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# EFFECTIVENESS OF WOUND TREATMENT WITH JUBIRKURILAH EXTRACTS IN RATS (RATTUS NORVEGICUS)

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#### **ABSTRACT**

The aim of this study was to identify the effectiveness of mixed extracts namely: dringo plant/jurangau (Acorus calamus), gambier/gambir (Uncaria), cassumunar ginger/kunyit bolai (Zingiber purpureum), betel leave/sirih (Piper betle linn), and black pepper/lada hitam (Piper nigrum) which is abbreviated as Jubirkurihlah toward the incision wound healing in rats (Rattus norvegicus). This research is quantitative research by using experimental research design. The samples were 45 of Rattus norvegicus in which 15 rats were treated by the extract of 15%, 15 rats were treated by the extract of 20%, and 15 rats were treated by the extract of 25%. The incision was as deep as 2 mm with 1 cm length on the back, then the wound was treated with Jubirkurihlah extract (15%, 20%, 25%) once a day and observed for infection mark until the wound healed. The results of the study found that there were no sign of infection on the three treatment groups, the average of wound healing time treated by the extract of 15% was 5.66 days, the extract of 20% was 3.84 days, and the extract of 25% was 3.43 days. The difference level in the length of wound healing time between the extract of 15% compared to the extract of 20% was found p value as much as 0.0049 (there was a significant difference level between the extract of 15% and the extract of 20%), the extract of 15% compared to the extract of 25% was found p value of 0.00165 (there was a significant difference level between the extract of 15% and the extract of 25%), the extract of 20% compared to the extract of 25% was found p value 0.318 at  $\alpha$  0.05 (there was no significant difference level between the extract of 20% and 25%). Conclusion: the extract of 25% was the most effective toward the wound healing process in Rattus norvegicus. Advanced research with the extract of 25% compared to Iodine and Betadine in Rattus norvegicus is required.

**KEYWORDS:** Jubirkurihlah extract, wound, treatment, *Rattus norvegicus*.

#### INTRODUCTION

Nowadays, understanding the wound healing involves three phases: "inflammation, proliferation, and maturation." Wound healing is a series of reactions and complex interactions between cells and "mediators." (George Broughton II *et al.* 2006).

Related research by Jean O. Latuheru (2013) concluded that there was an effect of betel leaves on the incision wound healing on rabbit skin (*Oryctolagus cuniculus*), the macroscopically results on the third day showed that there were the differences on rabbit skin which was given betel leaves and which was not given betel leaves. The wound that was given betel leaves got dried, the color was dark, the area of the wound began to diminish and the wound union occurred. On the seventh day, the wounds that were given betel leaves showed a black crust which was formed due to the sticking of the remaining betel leaves, and the length of the wound was shorter. The wounds that were not given betel leaves

looked reddish on the inside of the wound and the edges of the wound were still irregular. On the fourteenth day, the wound on the rabbit's skin had diminished. The wound that was given betel leaves showed that the wound had completely closed, while the wound that was not given betel leaves seemed smaller, but the wound with red color in the middle was seen.

The people tradition still using the ingredients of *Jubirkurihlah* which is a mixture of spices consisting of: dringo plant, gambier, spicy turmeric/cassumunar ginger, betel leaves, and black pepper for umbilical cord treatment is apparently faster in healing the newborn's umbilical cord, but it has not been scientifically proven if it is effective in wound healing of umbilical cord treatment. An incision of 2 mm and 1cm length on the back was made and then was treated with *Jubirkurihlah* extract once a day, then the signs of infection and the length of wound healing are observed. Assessment of wound area is a truly important parameter for predicting

and estimating the healing time (Smith KP, Zardiackas LD, Didlake RH, 1986; Flanagan M, 2003).

Further research to see the effect of *Jubirkurihlah* extract on the incision wound performed in experimental animal of *Rattus norvegicus* is required.

#### RESEARCH METHODS

This study used an experimental research design. The experiments were observation under artificial conditions where these conditions were created and regulated by the researcher (Nazir, M. 2011). Before the research was carried out, the first thing to do was made *Jubirkurihlah* 

extract in the following way: betel leaves and dringo plant were finely cut and aerated, gambier was finely ground in mortar, spicy turmeric and dringo plant were cut and finely sliced, and black pepper was finely ground.

The ingredients that had been cut, finely sliced and crushed, were put in a dark bottle with 96% of ethanol until the material was submerged, then the bottle was closed, left it for 5 days by stirred / shake / mixed it occasionally. After 5 days filtered, the result of filter (macerate) was thickened by using a tool "Rotary Evaporator" and repeated up to three times.



Figure 1: Rotary Evaporator equipment.

Furthermore, the treatment that began with anesthesia using chloroform by dropping on cotton as much as 20 drops was given after the experimental animals had adapted (7 days). Then, the cotton was inserted into the jar. After that, the experimental animals were inserted into the jar. After they got limb, then they were taken from the jar and conducted shaving on the backs of experimental animals.

Disinfection of the backs of the animals that had been shaved using alcohol swab was carried out, then incision on the backs of the animals with a depth of 2 mm and 1 cm length was conducted, then it was treated with *Jubirkurihlah* extract (15 rats with a concentration of 15%, 15 rats with a concentration of 20% and 15 rats with a concentration of 25%). The data were collected by

identifying and observing the effects of wound treatment by using *Jubirkurihlah* extract every day to see the signs of infection (red, swollen, pus) and the length of wound healing time.

The data were analyzed by univariate and bivariate. The hypothesis in this research was "There is an effect of *Jubirkurihlah* extract on the incision wound healing in rats (*Rattus norvegicus*)."

#### RESULT AND DISCUSSION

After incision wound care on the back of *rattus norvegicus* had been done once a day until it healed by using *jubirkurihlah* extract, the result and discussion were presented as follows:

Table 1: Identification Result of Infection Signs on the Cared Wound by using Jubirkurihlah Extract in Rats (*Rattus norvegicus*) in 2017.

	Infecti	%	
	Available	Unavailable	70
Concentration Extract 15%	0	15 rats	33.3
Concentration Extract 20%	0	15 rats	33.3
Concentration Extract 25%	0	15 rats	33.3
Total		45 rats	100

Among all treatment groups consisting of 45 rats, there were no infections signs found during the treatment

process, all extract concentrations used could really heal the incision wound administered on the rats. The

ingredients of the extract consisting of betel leaves, Uncaria, sweet flag, bangle, and black pepper could really heal the incision wound which were done to 45 rats. Betel leaves contained Kavakrol which was disinfectant and anti-fungus, so that it could be used as antiseptic medicine for bad breath and vaginal discharge. Other substances were eugenol and metal eugenol that could be used to relieve pain on teeth traditionally.

Betel leaves had distinctive aroma because of its content of essential oil. The high amount of eugenol determined the quality of the leaves. Terpene only functioned as spicy flavouring agent. The essential oil of betel leaves contained volatile oil (*betlephenol*), sesquiterpene, starch, diastase, sugar, tannin, and chavicol that had germicidal, antioxidant, and fungicide properties (Achyad, D.E. dan Rasyidah, R. 2000).

The research related to the research about astringent and haemostatic properties. A microbiologic test of uncaria leaves and twigs extract was done on several bacteria that caused in vitro diarrhoea. From the result of that research, the extract of uncaria leaves and twigs could inhibit the growth of bacteria causing diarrhoea. Traditionally, sweet flags were widely used as the medicine for stomach-ache and skin diseases (Zulfadli, 1989).

Sweet flags functioned as carminatives, spasmolytic, and diaphoretics which were useful for tranquilizers, stomach, gastrointestinal sedatives, spleen medicines, pain relievers, appetite stimulants, tonics, relieving inflammations, relieving nasal congestion, producing clearer voice, and antiseptic ingredients. Kinds of diseases that could be healed by using sweet flags were for instance swellings, scabies, ringworm, swollen spleen, cowpox, nosebleed, fever, and etc. (Prosiding Konferensi Nasional, 2006).

In India, sweet flags flour was used as anthelmintic and medical herbs. In medicine of native east, sweet flags were used to cure dyspepsia (a medicine for kids suffering diarrhoea), bronchitis, and lozenge (chewable medicines to relieve sore throat). Bangle (Zingiber montanum) was included as a beneficial plant, as rhizome plants in general. The benefits of bangle were supported by its components of constituting chemical compound which was mostly essential oil, in which it had positive effects for human body. Other compounds contained in bangle were for instance cineol, pinene and sesquiterpene, mineral, albumenoid, fat, bitter sap, and organic acids.

Black pepper contained antioxidants that detered the growth of bad bacteria, the chemical contents of black pepper were saponin, flavonoids, essential oil, chavicine, resin, albumen, amylum, piperine, piperiline, piperoleine, poparanine, piperonal, dihdrokarveol, caryophyllene oxide, kariptone, tran piocarrol, and pepper oil. The chemical properties of pepper were spicy and

distinctively flavoured. Flavonoid was a natural polyphenolic molecule that came from the plants known as antioxidant, anti-inflammatory, and anti-carcinogenic (Pinent M *et al.* 2008).

The trial using the combination of herbal plants could promote traditional medication to be more effective and gave significant proof from rats model application, particularly in terms of wound healing duration (T.W. Lau *et al.* 2008).

Table 2: Identification Result on Wound Healing Duration by Using Jubirkurihlah Extract in Rats (rattus Norvegicus) in 2017.

	Quick f %		S	Slow	Moon	
			f	%	Mean	
Concentrate	8	53.3	7	46.7	5.66	
extract 15%	0	33.3	/	40.7	days	
Concentrate	11	11 73.3	4	26.7	3.84	
extract 20%	1.1	73.3	4	20.7	days	
Concentrate	12	2 80	3	20	3.43	
extract 25%	12				days	

From the table 2 above, it could be concluded that in the group in which the wound was treated using 15% extract concentration on 15 rats, with the mean of the wound healing time needed of 5.66 days, there were 8 rats (53.5%) that experienced quick wound healing process (under the mean of wound healing time). In the group in which the wound was treated with 20% extract concentration with the mean of 3.84 days, there were 11 rats (73.3%) experienced quick wound healing process. And in the group in which the wound was treated using 25% extract concentration with the mean of 3.43 days, there were 12 rats (80%) that experienced quick wound healing process.

From the treatment result, it could be seen that the higher the concentration was, the faster the wound healing process in the rats would be, which were 15% concentration with the mean of 5.66 days, 20% concentration with the mean of 3.84 days, and 25% concentration with the mean of 3.43 days, there was only a little difference in the duration of wound healing between 20% concentration and 25% concentration which was 0.41 days, while the difference between 15% concentration and 20% concentration was only 1.82 days.

Wound healing was a truly complex and regulated process that could be compromised by endogenous factor (pathophysiologic) and exogenous factor (microorganism). The microbial colonization from acute and chronic wounds could not be avoided, and in most situations, endogenous bacteria was dominating, many among them have pathogenic potential in the wound environment (Bowler PG, 2002).

The rich herbal products such as phenolic, terpenoid, coumarin, glycoside, saponin, bitter principle, and carbohydrate compounds usually showed positive effects (Marles JR, Farnsworth NR, 1995; Tabatabaei-Malazy O, Larijani B, Abdollahi M, 2012; Li WL, 2004; Tanko Y *et al.*2007; Sherma RD, Sarkhar DK, Hazra MB, 2010; Sikarwar MS, Patil MB, 2010).

The type of wound in this research was sterile and not left open which was treated immediately using *Jubirkurihlah* extract in accord with the treatment group with 15%, 20%, and 25% concentration, it enabled the acceleration of wound healing process. The injury was done until the dermis layer which was the second layer of skin functioning as the supporter of epidermis layer. Dermis layer had the thickness around 0.25 mm up to 2.55 mm. The thickest layer structure was located in the area of palm and foot.

While the thinnest dermis layer structure was located in the area of eyelid skin, genital skin, and scrotum skin. The structure of dermis layer was composed of supporting tissue that consisted of white-coloured fibre and yellow-coloured fibre. The yellow fibre was elastic or flexible so that the skin could develop. In the structure of this dermis layer there were also sweat glands and hair roots. Hair roots were related to blood vessels carrying nutrients and oxygen, moreover they were also related to nerve fibres.

Table 3: The Statistics' Result of The Effectiveness of Wound Care in Rats (*Rattus norvegicus*) by Jubirkurihlah Extracts with concentration of 15% to 20% in 2017.

Concentration	Mean ± SD (day)	p Value
15%	$5.66 \pm 1.79$	0.00049
20%	$3.84 \pm 0.85$	0.00049

Based on the table 3, it showed the ratio between the concentration of 15% of the extract *Jubirkurihlah* and the concentration of 20%. On the extract of concentration of 15%, mean score of 5.66 was obtained, while the extract of concentration of 20%, mean score of 3.84 was obtained with p-value of 0.00049 at  $\alpha$  0.05. It meant that p value  $<\alpha$ . Thus, it could be concluded that there was a significant difference between extract with concentrations of 15% and that of 20% . It meant that the extract with concentration of 20% was more effective for the treatment or cure injuries than that of 15%.

Table 4: The Statistics' Result of The Effectiveness of Wound Care in Rat (*Rattus norvegicus*) by Jubirkurihlah Extracts with concentration of 15% and 25% in 2017.

Concentration	Mean $\pm$ SD (day)	p Value
15%	$5.66 \pm 1.79$	0.00165
25%	$3.43 \pm 0.90$	0.00163

Based on the table 4, it showed a comparison between extract with concentrations of 15% and that of 25%. The extract with concentration of 15% obtained mean score of 5.66, and the extract with concentration of 25% obtained mean score of 3.43 with p value of 0.00165 at  $\alpha$  0.05 which meant that p value <from  $\alpha$ . It could be concluded that there was a significant difference between the extract with concentrations of 15% and that of 25%. It meant that the extract with concentration of 25% was more effective for the treatment or cure injuries than that of 15%.

Table 5: The Statistics' Result of The Effectiveness of Wound Care in Rat (*Rattus norvegicus*) by *Jubirkurihlah* Extracts with concentration of 20% and 25% in 2017.

Concentration	Mean ± SD (day)	p Value
20%	$3.84 \pm 0.85$	0318
25%	$3.43 \pm 0.90$	0318

Based on the results of Table 5 above, it was obtained the comparison between extract with concentration of 20% and that of 25%, the extract with concentration of 20% obtained mean score of 3.84 and the extract with concentration of 25% obtained mean score of 3.43 with p value of 0.318 at  $\alpha$  0.05. It meant that p value >  $\alpha$ , and it was concluded that there was a significant difference between extract with concentrations of 20% and that of 25%. It indicated that the extract with concentration of 20% and that of 25% were equally effective for wound healing process and there was no difference in effect. Both of them could be used to heal with a difference mean only 0.41.

The basic ingredients of jubirkurihlah extract contained useful compound to accelerate wound healing time. By the presence of flavonoid compound that acted as an anti-bacterial and saponins in a betel leaves, it contained a protein structure that could stimulate the formation of collagen. Thus, it could be used in healing process and antibacterial properties. Moreover, the other base material extract (jurangau, Gambier, Bolai turmeric and black pepper) also had similar benefit.

#### CONCLUSION AND SUGGESTION

The conclusion of the research is that the existence of infection signs (aflame, swollen, and pus) cannot be found on the incision wound of the three treatment group, the average of time duration of wound healing in the treatment group by using *Jubirkurihlah* extract with concentration 15% of was 5,66 days, with concentration of 20% was 3,84 days and with concentration of 25% was 3,43 days.

The contrast level of time duration of wound healing between 15% concentration extract was compared with 20% was found 0,00049 value p and can be concluded the existence of significant contrast between 15% extract with 20% which mean that extract with 20% concentrate

was more effective to heal incision wound compared with 15% concentration.

The level of difference in the length of time the healing wound process between extract with concentration of 15% and that of 25% obtained p value of 0,00165. It can be concluded that extract with concentration of 15% and that of 25% is significantly difference. It meant that extract with concentration of 25% is more effective to heal incision wound than that of 15%.

The level of difference in the length of time the healing wound process between extract with concentration of 20% and that of 25% obtained p value of 0,318 at  $\alpha$  0.05. It can be concluded that extract with concentration of 20% and that of 25% is not significantly difference. It meant that extract with concentration of 20% and that of 25% are equally effective in incision wound healing process and there was no difference effect, equally can heal incision wound on rats (*Rattus Norvegicus*).

It is required further research by giving treatment to the three treatment group as follows: the treatment 1 was wound treatment using *Jubirkurihlah* extract with concentration of 25% since it is more effective in incision wound healing on rats than that of 15% and 20%, the treatment 2 is wound treatment using iodine and the treatment 3 is wound treatment using butadiene with control group.

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