Comparison of the Effectiveness of Honey and Bee Pollen in the Repair of Burn in Mice based on Diameter of Burns

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Abstract: Introduction: Burn is a type of trauma that can cause distress, because there is damage and changes in the body's system, causing a variety of complex problems. Burns also have higher morbidity and mortality rates than other injuries. In traditional medicine, honey has been used to treat burns that occur on the skin. When rubbed on the burned area, honey will reduce pain and prevent blisters. Therefore, bee products such as bee pollen thought to have the same effect as honey to treat burns. Method: The purpose of this study is to look at bee keeping products more effectively between honey and bee pollen in treating burns. The methode used in this experimental research is randomized designs post test only control group using experimental animals which were divided into two study groups. Animals were injured on the back, and the handling is done on the burn after some time. Apply honey for one group of mice and bee pollen on the other group of mice which suffered from burns until the wounds are fully covered. Apply daily for 14 days. Measuring the diameter of the wound is done every day in centimeters. The data was processed using the Mann-Whitney U test hypotheses. Result: The results showed a mean diameter of wound repair in the group treated with honey is 0,311 centimeters, and the mean diameter of wound repair in the group treated with bee pollen is 0.333 centimeters. Results from the hypothesis test obtained, p value is 0.884 ($p > 0.05$). Conclusion: The hypothesis test results show that there is no significant difference in the effectiveness of honey and beepollen in treating burns

1 INTRODUCTION

The skin is the largest organ of the human body, of which 15% of the adult human body is skin.1 The skin often undergoes mechanical trauma due to the impact of sharp objects and blunt objects, resulting in discontinuity of skin tissue (wounds). The most common and widespread type of wound in the world is burns.

Burns are a type of trauma that can cause suffering, because of damage and changes in the body system, resulting in complex problems.3 Burns are also events that cause higher morbidity and disability rates than other injuries.

The National Institute of Burn Medicine collects statistical data from various burn centers throughout the United States noting that about 75% of burn patients are caused by self-acts.

Statistics show that 60% of burns occur due to domestic accidents, 20% due to accidents at work, and the remaining 20% for other reasons, such as bombs, volcanoes, etc.3 According to the World Health Organization (WHO), burns cause 195,000 death every year until 2012 around the world, especially in poor and developing countries. Burns that do not cause any death apparently cause disability to the sufferer.

In the United States in 1995-2000, about 2.5 million people suffered burns each year. Where 200,000 patients require outpatient treatment and 100,000 patients are hospitalized. Approximately 12,000 people die each year from burns and inhalation injuries associated with burns. One million working days are lost each year because of burns.

In Indonesia, the mortality rate is quite high, more than 250 people per year die from burns in 2004-2008, where children and the elderly are most likely to experience burn injuries. The most cause of burn incident in Indonesia was caused by explosion of LPG gas cylinder by 30,4%, followed by fire 25,7%,

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and hot water 19,1%. The average patient was treated for 13.72 days with a mortality rate of 34% .

Deaths generally occur in burns with an area of more than 50% or in burns accompanied by respiratory tract injury, and 50% occur in the first 7 days of treatment.3 However, in almost all patients with severe burns or other exfoliation, there is a huge loss of plasma through the peeling areas of the skin, resulting in a decrease in plasma volume. Loss of plasma volume from the circulatory system can significantly lead to shock.

Currently, honey plays a role as an antibiotics as well as topical medicines currently circulating in the community, this is reinforced by some studies of honey as an antibacterial and accelerate the regeneration of damaged tissue cells.9 The content of flavonoids and phenolic acids in honey that function as antimicrobials. Honey has been used since ancient times for wound repair. Indian medicine states honey as a source of life and medicine. Honey has also been used to treat wounds in World War I. Other literature has also stated that honey is useful in reducing infection rates13 and has it is known that honey has antibacterial properties.

In traditional medicine, honey has been used to withstand skin burns. If applied to a burning area, honey will reduce pain and prevent blister formation. High osmolarity content in honey is a preventative of infection and speeds wound healing time. The content of honey acting as an antibiotic is hydrogen peroxide. Other honey content: 17.1% water, total carbohydrates 82.4%, and 0.5%, amino acids, vitamins, and minerals.

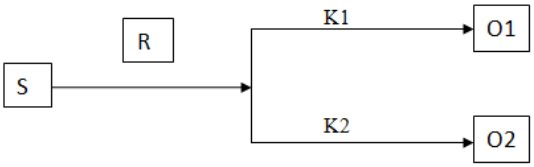
Because of the above, beepollen bee products are thought to have anti-bacterial effects and can accelerate wound healing time just like honey. Thus, this is what underlies the research on the comparison of effectiveness of Honey and Bee Pollen on the process of repair burns.

2 METHODS

2.1 Research Design

2.1.1 Type of Reseach Design

This research is true experimental research with randomized post test design only control group design. The study used experimental animals that were divided into 2 research groups.



Explanation :
S = Sample
R = Randomization
K1 = Treatment group with topical Honey for 14 days
K2 = Treatment group with topical Bee Pollen for 14 days
O1 = Observation result of treatment group by giving honey
O2 = Observation result of treatment group by Bee Pollen

2.1.2 Research Location

The research will be conducted at Biology Laboratory of Faculty of Mathematics and Natural Sciences (FMIPA) Universitas Sumatera Utara

2.1.3 Study Plan

The study is planned to begin in September - December 2016. Research will be conducted after Ethical Clearance approval from the Ethics Commission of the Faculty of Medicine, University of North Sumatra.

2.2 Population and Sample Research

In this study we will use Duoble Distsch Webster mice, 2-3 months of age, 20-30 gram weight, healthy, has never been used for other studies. The number of group try animals was obtained based on Federer's (1963) formula, as follows:

$$(t-1)(n-1) \geq 15$$

Explanation :
n = sample size
t = number of groups of experimental animals
Therefore, the required sample size is:

$$(t-1)(n-1) \geq 15$$
$$(2-1)(n-1) \geq 15$$
$$(n-1) \geq 15$$
$$n \geq 16$$

Sample selection and grouping were done using simple randomization method (simple random sampling). Based on Federer's formula, a sample is required for at least 16 animals in each group. With

the addition of the estimated drop out, then each group of animals required as many as 17 mice. So the number of experimental animals required for this study amounted to 34 individuals who met the inclusion criteria to be numbered and divided into 2 groups. For drop out groups, mice which die during the study period will be excluded from the study sample group.

2.2.1 Inclusion Criteria

1. Mice strain Duoble Distsch Webster, age 2-3 months, weight 20-30 grams.
2. Healthy mice.

2.2.2 Exclusion Criteria

1. Mice with degree I burns
2. Mice with degree III burns

2.3 Maintenance and Preparation of Animal Samples

Prior to the study, the adaptation of animals was tried for 24 hours with a standard diet of eating and drinking ad libitum. Animals sample to be maintained during the study period.

2.3.1 Procedure

Making Burns Degree II. Shave the backs of mice. Perform anesthesia on the back with a dose of 0.2 cc Lidocaine. The skin is injured by dripping the skin of mice with water 100o Celsius temperature using 5 cc syringe for 7 seconds. After 7 seconds, we suck back the water using spiut. In order to cut circular wound, we limit it with iron or metal in the form of a circle with a diameter of 2 centimeters.

Handling Procedures for Burns Degree II. Handling done twice a day is always cleaned before applying honey and Bee Pollen to mice. Before smearing honey and beepollen, researchers first observed the mice burns to see the repair of wounds in the mice every day. After the observation, the researchers measured the diameter of the wound in the mice to see if there was an improvement in burn wound diameter. Measurements are made by measuring the distance farthest from one side to the burn side by using a ruler in centimeters. Measurements are made only once a day. After measuring the diameter of the wound, then the honey and beepollen are smeared. Splash honey in the group with honey (K1) and Bee Pollen in the group with Beepollen (K2) to the back of the mice that burns until the entire wound is covered. Observations, measurements and honey and beepollen were done daily for 14 days in centimeters.

2.4 Operational Definition

Research Title: Comparison of the effectiveness of honey and bee pollen in the repair of burns in mice based on diameter of burns.

1. The diameter of burns
   a. The diameter is the furthest distance formed by a burn
   b. Means of Measure: two dimensional assessments (calculation length x width)
   c. Measurement tool: ruler
   d. Measure result: value by kilogram
   e. Measurement Scale: ratio
2. Repair Burns
   a. Repair of burns is a reduction in burn wound diameter
   b. Means of Measure: two dimensional assessments (calculation length x width)
   c. Measurement tool: ruler
   d. Measure result: value by kilogram
   e. Measurement Scale: ratio

2.5 Data Analysis

The data will be analyzed by ANOVA if the parametric data is normally distributed. If the data is not normally distributed, then the data taken is non-parametric and tested with Mann-Whitney U.

3 RESULT

3.1 Reseach Result

3.1.1 Description of Research Location

The research was conducted at Biology Laboratory, Faculty of Mathematics and Natural Sciences University of Sumatera Utara (FMIPA USU) Medan, located at Jalan Bioteknologi no. 1, University of Sumatera Utara Campus, Padang Bulan Village, Medan Baru District, Medan. This location is chosen by considering the completeness of facilities and supporting facilities are good to support the success of this research.

3.1.2 Description of Research Location

In this study, the samples used were 18 male Double Distsch Webster male strips.

Table 1. Table Repair the Hydraulic Diameter on Group Honey for 14 Days in Centimeter units

Days	Mice 1	Mice 2	Mice 3	Mice 4	Mice 5	Mice 6	Mice 7	Mice 8	Mice 9
1	2,0	2,0	2,0	2,0	2,0	2,0	2,0	2,0	2.0
2	1,8	1,9	1,7	1,8	1,9	1,5	1,8	1,8	1,6
3	1,8	1,7	1,7	1,7	1,7	1,4	1,6	1,6	1,6
4	1,5	1,5	1,5	1,5	1,5	1,3	1,5	1,5	1,2
5	1,4	1,5	1,5	1,5	1,5	1,3	1,5	1,5	1,2
6	1,4	1,4	1,5	1,5	1,5	1,3	1,4	1,5	1,1
7	1,3	1,3	1,5	1,4	1,5	0,9	1,3	1,3	1,1
8	1,2	1,1	1,3	1,4	1,4	0,7	1,3	1,3	1,1
9	1,2	1,1	1,2	1,3	1,4	0,7	1,3	1,2	1,1
10	1,2	1,1	1,2	1,3	1,3	0,7	1,3	1,2	1,1
11	0,9	1,0	0,7	1,0	1,0	0,4	0,9	1,1	0,8
12	0,7	1,0	0,5	0,7	0,7	0,2	0,7	1,1	0,5
13	0,5	0,9	0,0	0,5	0,4	0,1	0,6	1,0	0,3
14	0,0	0,9	0,0	0,5	0,0	0,0	0,6	1,0	0,0

Based on table 1, it can be seen that in the group of beepollen administration there is an improvement in progressive burn wound diameter in mice 1, 3, 5, 6, and 9. This is obvious because in mice 1, 3, 5, 6, and 9 have healing total at day 14. In mice 2, 4, 7, and 8 also happened improvement of burner diameter, but improvement of burn diameter not as good as in mice 1, 3,5, 6, and 9.

Table 2. Mean of wound diameter on group of honey and beepollen in centimeter

Sample group	N	Average Diameter	Standard deviation
Honey	9	0,311	0,4833
Beepollen	9	0,333	0,4213

Based on the results of the research in Table 2 above, the group with the meanest diameter is the group with the Honey, ie 0.311 ± 0.4833 centimeters. While the group with the largest mean diameter is the group with the provision of Beepollen, which is 0.333 ± 0.4213 centimeters.

A. Data Normality Test

In this study, the data normality test used is the Shapiro-Wilk test. Data is normally distributed if p value> 0,05 and is not normally distributed if p value <0,05. The results of normality test data can be seen in Table 3 below.

Table 3. Normality of Wound Repair Diameter with Shapiro-Wilk Test

Sample group	Statistic	Df	Sig.
Honey	0,732	9	0,003
Beepollen	0,772	9	0,010

Based on table 3, the significance value for the honey group is 0.003 and 0.010 for the beepollen group. Because the significance value of both groups <0.05, it is said that the data is not normally distributed.

B. Homogeneity Test

Homogeneity tests were conducted to see if the variants of some populations were the same (homogeneous). If the significance value is <0.05, then it is said that the variant of the population data group is not the same. Vice versa.

Table 4. Homogeneity test of impaired diameter of wound on giving group

Levene Statistic	df1	df2	Sig.
0,000	1	16	1,000

Based on homogeneity test results in the above table, obtained p value of 1,000, whose value> 0,05. It is known that the population data group in this study has the same variant (homogeneous).

C. Comparability Test

The comparability test was aimed to compare mean of wound diameter in honey and beepollen group. Comparative analysis used is a test for non parametric data, namely mann-whitney u.

Table 5. Data comparability test using the mann-whitney u hypothesis test

Sample Group	Day 14
Asymp. Sig. (2-Tailed)	0,884

After a nonparametric hypothesis test with mann-whitney u method with significance level of 0.05 (α = 5%,), p value was 0.884 (p> 0.05), which means no significant difference in effectiveness on honey and beepollen in repair burns

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4 DISCUSSION

This study is a true experimental design study with the aim to compare the effectiveness of honey and beepollen in the repair of burns based on burn diameter.

Based on the statistical test value in Table 2, it can be seen that the mean of wound diameter in honey group is smaller than the mean of beepollen group (0,311 <0,333). This is in accordance with previous research, Noori S. Al-Waili et al, where it is said that honey can accelerate the healing time of superficial burns and burns in .25

This may be due to previous studies, honey has been shown to be significant (p <0.001) in prospective, randomized, clinical trials, faster in curing superficial burns. In a previous study conducted on 52 burn patients treated with honey, 91% wound healed with steril within 7 days. Molan P.C found that healthy tissue granulation was faster in patients treated with honey (mean 7.4 days). Within 7 days, 84% of wound treated with honey showed a satisfactory epithelialization process, and recovered 100% within 21 days.14

The improvement of wound diameter in the honey group in Table 2 can also be due to the fact that Honey has a high sugar content and also high osmolarity, which can inhibit microbial growth.26 Honey can also reduce inflammation, swelling and pain in burns quickly, accelerate the release of necrotic tissue, accelerate granulation and reepithelization with minimal scarring.27 without causing resistance.25

In the group of beepollen based on Table 2 the mean diameter of 0.333 was found, which was larger than the honey group, but not so different from the honey group. This can be due to beepollen is also a bee product whose composition is not much different or almost the same as the composition of honey.

Researchers did not find previous studies that specifically looked for a comparison of the effectiveness between honey and beepollen in repair of burn wound diameter.

However, previous research by Vassev et al., Stated that beepollen also has a high anti-inflammatory effect that can prevent the activity of cyclooxygenase and lipoxygenase, an enzyme responsible for converting arachidonic acid into prostaglandins so as to minimize or eliminate pain after animals try to get burned. Beepollen also has a high antibiotic effect present in flavonoids and phenolic acids, which can shorten the wound healing time28.

It is said by Harbone, J.B, flavonoids are phenol compounds known to have potential as antioxidants and anti-inflammatory. In addition, flavonoids can also cause damage to the arrangement and changes in the permeability mechanism of the bacterial cell wall thus accelerating the wound healing process29.

From the results of the study it was found that there was no significant difference in effectiveness in honey and beepollen in the repair of burns, so this is not in accordance with the initial hypothesis. So it can be concluded that honey and beepollen have the same effectiveness in the repair of burns. However, further research on honey and beepollen related to the repair of burn diameter is required.

5 CONCLUSION

Based on the results of research conducted on the comparison of effectiveness of Honey and Beepollen in the repair of burns in mice based on the diameter of burns, can be drawn some conclusions as follows:

1. The mean of burn repair diameter in honey group is 0,311 centimeter.
2. Average diameter of burn repair in Bee Pollen group is 0,333 centimeter.
3. There was no significant difference between the mean time of repair of burn diameter in both groups because the obtained value p = 0.884.

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