Effect of Gamma Radiation on Dentin Bond Strength and Morphology

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Sterilization by gamma radiation is a method often used for bone and extracted teeth banking. The bond strength of human dentin submitted to gamma rays has not been reported. Therefore, the aim of this study was to assess the effect of gamma radiation on dentin shear bond strength and morphology. The roots were removed from extracted human bicuspids and their crowns divided into two groups: an untreated control and crowns submitted to gamma radiation sterilization. The crowns were mounted in epoxy resin and the buccal enamel removed exposing the subjacent dentin. SBMPP plus adhesive system was applied to a 3-mm diameter area after 15 s of 35% phosphoric acid etching. The samples were mounted in composite resin cylinders and stored in distilled water at 37°C for 24 h until the shear test. Dental fragments from both groups were prepared for SEM analysis. There was no statistically significant difference between the results of the shear test for the two groups according to the Tukey test (p>0.05). Scanning electron micrographs also did not show alterations. These results indicate that gamma radiation neither affected the shear bond strength of SBMPP plus nor altered the dentin surface morphology.

Key Words: sterilization, cross contamination, gamma radiation, bond strength.

INTRODUCTION

Investigations in vitro on dental materials and some restorative techniques in vivo (1,2) require a significant number of extracted human teeth. However, extracted teeth may harbor microorganisms causing cross-contamination. Thus, infection control procedures should be considered before manipulating extracted human teeth to eliminate the risk of disease transmission (3).

Several methodologies such as steam and dry autoclaving, ethylene oxide gas and gamma radiation have been used to achieve germ-free dental specimens (Table 1). Disinfectant solutions such as 2% glutaraldehyde, 10% formalin and sodium hypochlorite have also been used to reduce the number of microorganisms (11-13). DeWald (12) reported no significant alterations on bond strength when 10% formalin was used as a storage media.

Dentin bond strength is usually determined by shear and tensile tests but the results in human teeth submitted to different sterilization methods are uncertain, as shown in Table 1. These variations may be related to non-standardized time periods and storage solutions of teeth before their use (12,14).

Although most of these sterilization methods have been relatively well reported, the effects of gamma rays on dentin morphology and bond strength have not been studied (12). White et al. (10) concluded that gamma radiation is effective for elimination of microbes from dental specimens. Furthermore, they also showed that this sterilization method had no effect on dentin permeability. Dentin structural changes as measured by Fourier transform infrared spectroscopy or ultra-violet-visible-near-infrared spectra were also not detected. On the other hand, Cheung et al. (15) reported that collagen molecules in tissue are readily altered by gamma radiation.

The aim of this study was to assess the effects of
gamma radiation on both dentin bond strength and its surface morphology. Because the influence of sterilization was the main object of this investigation, rather than the material properties, a conventional two-bottle adhesive system was chosen.

**MATERIAL AND METHODS**

**Shear Bond Strength Test**

Forty extracted human premolars stored in 10% buffered formalin for seven days had their roots removed at the cementoenamel junction using a low-speed diamond disc under water cooling. The teeth were equally divided into two groups of 20 teeth each: 1) control group – teeth were kept in saline solution and 2) gamma radiation group – each tooth was adequately packed and then submitted to sterilization at 25 KGy for 6 h.

The crown segments were individually mounted in epoxy resin cylinders. Using 120-grit sandpaper, under running tap water, the buccal enamel was removed to obtain a dentin surface for bonding. Scotch tape with a 3-mm diameter perforation coated the exposed dentin surface and the circular area obtained was etched for 15 s with 35% phosphoric acid (Batch No. 7545BS, Scotchbond, Dental Products Division, 3M, St. Paul, MN, USA). Primer and bonding agent (SBMPPlus, Batch No. 7545BS, Scotchbond, Dental Products Division, 3M) were applied to the dentin surface according to the manufacturer’s instructions. By means of a teflon matrix, a 3-mm diameter composite resin cylinder (Batch No.8004B3, Z-100, Dental Products Division, 3M) was built in three layers and light-cured for 40 s each. The prepared specimens were stored in distilled water at 37°C for 24 h before the shear test (KRATOS testing machine). Shear force was applied at a constant rate of 0.5 mm/min (Figure 1). The bond strength (Mpa) was calculated and the data were analyzed statistically with Tukey’s test (p<0.05).

**Scanning Electron Microscopy (SEM)**

For scanning electron microscopy, the crowns of 4 molars were cross-sectioned at the pulp chamber plane perpendicular to the longitudinal tooth axis using a low-speed diamond disc under water cooling. The second cut was parallel to the first and exposed the occlusal surface of the dentin. Each tooth slice was cross-cut into 4 fragments: two for the control group (kept in saline solution) and two for the gamma radiation (Figure 2). After sterilization the dentin sections were demineralized with 10% citric acid plus 3% ferric acid for 6 h, followed by dehydration in graded alcohol solutions and critical point drying in liquid CO2. The sections were mounted on aluminum stubs coated with a thin carbon film and sputter-coated with gold. They were then analyzed with a Hitachi S-3000N scanning electron microscope at an accelerating voltage of 20 kV (Figure 2).

<table>
<thead>
<tr>
<th>Method</th>
<th>Test</th>
<th>Results</th>
<th>Reference</th>
</tr>
</thead>
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<td>Autoclave</td>
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</tr>
<tr>
<td></td>
<td></td>
<td>No effect</td>
<td>5</td>
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<tr>
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<td>No effect</td>
<td>7</td>
</tr>
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<td>Effectiveness as a</td>
<td>Positive</td>
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<td>oxide</td>
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Table 1. Summary of the sterilization methods for extracted teeth.
chloride in water for 30 s. For dehydration, the dental fragments were immersed in an increasing concentration of ethanol (25% to 100%). They were subsequently immersed in 100% acetone, 50% acetone plus 50% HMDS (hexamethyldisilazane), 100% HMDS and placed in a desiccator for 24 h to obtain complete drying (16). The sterilized dentin discs and untreated controls were coated with gold and the dentin was examined by scanning electron microscopy.

RESULTS

Shear Bond Strength Test

Mean shear bond strengths (MPa; mean ± SD) following gamma radiation sterilization was 9.67 ± 5.11 (range 6.75-24.22) compared to control values (10.03 ± 2.44; range 3.34-18.05). Analysis of variance followed by Tukey’s test indicated no significant differences between the two groups (p>0.05).

Scanning Electron Microscopy (SEM)

The dentin sterilized by gamma radiation showed a microscopic pattern, color and shape similar to the control dentin. In the dentin next to the pulp chamber, the scanning electron micrographs showed organic matrix as a wide collagen fiber network surrounding the dentinal tubule openings in both control and gamma radiation groups (Figures 3 and 4). The alterations detected in the teeth submitted to gamma radiation probably consisted of cellular remnants due to the proximity to the pulp chamber.

DISCUSSION

Gamma rays, similar to X-rays, comprise ionizing radiation used to achieve sterilization of bone for banking (17) and dental enamel used in cariogenicity tests in vitro (18). It has been reported that sterilization by gamma radiation is an effective method to prevent the transmission of various strains of bacteria, fungi and viruses, including HIV (17). In this study a maximum radiation dosage of 25 KGy for 6 hours, which is enough to sterilize bone according to Salai et al (17), was used.

Gamma radiation at dosages commonly used to sterilize biomedical products is harmful to collagen molecules in tissue (15). Therefore, modification in the collagen fibres with consequent disorganization or collapse of the collagen network could be expected.

Figure 3. Scanning electron micrograph of the control dentin. The arrows indicate the collagen fiber network of the peritubular and intertubular dentin showing its normal organization surrounding the tubule openings (5,000X).

Figure 4. Scanning electron micrograph of the dentin sterilized by gamma radiation. The collagen fiber network organization was preserved although some isolated cells remained (arrows) on the surface (5,000X).
However, the methods used in this study were not suitable to clarify if those changes really occur. SEM analysis of the dentin surface showed a normal pattern characterized by an organized collagen network with fibers surrounding the tubule openings making up peritubular and intertubular dentin. Shear bond strength was not affected by gamma radiation, but further investigation is required to establish comparisons and agreements.

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RESUMO


A radiação gama é um método de esterilização geralmente utilizado em banco de ossos e de dentes humanos extraídos. A força de adesão de resina composta à dentina de dentes humanos submetidos aos raios gama não tem sido investigada na literatura. O objetivo deste trabalho foi avaliar o efeito da radiação gama na força de adesão de resina composta à dentina e na sua morfologia. Pré-molares humanos íntegros extraídos, após remoção de suas raízes, foram divididos em dois grupos: grupo controle e coroas dentárias submetidas à esterilização pela radiação gama. As coroas dentárias foram incluídas em resina epóxica e o esmalte da face vestibular removido expondo a dentina subjacente. O sistema adesivo SBMPPlus foi aplicado sobre uma área de 3 mm de diâmetro após 15 segundos de condicionamento com ácido fosfórico 37%. Um cilindro de resina composta foi obtido sendo os espécimes armazenados em água destilada a 37°C por 24 horas até o teste de cisalhamento. Fragmentos dentários de ambos os grupos foram preparados para análise em microscopia eletrônica de varredura. Os resultados do teste de cisalhamento demonstraram que não houve diferença estatisticamente significativa entre os dois grupos de acordo com o teste de Tukey (p>0.05). Nenhuma alteração marcante foi detectada na morfologia dentinária avaliada ao microscópio eletrônico de varredura. Os resultados indicaram que a radiação gama não afetou a força de adesão do SBMPPlus à dentina e também não alterou a morfologia dentinária.

Unitermos: esterilização, contaminação cruzada, radiação gama, força de adesão.

REFERENCES


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