunders 1999). When used as a vehicle for calcium hydroxide, there is the possibility of rapid carbonation of calcium hydroxide from the atmospheric carbon dioxide or that generated by tissue decomposition resulting into the formation of carbonates that do not possess any therapeutic value (Simon *et al.* 1995). This is not favourable clinically, because it may delay the resolution of infection (Esberard 1992).

Bacteria on the surfaces of root canals may be easily removed and killed by various endodontic procedures, such as ultrasonic irrigation with NaOCl (Huque *et al.* 1998). However, bacteria which invade and reside deeply within dentinal tubules (Ando & Hoshino 1990) may survive such irrigation procedures (Huque *et al.* 1998) if the medicaments introduced into the root canal are not delivered efficiently. These remaining bacteria may be capable of causing infections once they reach the periradicular tissues.

Sodium hypochlorite, the most commonly used irrigant in endodontic therapy (West & Roane 1998), in conjunction with ultrasonics, is capable of eliminating bacteria in smear layer and artificially prepared reservoir channels in root dentine (Huque *et al.* 1998). In order to closely simulate clinical situations, sodium hypochlorite was used as irrigant in this study. However, since sodium hypochlorite alone may not effectively remove smear layer, the use of ultrasonics was employed to enhance the cleansing effect of sodium hypochlorite and improve its ability to remove smear layer as demonstrated by Huque *et al.* (1998).

Some of the root canal medicaments commonly used, such as formocresol, are volatile, protein-denaturing agents in liquid form and thus, do not require the use of vehicles to exert their bactericidal effect. However, rather high concentrations are necessary to kill bacteria in infected canals. Thus, use of these agents not only leads to elimination of microorganisms but to damage of

periradicular tissues, as well. The antibacterial efficacy of 3Mix had been confirmed against oral infections both *in vitro* and *in vivo* (Hoshino *et al.* 1988, Hoshino *et al.* 1989, Sato *et al.* 1992, Sato *et al.* 1993, Hoshino *et al.* 1996, Sato *et al.* 1996). Thus 3Mix could effectively eliminate microorganisms remaining in infected canals, particularly those residing in areas that could not be reached by root canal irrigants. However, an efficient vehicle may be helpful to allow this medicament to more effectively penetrate such areas and thus kill the remaining bacteria. It has been demonstrated that when propylene glycol was used to deliver the dye into the root canal, greater amounts of solute were allowed to penetrate through the dentinal tubules. The results obtained with this method were much higher than when distilled water was used as vehicle. Smear layer delayed the release of dye, even when propylene glycol was used as the carrier. This underlines the need to remove smear layer before introducing medicaments into the root canal if the maximum effect is to be achieved.

Results of the present study also indicated that dye in propylene glycol passed through the main canal of the root and out of the apical foramen with relative ease. In contrast, the dye in distilled water was slow, or even failed, to exit through the apical foramen. Before splitting the tooth, it was noted that in samples where dye in distilled water was used, the solution remained in the orifice. The high surface tension of distilled water may have delayed the efficient penetration of dentinal tubules significantly (Tasman *et al.* 2000). This accounts for the limited staining, as well as the much reduced area and depth of dye penetration of these samples which was confined mainly in the coronal end of the roots. Propylene glycol, although viscous as compared with distilled water, has a low surface tension. This gives it an advantage of being able to penetrate through dentinal tubules that was not observed when distilled water was used as a vehicle.

In observing the time and depth of dye penetration through the root canal system, including dentinal tubules, the root specimens were either covered entirely with inlay wax (except for the apical foramen) or left uncovered (except for the coronal opening). The results of the present study showed that there was no difference in the results obtained whether the roots were covered or not. In both instances, propylene glycol managed to penetrate deeply into the deeper layers of root dentine.

Another aim of the present study was to develop a standardized procedure that would allow testing of the efficiency of vehicles for intracanal medicaments to diffuse through dentinal tubules. A part of the study used the same set of specimens that were subjected to different

vehicles and different experimental conditions. This allowed for a more accurate observation and comparison of the results that would not have been possible if different sets of teeth, whose histories and physical condition varied greatly, were used. Analysis of the split halves of the specimen was done using Adobe Photoshop and NIH Image Program. This enabled a uniform, efficient and bias-free observation of the actual depth of dye penetration in the individual specimens. In addition, the procedure permitted the actual measurement of the total surface area that was penetrated by the dye. This method would therefore be a very useful tool to visualize and measure the areas and extent of microleakage in dye penetration studies.

Conclusion

The results of this study indicate that propylene glycol could quickly and efficiently deliver dye through the root canal system. This suggests that propylene glycol may be useful in delivering intracanal medicaments into the root canal. The combination of propylene glycol and dye has the potential to allow for the visualization of tooth structures that could possibly be invaded and colonized by bacteria.

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