Pomegranate (Punica granatum L.) gel extract as an antioxidant on the shear bond strength of a resin composite post-bleaching application with 40% hydrogen peroxide

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ABSTRACT

Background: Tooth discoloration can be treated with tooth bleaching. Bleaching using 40% hydrogen peroxide can reduce the shear bond strength of resin composite because there are free radicals on the tooth surface, so it can delay the restoration. The application of antioxidants can eliminate free radicals after the bleaching procedure and increase the shear bond strength of the composite resin. The common antioxidants are ascorbic acid and natural ingredients, such as pomegranate (Punica granatum L.). Purpose: To determine the effect of pomegranate extract gel on the shear bond strength of composite resin after 40% hydrogen peroxide bleaching application. Methods: This research used 32 maxillary first premolars that were divided into four groups. The samples were bleached, then the labial was prepared and antioxidant gel was applied: group P1 pomegranate gel extract of 5%, group P2 pomegranate gel extract of 10%, group K1 positive control ascorbic acid gel of 10% and group K2 as the negative control. The samples were restored with a nanohybrid composite resin. The shear bond strength was tested using a universal testing machine. The data were tested using a one-way ANOVA followed by a post-hoc LSD test. Results: The pomegranate gel extract increased the shear bond strength of the composite resin after the bleaching procedure of 40% hydrogen peroxide compared with the ascorbic acid gel group and the negative control group. The one-way ANOVA test showed a significant difference (p<0.05). The post-hoc LSD test showed significant differences between the treatment and negative control groups (p<0.05). Conclusion: The pomegranate gel extract as an antioxidant increased the shear bond strength of the composite resin restoration after the 40% hydrogen peroxide bleaching application.

Keywords: antioxidant; bleaching; pomegranate extract; shear bond strength

INTRODUCTION

Tooth discoloration is a problem that can occur in teeth due to intrinsic or extrinsic factors. Intrinsic tooth discoloration is of internal origin, such as tooth necrosis and drug consumption. Meanwhile, extrinsic discoloration is a colour change that is influenced by external factors, such as the consumption of tea or coffee, cigarette stains and mouthwash that leaves colouring on the tooth surface.1

The treatment for tooth discoloration is tooth bleaching. Bleaching is widely chosen because it is easy, fast and does not cause a reduction of the hard tissue in teeth.2 Tooth bleaching is classified into internal and external. External bleaching is divided into in-office and home-bleaching. Internal bleaching or walking bleach is a procedure performed on non-vital teeth.3 The bleaching materials commonly used in in-office bleaching procedures are 30-50% hydrogen peroxide or 35–37% carbamide peroxide.4

Using bleaching agents can have adverse effects, one of which is the reduction of the shear bond strength of the tooth surface with composite resin restorations. The bond strength of composite resin is very important for increasing the quality and durability of the restoration, so any reduction would be undesirable.5 The bond strength is influenced by various factors, such as the cause of discoloration, type of bleaching material, curing conditions and storage. The use of antioxidants can improve the bond strength, but the effectiveness of each type of antioxidant is limited.6

The common antioxidants used in dental research are ascorbic acid and natural antioxidants, such as pomegranate. Pomegranate (Punica granatum) is a common fruit grown in several countries, such as India, Turkey and China. Pomegranate juice and seed extracts have been investigated for their antioxidant and anti-inflammatory properties.7

In the present study, the effect of pomegranate extract gel on the shear bond strength of composite resin after 40% hydrogen peroxide bleaching application was investigated.
if it given pressure. The bond strength of composite resin can be seen from the shear bond strength of the attachment, which is the maximum resistance of a material to withstand loads that cause shear movements before the composite resin material is released. Several methods have been suggested to increase the bond strength after the bleaching procedure, including delaying the restoration procedure for 24 hours to three weeks.

The delay of restoration procedures can be accelerated by eliminating free-radical peroxides, which are free radicals formed by residual peroxide from bleaching materials that were left on the tooth surface. A waiting period may not always be possible for many reasons, such as lack of time, personal circumstances or aesthetic reasons. Many techniques have therefore been proposed to decrease the bond strength that is caused by the bleaching enamel, such as suggesting the use of adhesives containing organic solvents. The use of an antioxidant has been proven in many studies to be safe and effective to increase the bond strength directly after bleaching, with no significant difference with unbleached teeth. Natural antioxidants have benefits in the field of dentistry; for example, they can increase the bonding strength of the composite resin fill, which decreases after the bleaching procedure. Sodium bicarbonate, rosemary extract and aloe vera are already used to remove free radicals of hydrogen peroxide after whitening. Some vitamins, like α-tocopherol (an active component of the vitamin E complex) and sodium ascorbate (SA) or ascorbic acid (vitamin C), are known as neutral, biocompatible and potent antioxidants that have the ability to reduce various oxidative compounds.

The antioxidant material that is often used to eliminate free radicals after the bleaching procedure is 10% of ascorbic acid before the restoration procedure. This compound has high antioxidant potential, is biocompatible and has low toxicity. One of the fruits that is used as an antioxidant is pomegranate (Punica granatum L.) because it shows a homogeneous structure; there are no visible or coarse grains. The results of the gel adhesion test made and that it still met the positive criteria parameters. The homogeneity test results show that the gel is homogeneous in each group showed that all the groups met the adhesion requirements, namely having an adhesion time of more than one second. The longer the stickiness the longer the active substance is attached so that it will have a more optimal effect. The spreadability test was carried out to determine the gel’s ability to spread when applied. The dispersibility

MATERIALS AND METHODS

This research received the approval from The Health Research Ethics Commission of Faculty of Medicine, Universitas Jenderal Soedirman (Number 201/KEPK/IX/2020). The type of research was true experimental laboratory using a simple randomised sampling technique using a posttest-only control group design. The materials used were pomegranate, 40% of hydrogen peroxide and a universal testing machine. The samples used were 32 maxillary first premolar teeth. The first premolar teeth were chosen because they represent the anterior teeth. The samples were divided into four groups, namely P1 (treatment group 1, the sample was applied with a concentration of 5% pomegranate extract gel after the application of 40% hydrogen peroxide), P2 (treatment group 2, the sample was a 10% concentration of pomegranate extract gel, which was applied after the application of 40% hydrogen peroxide), K1 (positive control group, the sample was applied 10% ascorbic acid extract gel after 40% hydrogen peroxide application) and K2 (negative control group, the sample was not given any antioxidant after the application of 40% hydrogen peroxide). The ascorbic acid was used because it has been used in previous journals as a positive control.

The data used in this study are primary data in the form of material or a collection of facts that were collected by the researcher during the research. The methods of data collection in this study can be divided into getting ethical clearance, managing the permits, determining the pomegranate growth, the extracting and gelling of the pomegranate extract, making samples, treating the sample groups and testing the shear bond strength using the Pearson Panke universal testing machine (UTM) (Pearson Panke, Ltd. London, UK).

The extract gel was made by macerating 1500 grams of pomegranate, which was mashed and soaked in six litres of a 96% ethanol solvent for two x 24 hours with stirring every 24 hours. Stirring aims to accelerate the contact between the sample and the solvent. The solvent will penetrate the cell wall and enter the cell cavity so that the active substance dissolves. The immersion results were evaporated with a rotary evaporator at a temperature of 78 ± 1°C at a speed of 55 rpm. The gel was made by mixing pomegranate extract and natrium carboxymethyl cellulose (Na-CMC), which were the ingredients of the gel base.

The gel had to be physically evaluated through a homogeneity test, an adhesiveness test and a spreadability test before the application to ensure that the gel preparations had the same physical appearance after the preparation was made and that it still met the positive criteria parameters during storage. The criteria of a good gel are when the homogeneity test results show that the gel is homogeneous because it shows a homogeneous structure; there are no visible or coarse grains. The results of the gel adhesion test in each group showed that all the groups met the adhesion requirements, namely having an adhesion time of more than one second. The longer the stickiness the longer the active substance is attached so that it will have a more optimal effect. The spreadability test was carried out to determine the gel’s ability to spread when applied. The dispersibility
requirements for the topical preparations were 5–7 cm and the three groups of preparations met the dispersibility requirements for the topical preparations. The preparations that are difficult to spread will reduce the comfort level of use and the effectiveness of using the preparation, while preparations that are too diluted will cause their adhesion to decrease so that the contact time of the active substance with the target is reduced.15

A sample preparation was done by making a resin block mould measuring 1.5 x 1.5 x 1 cm. The mould was used to fix the crown of the first maxillary premolar, which had been prepared in a labial position facing upwards. The resin block mould is shown in Figure 1.

The samples were treated by applying an Opalescence Boost PF 40% hydrogen peroxide gel (Ultradent Product Inc., USA) to the labial surface of the teeth by as much as 1 mg for two x 20 minutes. The next sample was prepared according to ISO / TS 11405: 2003 using silicon carbide abrasive paper size P600 under running water on the labial surface of not more than 1 mm, while ensuring that all the surfaces were level and flat. After that, each sample group was subjected to treatment, namely groups P1 (sample applied pomegranate extract gel with a concentration of 5% 1 mg for 10 minutes), P2 (sample applied pomegranate extract gel with 10% concentration of 1 mg for 10 minutes), K1 (the sample was applied 1 mg of 10% ascorbic acid extract gel for 10 minutes) and K2 (negative control group, antioxidants were not applied to the sample after the 40% hydrogen peroxide application). All the sample groups were then subjected to restoration procedures using a nanohybrid composite resin to form the block moulds measuring 2 x 2 x 2 mm. They were then subjected to light curing for 20 seconds. The entire sample group was immersed in distilled water for 24 hours at room temperature.

The sample test was conducted by the shear bond strength test with the UTM. A total of 32 tooth samples were subjected to a shear test to determine the value of the composite resin adhesion. The samples that had been implanted in acrylic were mapped on Pearson Panke equipment with a speed of 2.28 seconds/milli 16 and then the shear bond strength was tested with the lowest strength. The resulting (F) in newton (N) units is the shear strength inputted with the shear bonding strength formula \( T = F/A \), where \( T \) = shear adhesion strength (MPa), \( F \) = shear force (N) and \( A \) = cross-sectional area (mm²).17

The data analysis was performed using IBM’s SPSS Statistics version 23 (Armonk, New York). The resulting data were stated to be normally distributed (p > 0.05) and homogeneous after the data transformation was conducted. A parametric test using one-way ANOVA with a confidence level of 95% (p = 0.05) was carried out to determine the significant differences in each group. The research data showed that there were significant differences (p < 0.5). The data were analysed using the least significant difference (LSD) test to establish any significant differences.

RESULTS

The results of the determination test show that pomegranate comes from Punica genus, the *Punica granatum* L. species. The pomegranate extraction resulted in a thick extract of 300 g. The pomegranate extract and the ascorbic acid were then made into a gel based on Na-CMC, which were evaluated to determine its physical condition. The evaluation of the gel preparations was carried out before the application to ensure that they had the same physical appearance after the preparation was made and that they still met the good criteria parameters during storage. The evaluation results of F1 (pomegranate extract gel 5%), F2 (pomegranate extract gel 10%) and F3 (ascorbic acid gel 10%) showed homogeneous results. The results of the adhesiveness evaluation of the formulas were 20, 32 and 25 seconds. The results of the dispersibility evaluation of the formulas were in the range of 5–7 cm.

The shear bond strength of the composite resin that was tested using the UTM had mean values, which can be seen in Table 1. The results showed that the mean value in the P2 group (treated with 10% pomegranate extract) had the highest value, namely 42.5 Mpa. The result of the one-way ANOVA parametric test demonstrated a significant difference of p 0.000 (p <0.05).

![Figure 1. Resin block mould.](image)

### Table 1. One-way ANOVA test result: shear bond strength

<table>
<thead>
<tr>
<th>Group</th>
<th>Mean</th>
<th>Standard Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>P1</td>
<td>11.56 Mpa</td>
<td>3.5197</td>
</tr>
<tr>
<td>P2</td>
<td>42.50 Mpa</td>
<td>19.7303</td>
</tr>
<tr>
<td>K1</td>
<td>15.31 Mpa</td>
<td>3.6443</td>
</tr>
<tr>
<td>K2</td>
<td>4.50 Mpa</td>
<td>2.9881</td>
</tr>
</tbody>
</table>

Note: P1: Gel treatment/ pomegranate extract 5%; P2: 10% pomegranate extract gel treatment; K1: positive control 10% ascorbic acid gel; K2: negative control.

### Table 2. Post-hoc LSD test result: shear bond strength

<table>
<thead>
<tr>
<th>Group</th>
<th>P1</th>
<th>P2</th>
<th>K1</th>
<th>K2</th>
</tr>
</thead>
<tbody>
<tr>
<td>P1</td>
<td>**</td>
<td>0.000</td>
<td>0.154</td>
<td>0.002</td>
</tr>
<tr>
<td>P2</td>
<td></td>
<td>**</td>
<td>0.000</td>
<td>**</td>
</tr>
<tr>
<td>K1</td>
<td></td>
<td></td>
<td>**</td>
<td>**</td>
</tr>
<tr>
<td>K2</td>
<td></td>
<td></td>
<td></td>
<td>**</td>
</tr>
</tbody>
</table>

** = There is a significant difference (p <0.01) between the groups. Source: Primary data, 2020.
DISCUSSION

The application of the 5% pomegranate extract gel, the 10% pomegranate extract gel and the 10% ascorbic acid gel was proven to increase the shear bond strength of the composite resin restoration. These results indicate that the gel is capable of being an antioxidant agent that can remove free radicals that are left on the tooth surface after the bleaching procedure using 40% of hydrogen peroxide.

The 10% pomegranate extract gel treatment group (P2) had higher strength than all the groups, and there was a significant difference, which means that it had better antioxidant effectiveness and ability than other antioxidant ingredients. Different test results in the 5% pomegranate extract gel treatment group (P2) with the negative control (K2) had a significant difference, which proves that the 5% pomegranate extract gel could be an antioxidant agent, but the different test results with the 10% ascorbic acid gel (K1) did not have a significant difference; it proved that the 5% pomegranate extract gel with 10% ascorbic acid gel (K1) did not show any difference, which means that the use of the 5% pomegranate extract gel was no better than the 10% ascorbic acid gel.

A bleaching procedure can cause the shear bond strength of the composite resin of the enamel to decrease if the restoration procedure is carried out immediately after the bleaching process because there is peroxide residue that interferes with the resin adhesion, which inhibits the resin polymerisation. The reduction in bond strength is also caused by the loss of calcium caused by residual free radicals after the residual bleaching application; the peroxide ion is granular and porous with a bubble appearance and were trapped in the enamel.

Antioxidant agents are effective in increasing the shear adhesion strength and surface tension of the composite resin restorations after bleaching because they aim to deactivate free radicals. This accelerates the delay time of the restoration procedure because the antioxidants stabilise the electrons so that the enamel surface is uniform and nonporous. Pomegranate peel extract has been shown to be effective removing free radicals and reducing oxidative stress by donating hydrogen atoms to prevent chain reactions of converting superoxide to hydrogen peroxide. The high antioxidant activity of pomegranate extract is related to the potency contained in the extracted content. One of the factors in the extract are tannin compounds; for example, punicalagin, which is classified as an ellagitannin, which is a donor of antioxidant activity in pomegranate extract gel compared to other ingredients.

The 10% pomegranate extract gel and ascorbic acid gel can be an antioxidant agent that is capable of eliminating residual hydrogen peroxide after the bleaching procedure because the higher the extract concentration, the higher the antioxidant activity. The application of the gel will affect the polymerisation of the adhesive material so that the composite resin can penetrate the dentinal tubules. The enamel gap is also deeper; therefore, the bond between the tooth and the adhesive material is stronger. This is in line with previous research, which stated that the antioxidant agent applied after the bleaching procedure can bind free radicals from the effect of the bleaching procedure to minimise the occurrence of a bubble appearance, which can result in a decrease in the strength of the composite resin on teeth after the bleaching procedure using 40% hydrogen peroxide. It can be concluded that the pomegranate gel extract as an antioxidant increased the shear bond strength of the composite resin restoration after the 40% hydrogen peroxide bleaching application.

REFERENCES


