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Dentin hydraulic conductance with different application times of diamine silver fluoride/potassium iodide desensitizing solution

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Abstract

The aim was to evaluate the influence of time of application of diamine silver fluoride on the hydraulic conductance.

Methods: The sample of this prospective experimental study included 60 healthy human third molars with indications of extraction in malocclusion. These were extracted from patients between 18 and 30 years of age. 4 groups of 15 dentin disks were obtained: GC (no treatment), G1 (were actively treated with diamine silver fluoride for 15 s), G2 (30 s), G3 (60 s). Then, was measured hydraulic conductance of dentin. The rate of flow of the fluid through the dentin disks was measured by recording the initial position of a bubble of air inside a capillary in 20 min. With the Wilcoxon signed rank test, it was established that there is a statistically significant difference in hydraulic conductance between the control group and the samples treated with desensitizing solution based on diamine silver fluoride/potassium iodide. There was no statistically significant difference between the groups treated with different application times of the desensitizing solution. There was no difference in hydraulic conductance between disks with different application times of desensitizing solutions of diamine silver fluoride/potassium iodide.

Introduction

Dentin permeability is defined by the passage of various substances such as fluids, molecules, and bacteria through the dentin. The characteristic of being permeable is fundamental and enables the exchange of nutrients to and from the pulp through the odontoblastic processes in the dentin tubules. The permeability is also related to the main theory of the emergence of sensitivity.^[1]

Dental hypersensitivity is a painful condition that affects a large part of the adult population and is associated with dentin exposure to the oral environment. It can be initiated by tactile, chemical, thermal, or osmotic stimuli, and it can cause varying degrees of pain. The etiology may be due to the loss of substance of the teeth via abfraction, erosion, or abrasion generated by

aggressive oral hygiene, eating acid, exposure to cold stimuli, or periodontal disease causing the denudation of the root surface.^[2] Pashley *et al.* assessed the rate of fluid flow through dentin disks using a diffusion camera, which was modified and validated by Hevia *et al.*^[3] It operates through a diffusion chamber positioned at a disk of dentin held between two rubber rings. A micropipette quantifies the movement of fluid in the system through the entry of a bubble of air that moves due to the permeability of the disk. The solution reservoir is located at a height to produce a pressure equivalent to 20 cm of H₂O to simulate pulp pressure.^[4]

Dentin sensitivity can be reduced through the formation of intratubular crystals from minerals from saliva or dentinal fluid, peritubular dentin formation, reparative dentin formation, and invasion of the tubules by bacteria or plasma proteins. The reduction of permeability through tubular occlusion is a method

that is used by many of the existing desensitizing agents today for the treatment of sensitivity. The treatment is aimed at preventing the flow of dentinal fluid and the reduction of pain. This tubular occlusion can be generated by the deposit of salts or ions within the dentinal tubule and by precipitation of proteins or dentin sealant.^[2]

Desensitizing solutions based on diamine silver fluoride with a high concentration of fluoride generate an occlusion of dentinal tubules through the formation of a film of variable thickness of silver phosphate and calcium fluoride.^[5] This method has the disadvantage of producing a black pigmentation in remineralized parts. A new alternative solution to this problem is to use a second treatment with potassium iodide (KI), which produces a creamy white color from silver iodide reaction products, thereby reducing the formation of black precipitates.

The required application time for the product to be effective is not clear. This study uses an *in-vitro* model to compare the hydraulic transdental conductance of disks of human dentin treated with different application times of desensitizer based on diamine silver fluoride/potassium iodide. This was a modification of the experimental model by Pashley *et al.* used for the measurement of flow in a chamber of diffusion. The aim of this work was to evaluate the influence of time of application of diamine silver fluoride on the hydraulic conductance.

Materials and Methods

The sample of this prospective experimental study included 60 healthy human third molars with indications of extraction in malocclusion. These were extracted from patients between 18 and 30 years of age. Informed consent was obtained, and the procedure was approved by the Ethical Committee of the Faculty of Dentistry of the University of Chile (PRI-ODO/12-002).

The teeth were disinfected for 24 h in a solution of 0.1% Thymol (SIGMA, England). Then, they were manually cleaned to remove all traces of periodontal ligament using curettes (Gracey 13-14 HuFriedy, USA). The teeth were then preserved in 0.9% saline at room temperature. The teeth were etched with 35% orthophosphoric acid (Coltene Whaledent) over all the enamel for 30 s. Then, they were washed for 60 s and dried to proceed with the application of a layer of cyanoacrylate. We then made cylindrical silicone molds, where the teeth were suspended in blocks of epoxy resin. These were left to spend 48 h to complete polymerization. Etching with enamel acid was carried out to enable micromechanical adhesion of the enamel to the cyanoacrylate and to enhance bonding with the epoxy resin.

Disks of dentin that were 1 mm ± 0.2 mm thick were cut in a cutting machine at 750 rpm (ISOMET BUEHLER LER LAKEBUFF IL, USA 1000) under the intermittent load of 500 g and refrigeration. The cut disks were numbered and randomized for distribution in the study groups. Subsequently, the occlusal face of each disk of dentin was regularized with abrasive paper (silicone Paper N 600, SIA, Switzerland) under circulating water to standardize the thickness, regularize the surface, and

standardize the smear layer. Finally, the disks were washed with an air/water spray for 30 s.

The human dentin disks were treated on their occlusal face with desensitization based on diamine silver fluoride/potassium iodide (riva star Step 1: Silver fluoride 30-35%, riva star Step 2: Potassium iodide, SDI Limited). The following instructions from the manufacturer were followed for the application of the desensitizing solution.

1. The immediate implementation of the solution of potassium iodide after applying the diamine silver fluoride is essential. In addition to helping with the process of desensitization, potassium iodide also helps to counteract the negative effects of the diamine silver fluoride. Used alone, the solution of diamine silver fluoride will stain the tooth.
2. Keep the product between 2 and 8 C.
3. The ingredients are only active 5 min after opening the capsule. After this time, its content should not be reused.
4. Groups of 15 dentin disks were obtained and divided in the following way.
 - Control group: 15 dentin disks were etched with 35% orthophosphoric acid for 15 s and washed later with abundant water.
 - Group 1: 15 dentin disks previously etched with 35% orthophosphoric acid were actively treated with diamine silver fluoride for 15 s using a microbrush, followed by the passive application of potassium iodide for 5 s. Finally, the disks were washed with plenty of water.
 - Group 2: 15 dentin disks previously etched with 35% orthophosphoric acid were actively treated with diamine silver fluoride for 30 s with a microbrush followed by passive application of potassium iodide for 5 s. Finally, the disks were washed with plenty of water.
 - Group 3: 15 dentin disks previously etched with 35% orthophosphoric acid were actively treated with diamine silver fluoride for 60 s with a microbrush followed by passive application of potassium iodide for 5 s. Finally, the disks were washed with plenty of water.

In addition, two control groups were added to verify the operation of the device before placing the samples in the diffusion chamber.

- Positive control: Measurement of diskless dentin
- Negative control: Disk of resin epoxy.

The experimental model used was developed by Pashley *et al.* and modified and validated by Hevia *et al.*^[3] The model is simple, inexpensive, and easy to mount, and it allows for measuring the hydraulic conductance of dentin. The rate of flow of the fluid through the dentin disks was measured by recording the initial position of a bubble of air inside a capillary. After 20 min, it enters its final position, and the rate is measured in µl/min by visual observation. The hydraulic conductance (CH) corresponds to a formula that determines the permeability of the disks of dentin. The variable F corresponds to the flow rate of each experimental group. The variable A corresponds to the area of dentin exposed to the fluid. The variable P corresponds to the intrapulpal pressure, whose value corresponds to the height of the column

of distilled water (20 cm). The variable T is the time in minutes. The results for the hydraulic conductance are calculated using the following formula.

$$CH = \frac{F}{A \cdot P \cdot t}$$

The measurements of the exposed dentin from the disk area were calculated using Image J software, which is widely used in medicine, using a previous photographic record obtained with a Nikon p90 camera at a distance of 30 cm.^[6]

Statistical analysis

Through statistical analysis using SPSS 21.0 for Windows, we found the normality of the data distribution through the Shapiro-Wilk test. The Wilcoxon signed ranks were used to compare the average of two related samples and determine if there were differences between them. The sample size was 15 (n). The established confidence interval was 95%. A beta value of 0.2 was considered for error.

Results

Table 1 presents the values of the dentin for each disk area, which were obtained using Image J. The numbering of disks from Groups 1-3 has a random distribution due to previous randomization. Table 2 presents the values of calculated hydraulic conductance obtained from the area of dentin and the flow rate of each disk.

The graph in Figure 1 shows a comparison of the values of hydraulic conductance of each group. The vertical axis indicates conductance values. The axis x indicates the allocation of each group: Control Group (C), Groups (1-3) with the application of diamine silver fluoride for 15, 30, and 60 s, respectively.

With the Wilcoxon signed rank test [Table 3], it was established that there is a statistically significant difference in hydraulic conductance between the control group and the samples treated with desensitizing solution based on diamine silver fluoride/potassium iodide. There was no statistically significant difference between the groups treated with different application times of the desensitizing solution.

Discussion

Dental hypersensitivity, as any other feeling of pain in the body, is a major health problem that affects the quality of life of patients, which leads to a greater concern for their state of health. Despite investigations of various substances and materials, much is still unknown about the hypersensitivity of dentin, and even the terminology can be questioned. With this in mind, dental professionals need to develop new strategies for the management of dental hypersensitivity to improve the quality of life of patients.^[7]

Desensitizing solution based on diamine silver fluoride with a high concentration of fluoride began to be used in 1972 to treat caries and active enamel lesions. Its properties have already been

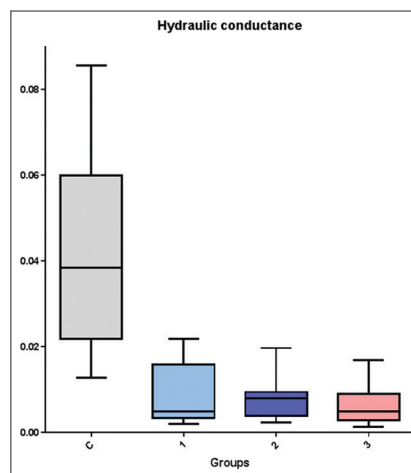


Figure 1: Hydraulic conductance of control group and experimental groups in $\mu\text{l}/\text{min} \cdot \text{cm}^2$

Table 1: Averages and SD of dentin disk areas of each group, expressed in cm^2

	Control	G1	G2	G3
Mean	0.537	0.598	0.573	0.534
SD	0.139	0.105	0.113	0.116

SD: Standard deviation

Table 2: Mean and SD values of hydraulic conductance (CH) group expressed in $\mu\text{l}/\text{min} \cdot \text{cm}^2$

	Control	G1	G2	G3
Mean	0.0433	0.0087	0.0082	0.0061
SD	0.0210	0.0063	0.0050	0.0040

SD: Standard deviation

Table 3: The range test with Wilcoxon sign for variable hydraulic conductance

Groups	Z	Asymp significant
Control-1	-3.408	0.001
Control-2	-3.408	0.001
Control-3	-3.408	0.001
1-2	-3.41	0.733
1-3	-1.193	0.233
2-3	-1.079	0.281

proven previously, and currently, its use is constantly growing. An *in-vitro* study by Quock *et al.* evaluated the effect of the treatment of dentin on caries with diamine silver fluoride at concentrations of 12% and 38%. The treatments affected the microtensile bond strength of the composite resin bond to dentin. The effect of the adhesive type used was also evaluated by comparing the subsequent rinse versus a self-etching adhesive technique. It was concluded that none of the experimental groups treated with diamine silver fluoride significantly reduced

the resistance of the microtensile bond of dentin in comparison to a control group without treatment. However, the application of 12% diamine silver fluoride after applying a self-etching technique significantly reduced the strength of tooth bond restoration compared to the subsequent rinse technique. With this result, it can be concluded that it is preferable to use an adhesive with an etching technique and rinsing instead of one self-etch prior to treatment with diamine silver fluoride.^[8]

Regarding effects on the resistance of tooth bond restoration, Knight *et al.* conducted an *in-vitro* study in order to compare the strength of adhesion of glass ionomer cement on dentin surfaces. Tooth surfaces were treated with diamine silver fluoride/iodide of potassium and compared to untreated dentin. This study found that it is preferable to wash the reaction products generated and subsequently dry them with air versus leaving the precipitate formed. Adhesion values similar to those of non-treated dentin were generated after such treatment.^[9,10]

Ariffin *et al.* studied the release of fluoride by glass ionomer cement after coating with silver fluoride. They concluded that the application of a layer of 10% silver fluoride improves the concentration of fluoride released from this material. This finding could be applied to improve protection against recurrent caries, especially in patients with high risk of caries, as well as for treatment of non-carious cervical lesions sensitivity with the loss of tooth structure or for teeth with post-operative sensitivity.^[11]

In 1972, Shimooka^[12] observed the penetration of diamine silver fluoride on the microstructure of healthy dentin and enamel. It was shown that the diamine silver fluoride has high penetration ability of 20 μ in healthy human enamel. Llodra mentioned that this compound penetrates the dentin by 50-100 μ , noting in addition that silver ions penetrate more deeply, coming close to the pulp chamber.^[13]

It is also important to consider the adverse effects of this compound. A study by Gotjamanos evaluated the response of primary teeth pulp with deep caries treated with 40% diamine silver fluoride followed by restoration with glass ionomer cement by applying an atraumatic restorative technique. 90% of the examined teeth showed a pulp response favorably inducing abundant formation of reparative dentin.^[14]

Nakamura *et al.* studied the cytotoxic impact of silver and diamine silver fluoride in six types of oral cells (human pulp cells, human periodontal ligament fibroblasts, human gingival fibroblasts, and human oral squamous carcinoma cells). The results showed a high cytotoxicity of diamine silver fluoride on oral cells, so the clinical application of this solution must be handled with special attention given to the tissues. When using the product, there should be caution in clinical handling to avoid contact with the gingival margin. Application may be contraindicated in deep cavities near the pulp tissue. In the future, it will be necessary to study the toxicity in greater depth to assess biocompatibility.^[15]

There have been no studies indicating the time during which the diamine silver fluoride/potassium iodide should be applied to make its action effective. The results in this study showed that

compounds formed by the desensitizing solution managed to decrease the permeability of dentin independently of the time applied. However, there was a statistically significant difference between the control group with no desensitizing solution treatment and the experimental groups to which desensitizing solution was applied.

Between experimental groups, the application of 15, 30, and 60 s of treatment was not relevant to measuring the hydraulic conductance of disks measured immediately after application. This can be explained by the 15 s of application generating saturation with salts of silver that precipitated on the dentin. This generated an occlusion of dentinal tubules. With these results, we conclude that an application for 15 s is effective for intratubular occlusion, which saves time. These results are consistent with the null hypothesis of the research that different application times would produce no statistically significant difference. However, it should be considered that the *in-vitro* results cannot be extrapolated to the possible outcomes *in-vivo*, since conditions in the oral environment are different from those in this experimental model.^[16]

In a recent clinical study, Craig *et al.* used a visual analogue scale (VAS) to compare the effectiveness of diamine silver fluoride/potassium iodide with a preparation based on oxalic acid to reduce dentin hypersensitivity. Measurements were performed 7 days after the application of the desensitizing agent, and it was concluded that the relief obtained by patients treated with diamine silver fluoride/potassium iodide was higher. Another clinical study by Castillo *et al.* (2010) used VAS to evaluate a population with teeth sensitive to cold air. Two 1-min applications of diamine silver fluoride were performed. As a result, there was a significant reduction of pain in response to cold air in comparison to a placebo. This was measured at 24 h and 7 days after application. It can be concluded that the diamine silver fluoride/potassium iodide has potential as a treatment for dentin hypersensitivity after immediate application. However, other studies are necessary to measure the maintenance of the effect over a longer time period and for comparison with other occlusive desensitizing agents.^[16]

It is known that diamine silver fluoride produces a black pigmentation, whether in carious or non-carious dentin. This produces an esthetic challenge for the material. This study used a formulation of diamine silver fluoride at various concentrations from 30% to 35% according to the manufacturer's specifications. After application, the use of potassium iodide is indicated to mask the staining produced by the diamine silver fluoride through the generation of a white precipitate of silver iodide. However, the effect of this treatment regarding effective stain reduction and the interaction with different restoration materials has not yet been determined. Because of this, we intend to carry out further studies to evaluate these variables.^[17]

The model with dentin disks has been widely used to assess intra-tubular occlusion and the effects on fluid flow through the dentin by desensitizing agents.^[18,19] However, there are some drawbacks. To measure the progress of air bubbles through a pipette, the observer can make mistakes in measuring depending on their visual ability. This makes the method more susceptible

to errors by the observer, who can unconsciously skew the readings. For example, the reading of the position of the air bubble located between two continuous calibration marks can be assigned either to the highest mark or to the lowest, depending on the researcher and their criteria. On the other hand, by using a millimeter pipette whose scale is 0.001 ml, smaller measurements are not obtained, so minor variations can be undetected, this is considered as a limitation of this trial. The conclusion is there was no difference in hydraulic conductance between disks with different application times of desensitizing solutions of diamine silver fluoride/potassium iodide.

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