Differences in effectiveness of water steeping from green tea leaf and black tea leaf as hemostasis effect on wound cut-tail mice (mus musculus)

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Abstract

Objective: Tooth extraction is a simple procedure that is frequently performed in the practice of dentistry. Bleeding is a common complication of tooth extraction. There are several materials or methods that may be used to control the bleeding. To minimize side effect, it might need natural ingredients as a replacement. Aims of this study are to determine the differences in the effectiveness of water steeping green tea leaves and black tea leaves as a hemostasis effect on wound cut-tail mice (mus musculus).

Material and Methods: This is an experimental research with study design post-test only control group design. Samples were 30 male mice which divided into three groups. One negative control group (aquadest) and two treatment group (green tea and black tea). Mice that match the criteria is cutted the tail equal to 3 mm from the tip of the tail, then the ingredients is applied to the wound. Blood is dripped on absorbent paper until bleeding stop and data were analyzed using Kruskal-Wallis.

Results: There are significant differences between negative control group and the treatment group. In the treatment groups there were significant differences in the bleeding time between green tea group and black tea group.

Conclusion: As a conclusion, water steeping from green tea leaves have a better effect than black leaves tea as hemostasis on wound cut-tail mice.

Keywords: Bleeding, Hemostasis, Water steeping, Wound cut-tail mice

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Introduction

Tooth extraction or tooth removal is a simple procedure that is frequently performed in the practice of dentistry. Tooth extraction is performed on teeth which can not be treated again, such as caries lesions, severe periodontal disease, or periodontal infections and also often performed in cases of orthodontic or prosthodontic treatment.

The ideal tooth extraction is an extraction of teeth without pain and minimal trauma to the soft tissues but this situation does not always happen according to existing theories. It’s not uncommon where tooth extraction causes complications, either prior to or after the procedure. Complications that can occur at the time of tooth extraction is dry socket, paresthesia, osteomyelitis, oedema and bleeding. In addition to those already mentioned, there are many other complications that can occur prior or after the extraction.

Bleeding is the most common complication of this procedure. Bleeding usually occurs as a result of pressure inside is higher than outside of the blood vessel. Bleeding normally occurs at the time of the extraction but if the bleeding did not stop for 30 until 60 minutes, then the operator must prevent excessive blood loss.

Action that is generally used to prevent bleeding is to apply pressure around the socket after the extraction, the pressure is done by biting sponge or gauze placed over the wound for 30 minutes.

There are several materials or methods that may be used as a control a bleeding, such as the use of cellulose tampons, sponges gelatamps, and cyanoacrylate glue. These materials is believed to reduce the level of bleeding quickly, but these material had side effects that are not good for body and also the price of these material not economically friendly.

Indonesia is a country that has many plants that can be used as herbal medicine, one of the plants that commonly used is tea. Tea has many benefits for our body, such as for the prevention of diabetes, lowering blood pressure, even better, it is used to maintain healthy teeth and mouth.

Tea consists of several types, such as green tea, black tea, oolong tea, red tea and white tea. Every kind of tea contains an active ingredient which is different due to the manufacturing process. In Indonesia, green tea and black tea is one of several types of tea that are very popular among the community.
In dentistry medicine, green tea and black tea are widely used to maintain oral health, as a substitute for mouthwash and to prevent caries. Its also used to cope with bleeding. Some journals say tannin content contained in tea can accelerate the process of haemostasis after tooth extraction. Additionally, antibacterial and anti-inflammatory effect on the tea is widely used as an alternative remedy for wound healing.\(^7,8\)

**Material and Methods**

The study was conducted in Entomology Laboratorium at Faculty of Medicine Hasanuddin University Makassar. These mice is divided into three groups. Each group consists of 10 male mice. One negative control group (aquadest) and two treatment group (green tea and black tea). This is an experimental research with study design post-test only control group design.

Green tea leaves and black tea leaves is prepared by making water steeping. Making water steeping tea leaves is made by boiling the tea leaves at the optimum temperature expenditures chemical content. Steeping of green tea is done by pouring water at a temperature of 80°C for 3 minutes, whereas black tea at 90°C for 3 minutes. Male mice is adapted to its new environment. Feeding is three times a day with a pellet material. Adaptation of mice were conducted during one week. Then, the male mice that had been prepared for experiment is removed from the cage. Cutting the mice tail is done with surgical scissors. The tail is cut about 3 mm from the tip of the tail. Dipping the mice that had been cut in the steeping green tea leaves for 5 seconds. The stopwatch start after blood is dripped on absorbent paper until bleeding stop (the wound should not touch absorbent paper). The stopwatch is stopped when the blood has not been seen on absorbent paper and the time recorded. For the next group, it doing the same procedure, but the treatment group using water steeping of black tea leaves and the control group using distilled water.

Mean and standard deviation were calculated for each group. Comparison were analyzed using Kruskal-Wallis test.

**Results**

A total of 30 male mice (mus musculus) were divided in three group and each group consists of 10 male mice. The mean bleeding time for the green tea group showed 38.84±4.35 seconds, the black tea group 66.97±31.62 seconds, and the negative control group for 183.3±27.83 seconds. This shows that the shortest bleeding time is in green tea group and longest bleeding time is in negative control figure 1.

Significance (probability) from Kruskal-Wallis test is 0.000. This shows that mean of each group is significant statistically because the probability is smaller than 0.05 (p<0.05).

**Discussion**

We use of mice as experimental animals because these animals have the advantage that mice can reproduce rapidly and adaptability to new environments well. Mice used are male mice because of biological conditions are more stable when compared with female mice that are influenced by uterus that can affect bleeding time.

The mean bleeding time in the treatment group showed a substantial difference. This difference shows that each tea contains a different concentration of ingredients. Green tea contains tannins and flavonoids that are quite high.\(^11,12\)

Tannin is an astringent that has a bitter taste when consumed. Tannins have an effect that is very good for the health of which is able to work as a vasoconstricor through astringentna effect, tannin

![Figure 1](image1.png)

**Table 1** Comparability analysis of bleeding time by LSD test

<table>
<thead>
<tr>
<th>Group (I)</th>
<th>Group (J)</th>
<th>Mean Difference (I–J)</th>
<th>Std. Error</th>
<th>Sig.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Green</td>
<td>Black</td>
<td>-28.130000*</td>
<td>10.93329</td>
<td>.016</td>
</tr>
<tr>
<td></td>
<td>Aqua dest</td>
<td>-144.460000*</td>
<td>10.93329</td>
<td>.000</td>
</tr>
<tr>
<td>Black</td>
<td>Green</td>
<td>28.130000*</td>
<td>10.93329</td>
<td>.016</td>
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<tr>
<td></td>
<td>Aqua dest</td>
<td>-116.330000*</td>
<td>10.93329</td>
<td>.000</td>
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<tr>
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<td>.000</td>
</tr>
</tbody>
</table>

Note: Statistically significant is shown in (*) if significance value is below 0.05.
can accelerate the release of proteins from the cell and precipitation, the precipitated protein is albumin. This protein deposition process will induce the synthesis of thromboxane A2 to increase platelet aggregation, thereby accelerating the formation of platelet plugs while the blood vessels are injured.\textsuperscript{8,9}

Flavonoids also one of the chemical compounds found in both types of tea (green tea and black tea) and most abundant in black tea 200mg/cup. Flavonoids play an important role in maintaining the permeability of blood vessels and increase vascular resistance capillary.\textsuperscript{10,11} Flavonoids can also improve endothelial function by reducing oxidative stress. Endothelial function will impact on the activity of platelets, the adhesion of leukocytes and blood vessel muscle cells function.\textsuperscript{12,13}

Tannin and flavonoids levels in black tea and green tea can affect hemostasis work activity by steeping the tea water. In green tea, it had higher levels of tannins and flavonoids that proven to reduce bleeding time compared to black tea which has a lower tannin levels. Shortening of bleeding time can also be caused by other compounds in green tea that work synergistically with tannins and flavonoids.

Cutting mice's tail may also affect bleeding time, as has been explained on research by Tedjasulaksana\textsuperscript{9} showed that the bleeding time of mice with a cut of 0.5 cm from the tip of the tail is shorter than the mice in this study. Tail form that were decreasing in size in the area where the tail end also has shrinking blood vessels that affects the flow of blood. The smaller the bloodstream hurt, the less blood flows out. Changes in blood vessels diameter will causes change in the blood's ability to deliver blood flow.\textsuperscript{15}

Study on mice's tail hemostasis effect is expected to be applied in the bleeding after simple tooth extraction, using mice as experimental animals based on the estimated size of the capillary blood vessels mice equal to the size of capillary blood vessels in human teeth.\textsuperscript{15}

**Conclusion**

Water steeping green tea leaves and black tea leaves have an effect hemostasis on cut-tail male mice. Water steeping from green tea leaves have a better effect than black tea leaves to effect hemostasis on cut-tail male mice.

**Conflict of Interest**

The authors report no conflict of interest.

**References**