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The Effect of Andaliman Fruit Extract on the Incision Wound Healing the Wistar Male White Rats and Collagen Type III

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ABSTRACT

The extract of andaliman fruit has antimicrobial activity. The active compounds of andaliman fruit are believed to positively contribute to human health, one of which is flavonoids, which can improve the wound healing process because of their function as an antioxidant and anti-inflammatory. This study aims to determine the effect of giving ethanol extract of andaliman fruit on the healing process of incision wounds in Wistar strain white rats and collagen type III. This research is an experimental posttest-only controlled group design. This study used five groups: NaCL treatment, betadine treatment, 1% andaliman ethanol extract treatment, 3% andaliman ethanol extract treatment, and 5% andaliman ethanol extract treatment, where each group had six rats. Data analysis used normality tests, homogeneity tests, and ANOVA tests. The results showed that the ethanol extract of andaliman fruit contains several active compounds such as alkaloids, saponins, tannins, and glycosides. The fastest incision wound healing was the fourth test group, the test group with 3% andaliman fruit ethanol extract treatment on day 14, with an average length of incision wound healing of 3.44mm. The 3% andaliman ethanol extract had the potential to accelerate wound healing activity.

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INTRODUCTION

A wound is the loss or damage of part of the body's tissue due to a factor that disrupts the body's protective system. Wounds often appear on the skin, causing damage to the skin epithelium or disruption of the standard anatomical structure of the tissue due to trauma. Cuts can occur intentionally (surgical wounds) or unintentionally (incidental wounds) due to sharp objects. Its characteristics are open wounds, pain, and the length of the wound being more significant than the depth of the wound (Nabeela, 2017).

A cut wound is a form of damage or loss of body tissue caused by a sharp object and can cause bleeding by involving the role of hemostasis and, eventually, inflammation. Cuts can be treated with chemical and traditional medicine. Traditional medicines that can help wound healing are plants that contain active flavonoids, alkaloids, tannins, and saponins. Cuts cause linear tears in the skin and underlying tissue. It cuts the infection risk and will worsen if not treated immediately. Wound management generally uses antiseptics. The disadvantage of antiseptics is that they can cause irritation and discoloration of the skin and cause scarring, leaving marks on the skin. In the healing process, plants can be used as drugs or sources of medicinal raw materials with the hope of having side effects (Nurihardiyanti et al, 2020).

Basic Health Research (RISKESDAS) data from 2018 show that the prevalence of cuts/slices/stab wounds in Indonesia is an average of 20.1%, with the highest rate in the Papua region at 38.5%. In North Sumatra, the incidence of cuts/slices/stab wounds is 23.9% (Ministry of Health RI, 2018). When a wound occurs, the skin requires the repair of damaged tissue, which must be replaced with a new matrix to rebuild the integrity of the epidermal tissue (Purwanto et al., 2022).

Plants are the most significant sources of new compounds with properties that include antimicrobial, antiviral, antioxidants, and others. According to World Health Organization, the properties of plants provide a way to combat disease (Tahir et al., 2019). The community increasingly favors treatment using natural

ingredients because it is cheap, easy to obtain, and has few side effects. Many plants around us have yet to be put to good use. This can happen due to limited information to the public; for this reason, it is necessary to develop research on traditional medicinal plants. One medicinal plant with many benefits is the Andaliman plant; all parts of the Andaliman, from the root to the tip of the fruit, including the flowers and fruit, have a function. Andaliman fruit has many benefits, including preventing infection in open wounds based on research, a gel preparation of 70% ethanol extract of Andaliman fruit affected healing wounds in rabbit test animals (Janice et al., 2023).

Andaliman (*Zanthoxylum acanthopodium DC*) is a typical plant found in North Sumatra. The fruit is often used as a seasoning for traditional Batak cuisine. Andaliman fruit extract (*Zanthoxylum acanthopodium DC*) has antimicrobial activity. The extract of andaliman fruit (*Zanthoxylum acanthopodium DC*) also has compounds of flavonoids, saponins, tannins, alkaloids, steroids, and terpenoids (Tanessa et al., 2023). Flavonoid content in andaliman fruit can also improve the wound healing process because of its function as an antioxidant and anti-inflammatory (Siregar, 2000). Flavonoids are compounds called quercetin, which are believed to increase the number of blood components and play an essential role in the body to stop bleeding caused by the rupture of a blood vessel. Flavonoids play a role in the wound healing process because they are useful as anti-inflammatory and antimicrobial (Nuari et al., 2017).

Collagen plays a crucial role at every stage of the wound healing process. It can regulate homeostasis, interact with platelets, interact with fibronectin, increase fluid exudation, increase cellular components, increase growth factors, encourage fibroplasia, and sometimes promote epidermal proliferation (Nining, 2020).

Collagen type III plays an essential role in skin wound healing, forming a temporary matrix that guides inflammatory cells and fibroblasts to the wound site. It is also found in adult human cartilage. It is thought to act as a modifier of the fibril network composed of collagen type II and other small collagens during tissue healing (Makuszewska et al., 2020). Accordingly, the study was needed to test the effect of an ethanol extract of andaliman fruit (*Zanthoxylum acanthopodium DC*) on the wound healing process in white male Wistar rats and the level of collagen type III.

METHOD

The research design with a post-test-only control group is a laboratory experiment on white Wistar rats with incision wounds. This design allows the researcher to measure the effect of the treatment (intervention) on the experimental group by comparing the experimental group with the control group.

This research took place from September 2023 to December 2023. The study was conducted in a laboratory with complete equipment and adequate experience. Extraction of andaliman fruit, maintenance of test animals, and skin histopathology examination were performed at the Pharmacology Laboratory, Faculty of Pharmacy, Universitas Sumatera Utara. Meanwhile, the collagen type III examination conducted in Univeristas Prima Indonesia, Faculty of Medicine Laboratory. The Health Research Ethics Committee from Universitas Prima Indonesia, No: 042/KEPK/UNPRI/X/2023, has approved all study procedures.

Several tools will be used in this research, including laboratory glassware, animal cages, injection syringes, tissues, scalpels and blades, surgical scissors, tweezers, fixation tools, stirrers, glass covers, microscopes, electronic gram scales, separating funnels, beakers, cameras, blenders, rotary evaporators, gloves, and documentation tools. The research materials used were andaliman fruit (*Zanthoxylum acanthopodium DC*), NaCl 0.9%, povidone-iodine, ethanol, and Elisa kit sandwich collagen type III.

The experimental animals used in this study were male white Wistar rats. The selection of rats as experimental animals is based on the consideration that genetically, rats are similar to humans and can adapt to the laboratory environment. Researchers chose to use 30 rats. The rats were divided into five groups, each with six rats. One positive group, one negative control, and three treatment groups. Negative control (K-) is a Wistar male white rat with a cut wound given 0.9% NaCl. Positive control (K+) is a male white rat with a Wistar wound and treated with Povidone Iodine 10% 2 mg. P1 is a male white rat Wistar strain with a cut wound and treated with andaliman fruit ethanol extract (*Zanthoxylum acanthopodium DC*) 1% of 1 mg given every 24 hours. P2 is male white rats from the Wistar strain with wounds and treated with andaliman fruit ethanol extract (*Zanthoxylum acanthopodium DC*) 3% of 1 mg applied every 24 hours. P3 is a Wistar strain male white rat with a cut wound and treated with andaliman fruit ethanol extract (*Zanthoxylum acanthopodium DC*) 5% of 1 mg applied every 24

hours. Observe the changes that occur in the wound. Inclusion criteria include male white Wistar rats aged 6-8 weeks, with a body weight of 150-200grams, and healthy conditions as long as the rats are given an incision in the back area along 2cm.

The andaliman extraction process begins with drying 8000grams of andaliman fruit by aerating for eight days. After the andaliman fruit is dry, it is mashed until it becomes simplisia. So, the dry andaliman powder (Simplisia) obtained is as much as 2,088.51grams. After obtaining dry andaliman powder (Simplisia), the resulting simplisia 2,088.51 is then macerated (extracted) using a 96% ethanol solvent of 15000ml for three days with two repetitions. In 1 time, 24 hours, stirred for 10 minutes. After that, the macerated extract was filtered. Then, the liquid extract of andaliman was obtained in as much as 10,820ml. After obtaining the andaliman liquid extract, the extract is carried out by a Rotary Vacuum Evaporator and placed in a water bath until the andaliman concentrated extract is obtained. The concentrated andaliman extract obtained was 108 grams (Djuang et al., 2022). The concentrated extract of andaliman fruit is tested using phytochemical methods, including phenotypic, flavonoid, tannin, alkaloid, triterpenoid, and steroid tests.

Before the wounding, rats were anesthetized using 10% ether by inhalation. Then, the fur around the back was shaved with a diameter of 3cm and cleaned with 70% alcohol. This treatment was done equally to all test animals. The incision was made on the rat's back using a sterile slingshot, making a 2 cm long incision with a depth of 2mm (Handajani, 2021).

Wound length was measured using a slide ruler on days 3, 7, 11, and 14. On day 14, fragments of skin tissue containing wounds with a 1cm margin were collected and fixed in a 10% standard formalin buffer solution for further histopathological assessment. Furthermore, rat blood was taken and converted into serum for further ELISA analysis of collagen type III in rats. The examination of collagen type III using an ELISA kit (Rats Collagen III, Col 3 BT Lab kit, Shanghai, China) was performed according to the instructions listed.

Data normality and homogeneity tests were performed. Normal and homogeneous data were continued with the ANOVA test. All data analysis was carried out using statistical software. This study's statistical test decision was taken at the actual 5% (p=0.05), which is considered significant. If the p-value<0.05 is obtained, the Levene's test is carried out.

RESULTS

Table 1. Phytochemical Screening of Andaliman Fruit Extract *(Zanthoxylum acanthopodium DC)*

Secondary metabolite compounds	Reagent	Result
Alkaloid	Bouchardart	-
	Maeyer	+
	Dragendroff	+
Steroida and	Salkowsky	-
Triterpenoid	Lieberman-Burchad	-
Saponin	Aquadest + Alcohol 96%	+
Flavonoiod	$Mg_{(s)}$ +n $HCI_{(p)}$	+
Tanin	$FeCI_3$ 1%	+
Glikosida	Mollish	+

Table 2. Average Length of Incision Wound Healing in Male White Rats Wistar Strain

Group Treatment	Size of wound diameter (mm)				
	Day 1	Day 3	Day 7	Day 11	Day 14
Group I	20.47	15.50	13.60	8.68	7.32
Group II	20.47	15.62	12.48	8.53	5.87
Group III	20.60	17.32	14.18	9.82	5.16
Group IV	21.68	17.30	12.97	9.78	3.44
Group V	21.10	19.60	15.77	12.28	6.08

Based on the statistics in the table 2, the shortest incision wound in group I that received NaCI treatment required an average healing time of 7.32mm or 14 days. The shortest incision wound in group II that received betadine therapy required an average healing time of 5.87mm or 14 days. The smallest average length of incision healing in group III treated with 1% ethanol extract of andaliman fruit was 5.16mm, or day 14. In group IV, treated with 3% ethanol extract of andaliman fruit, the smallest average length of wound healing was 3.44mm. In group V, treated with 5% ethanol extract of andaliman fruit, the smallest average length of wound healing was 6.08mm.

Table 3. Collagen Type III

Group I	Group II	Group III	Group IV	Group V
0.132	0.298	0.111	0.239	0.587
0	0.294	0.378	0.426	0.148
0	0.276	0.298	0.37	0.043
0.084	0.414	death	death	0.081
0.123	0.628	0.238	0.434	0
0.17	0.729	0.267	0.88	0.134

Based on table 3, it can be seen that the collagen type III content of each test group has significant variation among the groups. Group II showed collagen type III consistently high, with

values ranging from 0.298 to 0.729 in each group. Group IV had a significant variation, with some rats showing high levels. Still, some dead rats may indicate the non-detection of collagen type III in certain rats in this group. Group V had variable collagen type III; some rats showed low or undetectable levels. Groups I and III consistently showed lower levels when compared with Group II but higher levels with Group V.

The results of the Shapiro-Wilk normality test showed that the average data of the incision test group and the data on collagen type III in this study are normally distributed. Data is declared generally distributed if the significance value exceeds 0.05.

The results of the homogeneity test show that the average data of the incision test group and the data on collagen type III in this study obtained significant data. This indicates that the data obtained is homogeneous because the value (p> 0.05) indicates that the data obtained is homogeneous. Testing the distribution and variance of the data obtained normal results. If the variance is the same, then the data can be tested next using the One Way Anova parametric hypothesis test.

Based on using the One Way Anova parametric hypothesis test, there is a statistically significant difference (p<0.05) in the wound healing process of rats in each treatment group using andaliman ethanol extract, with a statistical significance value of 0.977. Further testing was carried out using the Levene test.

Based on the data from the levene test results, it is known that 5 treatment groups: group with NaCI administration, a group with betadine administration, a group with dose I (administration of 1% andaliman ethanol extract), a group with dose II (administration of 3% andaliman ethanol extract), group with dose III (administration of 5% andaliman ethanol extract), obtained a sig value of 0.988>0, 05 So it can be concluded that the group with NaCI administration, group with betadine administration, group with dose I (administration of 1% andaliman ethanol extract), group with dose II (administration of 3% andaliman ethanol extract), group with dose III (administration of 5% andaliman ethanol extract) have the same variant (homogeneous).

The One Way Anova parametric hypothesis test on collagen type III shows that the significance value is 0.012, so that (p <0.05), this value indicates that there is a significant difference between each group on collagen type III against incision wounds in white Wistar rats using andaliman ethanol extract.

The statistical test was continued using the LSD post hoc test because the data was normally distributed and homogeneous (Prabowo et al., 2021).

Table 4. Post Hoc LSD Variable Collagen Type III Test Results

Group	Comparison group	Mean difference	Significance
NaCI	Betadine[*b]	-.355000[*]	.001
(a)	Dose I	-.173567	.092
	Dose II[*d]	-.208967[*]	.045
	Dose III	-.080667	.401
Betadine	NaCI[*a]	.355000[*]	.001
(b)	Dose I	.181433	.079
	Dose II	.146033	.153
	Dose III[*e]	.274333[*]	.008
Dose I	NaCI	.173567	.092
(c)	Betadine	-.181433	.079
	Dose II	-.035400	.735
	Dose III	.092900	.357
Dose II	NaCI[*a]	.208967[*]	.045
(d)	Betadine	-.146033	.153
	Dose I	.035400	.735
	Dose III	.128300	.207
Dose III	NaCI	.080667	.401
(e)	Betadine[*b]	-.274333[*]	.008
	Dose I	-.092900	.357
	Dose II	-.128300	.207

Based on the post hoc test with LSD Variable collagen type III in table 4, it can be explained as follows: In group 1 (administration of NaCI), it can be seen that there is a significant comparison with group 2 (administration of betadine) and group IV (administration of 3% andaliman ethanol extract); this is because the considerable value <0.05. In group 2 (betadine administration), it can be seen that there is a significant comparison between group 1 (NaCI administration) and group 5 (5% andaliman ethanol extract administration). This is because the considerable value is <0.05. There is no significant comparison in group 3 (administration of 1% andaliman ethanol extract) because the significant value is> 0.05. In group 4 (administration of 3% andaliman ethanol extract), it can be seen that there is no considerable comparison with group 1 (administration of NaCI). This is due to the significant value of <0.05. In group 5 (administration of 5% andaliman ethanol extract), it can be seen that there is a significant comparison with group 2 (administration of betadine). This is due to the considerable value of <0.05.

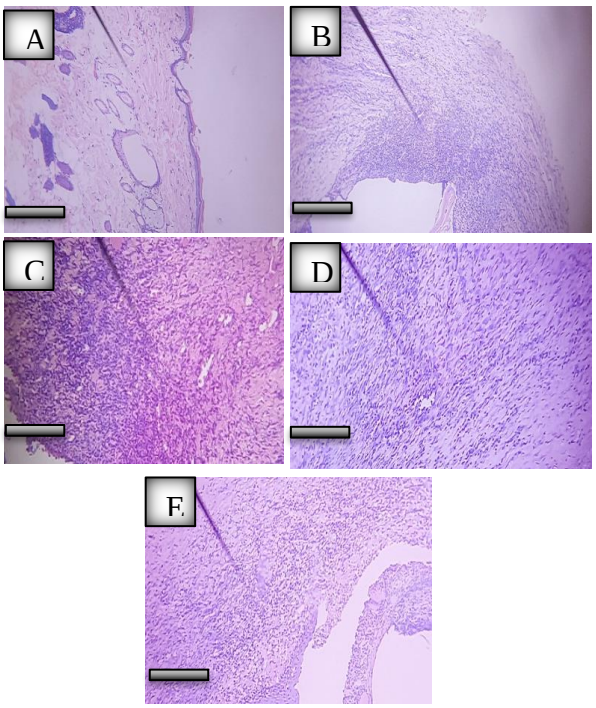

Figure 1. Representative images of the histological section of the five experimental groups.

It can be explained, A: Histopathology image of the skin in the group 1 (sNaCl). B: Histopathology image of the skin in group 2 (Betadine) C: Histopathology image of group 3 (1% andaliman ethanol extract). D: Histopathology image of group 4 (3% andaliman ethanol extract). E: Histopathology image of group 5 (5% andaliman ethanol extract). The black bar = 30µm

DISCUSSION

Based on the results of phytochemical tests on andaliman fruit extract (*Zanthoxylum acanthopodium DC*), it shows that there are Alkaloid compounds with Bouchardat reagent, which shows negative results, Maeyer reagent shows positive results, Dragendroff reagent shows positive results. In addition, there are steroidal and Triterpenoid compounds with the Salkowsky reagent, which shows negative results, and the Lieberman-Burchad reagent, which shows positive results. Saponin compounds with Aquadest + 96% alcohol reagent showed negative results. Tannin compounds with Mg (s) + HCI (p) reagent showed positive results, and Glycoside compounds with Mollish reagent showed positive results.

The andaliman fruit extract contains several active compounds, such as alkaloids, steroids, triterpenoids, tannins, and glycosides. The presence and type of these compounds can provide information about the bioactive potential of andaliman fruit extract, as well as indicate possible pharmacological effects or certain nutritional content it can have. This is in line with research conducted by (Muzafri, 2019), which states that andaliman extract contains alkaloid compounds, flavonoids, glycosides, saponins, tannins, triterpenes/steroids, and anthraquinone glycosides.

Based on the research, the test group with the fastest wound healing time is the test group with a treatment dose of andaliman ethanol extract of as much as 3%. On day 14, the average length of incision wound healing is 3.44mm. Andaliman has an antibacterial effect that helps wound healing and prevents secondary infections. Flavonoids, alkaloids, and tannins in andaliman can prevent the growth of pathogenic microorganisms such as Escherichia coli, Staphylococcus aureus, and Pseudomonas aeruginosa. The extract of andaliman can cause bacterial cells to undergo lysis by damaging the bacterial cell wall and increasing the permeability of the cell wall, resulting in leakage of bacterial intracellular metabolites (Xuliang & Ginting, 2022).

This study's results align with research conducted by Janice et al. (2023), which states that ethanol extract from Andaliman fruit can be formulated into a nano gel preparation with the best concentration of 20% as a wound healer. The nano gel of Andaliman fruit ethanol extract has a high level of stability. This finding has implications for the formulation and application of nanogels for topical delivery of Andaliman extract in various pharmaceutical and cosmetic applications.

This study's results indicate a significant difference in collagen type III in the healing of incision wounds in Wistar male white rats. In group 1 (administration of NaCI), it can be seen that there is a significant comparison with group 2 (administration of betadine) and group IV (administration of 3% andaliman ethanol extract). In group 2 (betadine administration), it can be seen that there is a significant comparison with group 1 (NaCI administration) and group 5 (5% andaliman ethanol extract administration). There was no considerable comparison in group 3 (administration of 1% andaliman ethanol extract). In group 4 (administration of 3% andaliman ethanol extract), it can be seen that there is a significant comparison with group 1 (administration of NaCI). In group 5 (5% andaliman ethanol extract administration), it can be seen that there is a significant comparison with

group 2 (betadine administration). This is because the considerable value <0.05.

Collagen plays a significant role in the wound-healing process. Its roles include hemostasis, interaction with platelets, interaction with fibronectin, increasing fluid exudation, increasing cellular components, increasing growth factors, and encouraging fibroplasia and sometimes epidermal proliferation (M Ricky et al., 2017).

This study's results align with research conducted by Rahman et al. (2021), which states that collagen type III also plays a role in wound healing. The mechanism of action of this collagen has yet to be discovered. However, this collagen decreases with age, which changes the skin's tension, elasticity, and healing. The lack of this collagen in mice causes more scarring than in mice that do not lack this collagen.

CONCLUSION

Overall, the 3% andaliman ethanol extract accelerates incision wound healing effects compared to negative control.

REFERENCES

Djuang, M. H., Syahputri, N. R., Silitonga, R., & Chiuman, L. (2022). Antimicrobial effectiveness of fruit extracts andaliman (Zanthoxylum Acanthopodium DC) AGAINST Staphylococcus epidermidis Bacteria. *Journal Health & Science: Gorontalo Journal Health and Science Community*, *6*(2), 68–75. https://doi.org/10.35971/gojhes.v5i3.13792

Handajani, F. (2021). *Metode pemilihan dan pembuatan hewan model beberapa penyakit pada penelitian eksperimental*. Sidoarjo: Zifatama Jawara.

Janice, J., Putri, R. E., & Fioni, F. (2023). Tests of Burn Wound Healing with Nanogel Preparations from Ethanol Extract of Andaliman Fruit. *Jurnal EduHealth*, *14*(04), 195–203. https://ejournal.seaninstitute.or.id/index.php/healt/article/view/3102

Ministry of Health RI. (2018). *Proporsi Jenis Cedera Jenis luka, Terkilir, Patah Tulang, Anggota Tubuh Terputus) Menurut Provinsi, Riskesdas 2018.* Jakarta.

M Ricky, R., Tri, U. S., Rizki, H., & Hanarisha, P. A. (2017). Pengaruh ekstrak metanol daun ketapang (Terminalia catappa L.) terhadap kepadatan serabut kolagen pada penyembuhan luka sayat mencit (Mus musculus). *Jurnal Agromedicine*, *4*(1), 17–24.

Makuszewska, M., Bonda, T., Cieślińska, M., Bialuk, I., Winnicka, M. M., & Niemczyk, K. (2020). Expression of collagen type III in healing tympanic membrane. *International Journal of Pediatric Otorhinolaryngology*, *136*, 110196. https://doi.org/10.1016/j.ijporl.2020.110196

Muzafri, A. (2019). Uji aktivitas antimikroba ekstrak Andaliman (Zanthoxylum acanthopodium DC.) pada Staphylococus aureus. *Jurnal Sungkai*, *7*(1), 122–126. https://doi.org/10.30606/js.v7i1.1742

Nabeela, S.A (2017). Efektifitas Gel Ekstrak Daun Binahong (Anredera Cordifolia (Ten) Steenis) Pada Luka Insisi Mencit (Mus Musculus) Melalui Pengamatan Panjang Area Luka Dan Penyembuhan Luka. *[Undergraduate thesis]. Surabaya: Universitas Airlangga*.

Nining, N. (2020). Pemanfaatan Kolagen Laut dalam Sistem Penghantaran Obat. *Majalah Farmasetika*, *5*(5), 245–256.

Nuari, S., Anam, S., & Khumaidi, A. (2017). Isolasi dan identifikasi senyawa flavonoid ekstrak etanol buah naga merah (Hylocereus polyrhizus (FAC Weber) Briton & Rose). *Jurnal Farmasi Galenika (Galenika Journal of Pharmacy)(e-Journal)*, *3*(2), 118–125. https://doi.org/10.22487/j24428744.0.v0.i0.8771

Nurihardiyanti, N. , W. A. , K. F. , & A. R. (2020). Uji Farmakologi Plester Patch Tanaman Toxic Jatropha Curcas L Untuk Penyembuhan Luka Sayat Dari Berbagai Literatur. *PROSIDING SENANTIAS*, *1*(1), 1373–1380. http://openjournal.unpam.ac.id/index.php/Senan/article/view/8488

Prabowo, Y. B., Ibrahim, N. P., & Saraswati, I. (2021). Pengaruh Variasi Waktu Perendaman dalam Ekstrak Daun Teh Hijau (Camellia sinensis) terhadap Stabilitas Dimensi Alginat. *E-GiGi*, *9*(1). https://doi.org/10.35790/eg.9.1.2021.32307

Purwanto, E., Tahir, T., & Syam, Y. (2022). Efek penggunaan ekstrak buah naga (Hylocereus

Sp) pada penyembuhan luka: Tinjauan sistematis. *NURSCOPE: Jurnal Penelitian dan Pemikiran Ilmiah Keperawatan*, *8*(1), 59-69. https://jurnal.unissula.ac.id/index.php/jnm/article/view/23135

Rahman, V. R., Bratadiredja, M. A., & Saptarini, N. M. (2021). Artikel Review: Potensi Kolagen sebagai Bahan Aktif Sediaan Farmasi. *Majalah Farmasetika*, *6*(3), 253–286. https://doi.org/10.24198/mfarmasetika.v6i3.33621

Siregar, B.L. 2003. Andaliman (Zanthoxylum acanthopodium DC.) di Sumatera Utara: Deskripsi dan Perkecambahan. *Hayati 10(1): 38-40*..

Tahir, T., Febrianti, N., Wahyuni, S., Syam, Y. (2019). Evaluation of acute wound healing potential of red dragon fruit *(Hylocereus Polyrhizus)* extract cream on type III collagen and Epidermal Growth Factor (EGF) levels: An animal study. *Elsevier Medicana Clinica. Volume 3.* https://doi.org/10.1016/j.mcpsp.2020.100091

Tanessa, M., Chiuman, L., & Kotsasi, F. (2023). Effectiveness Of Andaliman Extract Nanoemulsion (Zanthoxylum Acanthopodium Dc) Against Lipid Profile In Streptozotocin-Induced Wistar Male Rats (Stz). *Journal Health & Science: Gorontalo Journal Health and Science Community*, *7*(1), 27–34. https://doi.org/10.35971/gojhes.v7i1.17387

Xuliang, X., & Ginting, J. B. (2022). Analysis of wound healing from andaliman fruit essential oil ointment (zanthoxylum canthopodium dc.) on wistar rats (Rattus norvegicus). *Britain International of Exact Sciences (BIoEx) Journal*, *4*(1), 31–42. https://doi.org/10.33258/bioex.v4i1.542