The Effect Of Giving Salak Skin Extract (*Salacca Zalacca*) On Hair Growth And Histopathological Picture Of Skin Tissue In The Healing Process Of Dermapen Wounds In White Rats Of Male Wistar Strain In Obesity Model

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ABSTRACT

Vitamins, minerals, and antioxidants found in the skin of snake fruit (salacca zalacca) contribute to better hair health. The vitamins and minerals found in the skin can promote hair growth, improve hair quality, and reduce hair damage.The purpose of this study was to determine how snake fruit skin extract can accelerate the healing of wounds from dermapen and hair growth in obese Wistar white rats. This study used a quantitative experimental design research method with a post-test only group design.Treatment group 1 was given topical treatment with 5% salak skin extract cream given once a day for 21 days. Treatment group 2 Treatment group 2 mice were given topical treatment with 10% salak skin extract cream given once a day for 21 days. Treatment group 3 mice were given topical treatment with 15% salak skin extract cream given once a day for 21 days.. The results of this study, namely the extract of snake fruit skin, showed the presence of alkaloids and flavonoids that play a role in hair growth.With an average result of applying the cream extract in treatment group 3 which experienced the longest hair growth, namely 0.85 cm..So it can be assessed that the extract of snake fruit skin cream can accelerate the growth of hair follicles in male Wistar strain rats (Rattus norvegicus).

Keywords: *Salak Peel Extract, Wound Healing, Hair Growth*

INTRODUCTION

A series of studies have shown that various types of herbal plants have the ability to overcome the problem of hair loss by activating hair growth, including katuk leaves, brahmi, betel leaves, bitter melon leaves, shallots, licorice, bhringhaj and guava leaves. Knowing about plants that have the ability to fertilize hair, testing was continued until the manufacture of shampoo for hair was made. Healthy hair is an important factor for a healthy physique and women can display charm, beauty, and personal strength using healthy hair. Therefore, hair loss can be a psychological problem for women compared to men. Hair loss is a universal problem for many people in the world, this condition can occur on skin areas such as the scalp, body and face. Various aspects contribute to hair loss such as genetics, hormones, nutritional status, and environmental exposure (exposure to radiation, environmental toxins), and drugs(Mercedes, 2021). Hair loss is also called alopecia. Alopecia is a condition, where hair loss is found from the head or body parts of the hair naturally. This condition can cause a lack of psychological and social confidence. There are different categories of alopecia but the most common are androgenic alopecia (generalized baldness), alopecia areata, and chemotherapy-induced alopecia. The causes of these conditions are many, such as stress, heredity, hormones, nutrition, some diseases, and certain medications.(Rambwawasvika, H., 2021). In general, all forms of alopecia shorten the hair growth cycle and cause hair loss in two ways. Firstly, shortening of the anagen phase as is characteristic of androgenic alopecia. This results in a decrease in the anagen:telogen hair ratio from the normal 6:1 to as low as 2:1 and thus prolonging the telogen phase. Secondly, shrinkage of the dermal papilla, which is responsible for hair follicle cell differentiation and growth through nutrient supply. The shrinkage of the dermal papilla is caused by vasoconstriction of the blood vessels that supply nutrients and oxygen to the hair. This causes a change in the diameter and appearance of the hair resulting in a sudden change from thick, pigmented hair to distorted vellus hair (thin and white).(Orasan, MS, Roman, II, Coneac, A., Muresan, A., 2016)The main characteristics consumers want in the hair care category are natural ingredients, botanical ingredients, and ‘free from’ claims (such as free from salts, sulfates, silicones, parabens, and other components considered harmful)(Abelan, US, 2022)This is also supported by the many components of nature that have many functions, one of which is black rice. The vitamins, minerals, and antioxidants found in the skin of the snake fruit (salacca zalacca) contribute to better hair health. The vitamins and minerals found in the skin can promote hair growth, improve hair quality, and reduce hair damage. Overall(Andarwulan, 2010). Salak skin (salacca zalacca) has many benefits that can help improve hair growth. One of them is the presence of vitamins and minerals that help improve hair quality and reduce damage.

LITERATURE REVIEW

Hair shaft pigmentation ensures many benefits including UV protection, thermoregulation, and sexual perception. In addition, the hair pigment, melanin, is a powerful free radical scavenger. Therefore, melanin production within the active anagen hair bulb may help withstand cellular stress caused by reactive oxygen species.(Erdogan, 2017).Hair growth occurs in a continuous process characterized by three phases: anagen, growth; catagen, regression; telogen, resting. The length of the hair cycle for each phase can vary between different areas of the body. Unlike other mammals, human hair growth and loss occur in a mosaic and asynchronous pattern. At any given time, a percentage of hairs depending on a particular location will be in one of three main hair cycle stages, with the duration of anagen on the scalp being calculated to be 4–7 years, catagen 2–3 weeks, and telogen approximately several months.(Orasan, MS, Roman, II, Coneac, A., Muresan, A., 2016). The hair shaft is divided into three layers, namely the cuticle, cortex to the medulla case fragment. The formation of flat and square cuticle cells adheres tightly to the cortex cells proximally. Peripheral activity of cuticle cells makes the distal free edge direction upward and produces a widespread overlap. This imbrication is very critical. In locking with cuticle cells on the inner root cap, imbrication plays a role in creating an anchor for growing hair follicles. This imbrication background has a facility for removing dirt to cells that are peeled off from the scalp.(Erdogan, 2017)argues that the cuticle also has a function that protects important parts and the usefulness of the cover on physical and chemical problems. On the hair shaft there is also a hair shaft surface in the form of:

a. Cuticle or hair membrane is the upper surface of the hair shaft, composed of 7 to 10 flat horn cells, hard to transparent or able to penetrate light. The cuticle has a protective function in the hair, so that the hair looks shiny and the combing process becomes easy. The cuticle has a function as a hair elasticity tool, namely the activity of stretching the hair shaft to make the hair shaft position return to its original state without breaking. The cuticle is fragile when exposed to sharp friction. A healthy hair shaft has a loose and emerging cuticle, in contrast to damaged hair which will look very loose.

b. The cortex or hair skin is composed of a collection of fine threads with keratin or horn cells. Each horn cell can be broken down into a finer unit, namely protofibril. In microfibrils, there are 11 keratin molecules, namely protofibrils.

c. Medulla or hair marrow is the central organ of hair composed of small horn cells with an uncertain shape. Medulla contains hair keratin. Cells have a poly-global shape, look sparse with each other. Has the function of delivering sebum to the hair shaft and regulating the evaporation of the hair shaft.

Hair follicles and hairs can be identified in skin biopsies under the light microscope. Under the light microscope, hair follicles are easily distinguished from the surrounding dermis. The hair-bearing tissue can reveal several longitudinal, oblique, and cross-sectional views of the hair follicle. Associated structures such as muscle fibers of the arrector pili muscle and/or sebaceous glands of the pilosebaceous unit can be helpful in defining structures unknown to be hair follicles.(Brown, TM, & Krishnamurthy, 2018). Salak skin can be used as herbal medicine to prevent and cure diseases. Bioactive components in the skin of this fruit can be beneficial for health. It is known that salak skin has antioxidant, antimicrobial, antidiabetic, anticancer, antihypertensive, anticholesterol, antihyperuricemia, and immune system maintenance properties. The results of phytochemical screening of salak skin show that there are compounds such as flavonoids, saponins, phenols, tannins, triterpenoids, and alkaloids. Flavonoids in salak skin are among the compounds that have an important role in carrying out various pharmacological functions. For years, flavonoids have been used as a base ingredient for pharmaceutical and cosmetic products. However, most flavonoids do not mix with water, meaning they cannot be used well in oral form and are not effective as drugs.(E. Susilowati S., A. Rahmadani, L. Meylina, 2020)

METHODS

The type of testing used is quantitative experimental. The purpose of quantitative experimental is the use of true experiment design or laboratory experiment. True experiment is an experimental test that is carried out seriously and all external variables are controlled by the experimental activities influenced. This study was conducted post-test only group design to determine and analyze the effect of giving snake skin extract (*Salacca zalacca*) in accelerating the procedure of dermapen scars on the backs of white rats (*Rattus norvegicus*) wistar strain suffering from obesity healed. The test sample used was a male rat (*Rattus norvegicus*) wistar strain weighing 160 to 200 grams and aged 2 to 3 months. The purpose of selecting male wistar rats as test material is because rats have specifications to physiology that resemble humans and are used as dominant animals used in biomedical testing. *In vivo* studies of this research were conducted and supervised under Universitas Prima Indonesia ethical committee, number: 045/KEPK/UNPRI/X/2024. Mice are test animals that are kept in groups in experimental animal cages in the laboratory. The mouse cages are made of plastic measuring 30 cm x 20 cm x 10 cm covered with fine wire mesh. Rice husks 0.5 to 1 cm thick are installed at the bottom of the cage and replaced every day during the study. The lights in the room are set to produce a 12-hour light/12-hour dark cycle. The temperature is set to 25–27 °C, and the humidity is set to normal 35–50%. Mice are given distilled water ad libitum and regular mouse pellets. The test animals that have passed the acclimation phase are then divided into 4 groups

in no order. Each group has 6 mice. Each mouse is labeled with a waterproof marker on its tail. In the control group, mice were only given 0.9% NaCl. However, the group was given mice given a solution of snake skin extract (*Salacca zalacca*) at different doses:

1. Control Group (P0): fed rat pellets + 0.9% NaCl/day/head for up to 14 days.
2. Treatment Group I (P1): fed rat pellets + Salak skin extract at a dose of 300mg/kgBW plus distilled water/day/head for up to 14 days.
3. Treatment Group II (P2): fed rat pellets + Salak skin extract at a dose of 500mg/kg BW and given distilled water/day/head for up to 14 days.
4. Treatment Group III (P3): consuming rat pellets + Salak skin extract with a dose of 700mg/kg BW given distilled water/day/head for 14 days. The doses in this study were made differently to determine which dose was more effective from all the variations of doses given.

Dermapen wound healing consists of several phases, namely the inflammation, proliferation and maturation phases. In the proliferation phase, fibroblasts play an important role in producing proteins for wound healing, one of which is collagen.

RESULTS

Based on the results of observations made on all groups as in **Table 1**, it shows that there is a healing process of dermapen wounds in white rats (*Rattus norvegicus*) Wistar strain. There is a difference in the average percentage of healing from the control and treatment groups. The average percentage of healing of dermapen wounds on the last day of the control group was 70.5%. Treatment group 1 got a wound healing percentage of 100% on the 13th day, while treatment groups 2 and 3 on the 12th day. The researcher concluded that the control group did not experience total healing and treatment groups 2 and 3 required the fastest time to experience total healing than treatment 1.

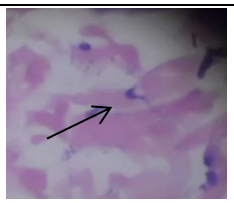
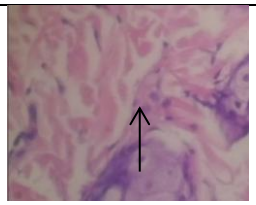
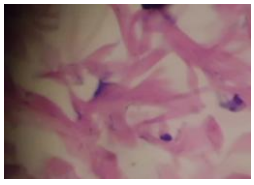
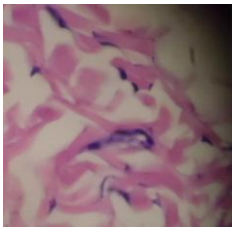
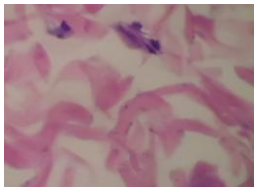
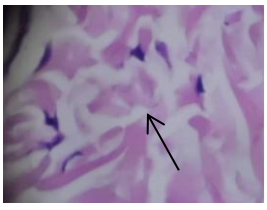
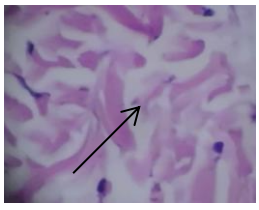
Table 1. Average Percentage of Wound Healing (%)

Day	Control	P1	P2	P3
1	0	0	0	0
2	1.5	2	3	4.5
3	2.5	4.5	14.5	17.5
4	4.5	7.5	23.5	24.5
5	9	14.5	34.5	37
6	11.5	17.5	39.5	67.5
7	16	34	41	69.5

8	19	39.5	47.5	73.5
9	20.5	44.5	56	89.5
10	38.5	57.5	76	92.5
11	44.5	74.5	94.5	99
12	53	87.5	100	100
13	57	100	100	100
14	70.5	100	100	100

Dermapen wound healing consists of several phases, namely the inflammation, proliferation and maturation phases. In the proliferation phase, fibroblasts play an important role in producing proteins for wound healing, one of which is collagen. The results of histopathological within **Table 2,** examination under a 400x magnification microscope shown that wound healing concurrent with the number of differences in collagen density:

Table 2. Histopathological Description of Skin Tissue

No	Group	Histopathological Image of Skin Tissue	
1	Control (0%)		
2	P1 (10%)		
3	P 2 (15%)		
4	P 3 (20%)		

Phytochemical Screening Test Results

Screening test results content of compounds in the salak plant in **Table.**3, is widely used by the community. From various studies, the salak plant is considered a fruit plant that has properties from the flesh and skin of the fruit. The sweet flesh of the fruit and its nature that grows in the wild are useful as medicine for people with diabetes and others. The results of the phytochemical test of the salak skin extract can be seen in the following.

Table 3 Results of Phytochemical Test of Salak Skin Extract

Secondary Metabolites	Results
Alkaloid	+
Flavonoid	+
Saponins	+
Tannin	+
Steroids/Triterpenoids	-
Glycosides	+

Antioxidant Activity Test

Dtermining the antioxidant activity test, it starts from determining the wavelength, determining the operating time and determining the antioxidant activity of the ethanol extract of snake fruit skin. The levels of antioxidant activity can be explained in the following in **Table 4** :

Table 4. Antioxidant Activity Levels of Salak skin extract

Mark	Levels
IC50 < 50 µg/ml	Very strong
IC50 50-100 µg/ml	Strong
IC50 101-150 µg/ml	Currently
IC50 > 150 µg/ml	Weak

In making 0.5 mM DPPH solution (200 ppm), 10 mg of DPPH powder was weighed and dissolved in methanol up to 50 mL. Meanwhile, to measure the maximum absorption wavelength of DPPH by pipetting 1 mL of DPPH standard solution. put into a 5 mL volumetric

flask. then methanol was added to the mark so that a solution with a concentration of 40 ppm was obtained. The maximum wavelength was measured using a UV-Vis spectrophotometer (400 nm - 800 nm). The maximum wavelength was obtained at 515 nm. To make the extract test solution, 10 mg of thick extract was weighed and dissolved in methanol up to 10 mL. a solution with a concentration of 1000 ppm was obtained. Taken 0.1 mL; 0.2 mL; 0.3 mL; and 0.4 mL; from the 1000 ppm extract solution. then added 1 ml of DPPH solution (concentration 200 ppm) to each concentration and added with methanol to the mark limit (5 mL measuring flask). obtained concentrations of 20, 40, 60, and 80 ppm. Incubated for 30 minutes then measured the absorbance using a UV-Vis spectrophotometer at a maximum wavelength of 515 nm. Determination of the free radical trapping process by the test sample using the DPPH free radical trapping method obtained different results. The results of the 20 ppm concentration were 33.6604%, the 40 ppm concentration was 52.6987%, the 60 ppm concentration was 70.0686%, the 80 ppm concentration was 74.5892%. For the calculation of the results of the DPPH trapping method is to calculate IC50. This value indicates that the plant extract can cause a reduction of 50% of DPPH activity. The results obtained were 44.0931 ppm, which means that based on the antioxidant activity level table, it is included in the very strong category. This can also be seen from the color change of the test sample which is dark purple when DPPH is added, it will turn yellowish if the extract has a reduction.

DISCUSSION

The normally distributed data were then retested using the paired T test. This test was conducted to see the effectiveness of the treatment, marked by the difference in the average before and after the treatment. The results of the paired t test at the sig value. (2 tailed) were 0.00 <0.05 which explained that there was a difference in the average growth of hair follicles resulting from the treatment of snake fruit skin extract cream in each treatment group and the final results on the 21st day with the control group category, treatment 1 with 5% snake fruit skin extract, treatment 2 with 10% snake fruit skin cream extract, treatment 3 with 15% snake fruit skin cream extract. By administering snake fruit skin cream extract, it is effective in the growth of hair follicles in white rats (Rattus norvegicus) Wistar strain. Normality test results usingKolmogorov-Smirnov test it is known that the significant value of the control group is 0.147> 0.05 then the data is normally distributed, the significant value of treatment group 1 is 0.227> 0.05 then the data is normally distributed, for the significant value of treatment group 2 is 0.197> 0.05 then the data is normally distributed, then the significant value of treatment group 3 is 0.212> 0.05 then the data is distributedin a waynormal.homogeneity results using Levene's Test. For decision making, the guideline is if the significance value <0.05 means the

data is not homogeneous, conversely the significance value > 0.05 means the data is homogeneous. From the homogeneity test table data above, it can be seen that the sig. value (2 tailed) is 0.01 <0.05, so it can be said that there is a significant difference in hair growth between groups. The results of the One-Way Anova test in the table above show that the significance value produced is 0.000 or <0.05. Based on these data, it can be concluded that there is no significant difference between the control group and the treatment group. Further Post-hoc LSD tests were conducted to analyze the differences in average wound healing between groups. Further Post-hoc LSD test results The results of the analysis showed that there was a significant difference between the control group and treatment groups 1 (p = 0.000), 2 (p = 0.000) and 3 (p = 0.000). While treatment group 2 and treatment group 3 did not show any significant difference (p = 0.000).

CONCLUSION

1. Active substance content In the phytochemical test of snake fruit skin, it can be seen in the Alkaloid Examination, showing positive results for alkalolides and flavonoids in the snake fruit skin extract. Where this content is a secondary metabolite content that is suspected as a compound responsible for hair growth. The antioxidant properties of flavonoids can stimulate hair growth by relaxing the muscles in the blood vessels in the hair follicles.
2. In the antioxidant test of snake fruit skin using the DPPH method, the results showed that the antioxidants in snake fruit skin extract were weak with a ppm concentration ranging from 151-200. This may be due to the influence of oxidation, light and chemical changes in snake fruit so that if it is oxidized, its structure will change its function as an active ingredient and will decrease.
3. The results of this data were taken with observations for 21 days for the control group, treatment 1 and treatment 2 and treatment 3. With an average result of applying the cream extract in treatment group 3 which experienced the longest hair growth of 0.85 cm, the control group got a result of 0.52 cm, treatment group 1 got a result of 0.68 cm, and treatment group 2 with hair extension of 0.79 cm on the 21st day of observation. So it can be assessed that the extract of salak skin cream can accelerate the growth of hair follicles in male wistar rats (Rattus norvegicus).

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