Formulation of Cassava (*Manihot esculenta* Crantz) Tuber Extract Gel and Effectiveness Test on Burns in Rabbit (*Oryctolagus cuniculus*)

Formulasi Gel Ekstrak Umbi Singkong (*Manihot esculenta* Crantz) dan Uji Efektivitas terhadap Luka Bakar pada Kelinci (*Oryctolagus cuniculus*)

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ABSTRACT

Burn is heat transfer in which the source can be direct or indirect contact with fire, electricity, friction chemicals or radiation. It can cause damage of skin tissue even in a state of multisystem injury leading to serious disorders. Systemic effects and mortality caused by burn is generally determined by the extent and depth of affected skin. Cassava (*Manihot esculenta* Crantz) contains carbohydrates, phosphorus, calcium, vitamin C, vitamin B1, protein and iron. Furthermore, substances contained in cassava are able to give effect to the re-epithelialization of skin damaged cell tissue due to burns. The aim of this study was to find out the cassava tuber extract in gel preparation formulation and the effectiveness of burn healing in rabbits. This study was an experimental study to examine the concentration of cassava tuber extract gel formula which is the most effective in healing superficial burns of degree 4 in male rabbits. From the observations of giving gel preparations once a day by using positive control comparison (Bioplaceton) could be concluded that the extract of cassava provided healing which was not significantly different from the positive control.

Keywords: burns, cassava, gel, rabbit

ABSTRAK

Luka bakar merupakan peristiwa perpindahan panas, yang sumber panasnya dapat bervariasi seperti kontak langsung atau tidak langsung dengan api, listrik, bahan kimia gesekan atau radiasi. Akibat yang ditimbulkan berupa kerusakan jaringan kulit, bahkan pada keadaan cedera multisistemik dapat menyebabkan gangguan yang serius. Efek sistemik dan mortalitas yang disebabkan karena luka bakar sangat ditentukan oleh luas dan dalamnya kulit yang terkena luka. Tanaman singkong (Manihot esculenta Crantz) memiliki kandungan karbohidrat, protein, fosfor, kalsium, vitamin C, vitamin B1, dan zat besi. Kandungan yang terdapat dalam singkong mampu memberikan efek epitelisasi kembali kulit yang mengalami kerusakan sel kulit akibat luka bakar. Tujuan dari penelitian ini adalah untuk mengetahui ekstrak umbi singkong dalam formulasi sediaan gel dan efektivitas penyembuhan luka bakar pada kelinci. Penelitian ini merupakan penelitian eksperimental untuk mengetahui konsentrasi formula gel ekstrak umbi singkong yang paling efektif dalam penyembuhan luka bakar superfisial derajat 4 pada kelinci jantan. Dari pengamatan pemberian sediaan gel sekali sehari dengan menggunakan perbandingan kontrol positif (Bioplaceton) dapat disimpulkan bahwa ekstrak singkong memberikan kesembuhan yang tidak berbeda nyata dengan kontrol positif.

Kata kunci: gel, kelinci, luka bakar, umbi singkong

Introduction

The skin as the outermost defense of the body is more susceptible to damage like burns. Burn can be dangerous to the tissue that does not only occur on the surface of the skin, but can occur underneath the skin. Burned tissue is even damaged causing body fluids to leak out through the capillaries of the blood vessels in the tissue that is swollen due to burns. In large burns, you will experience a large amount of fluid loss due to leakage of fluid from the skin, causing shock (Rahmawati *et al.*, 2018).

Burn is usually expressed as a degree determined by the depth of the burn. The severity of the wound depends on the depth, extent and location of the wound. The age and health of the previous sufferer also influences the prognosis. The depth of the burn is determined by the high temperature (Mz A, 2017). Therefore, burn treatment plays an important role in the wound healing process. Wound healing is a form of an effort to repair damage that occurs to the skin. The physiology of wound healing naturally goes through several phases, namely the hemostasis phase, the inflammatory phase, the proliferation phase and the maturation phase.

One of the herbal plants used for healing burns is cassava tubers. Cassava is one of the most widely grown plants in Indonesia, and is a plant that contains nutrients in the form of carbohydrates, phosphorus, calcium, vitamin C, protein, iron and vitamin B1. Besides having high nutritional content, cassava tubers also contain flavonoid compounds and saponins. The content of flavonoids and saponins in plants can be used as antiinflammatory and antibacterial (Nugroho, 2013). The administration of cassava root extract has an effect on increasing the thickness of the granulation tissue in the treatment of fourth degree burns in male rabbits. Therefore, it is necessary to conduct research on carbopol bases with different concentration variations, namely 0.5%, 1%, and 2% to determine the most stable concentration, then test the effectiveness of the most stable gel against grade IV burns in male rabbits.

Method

This study used experimental approach which aims to determine a symptom or effect that arises as a result of certain actions.

Instruments and Materials

The instruments used in this study were simplicia sieve, glass tools, homogenezer, freeze dryer (Scanvac[®]), Climatic chamber (MMM[®]), analytical scales (Matrix[®]), Viscometer Brookfield (LV[®]), a set of gel evaluation tools, extraction kit, shaving razor, heatinducing metal. The materials used in this study were cassava tubers, carbopol 940, methyl paraben, propylenglycol, aquadest, 70% ethanol and male rabbits.

Experiments

1. Sampling

The samples were cassava tubers, which were obtained from Bitingan Market.

2. Population

The study samples used were cassava tubers and male rabbits.

3. Sample processing

The collected cassava tubers were sorted wet to separate other parts of the plant which were not used and then washed with clean running water. Then conducting the chopping by means of the sample cut into small pieces, after that it was dried by airing it out of direct sunlight, then carried out by the maceration extraction process.

4. Preparation of cassava ethanol extract

The extract of cassava roots using the extraction method by maceration and using 70% ethanol as a solvent. The simplicia is weighed as much as 500 grams then macerated with 70% ethanol until the sample is submerged then allowed to stand for x 24 hours while stirring 3 occasionally. The phytrate was filtered, the waste was extracted again using 70% ethanol solvent. The liquid extract obtained is evaporated until it gets a thick extract, then freezer drying it until the dry extract is obtained.

5. Selection and preparation of experimental animals

The animals used were healthy male rabbits, not defective, white, weighing 1.5-2.0 kg, 4-10 months old. The number of rabbits used is three. Before being treated with experimental animals, rabbits must be adapted to their environment.

6. Testing of burn healing effects

The experimental animal (rabbit) was shaved on its back for 3 parts and induced with a heat induction device in the form of a metal plate (3 x 3 cm) for 1 minute. Each test animal was given 3 parts of the wound. Wound I was positive control (Bioplasenton[®] gel), wound II (stable gel of cassava tuber extract, formulation in Table 1) and wound III was negative control (gel base). Apply this once a day. Every day the experimental animals were observed for 21 days.

Data Analysis

Wound healing data were analyzed statistically using One way ANOVA (Analysis of Variants) and physical evaluation of the gel preparations were analyzed descriptively.

Results and Discussion

Crude drugs of 500 g of refined cassava tubers are soaked using 70% ethanol. Extraction uses the maceration method which is a cold extraction method that is suitable for cassava tubers to maintain the contents in the cassava tubers which are easily damaged by heat and allow all simplicia to come into contact with the pollen liquid. Ethanol 70% is used as a solvent because it can dissolve almost all substances, both polar, semipolar and non-polar, such as flavonoids, tannin alkaloids and saponins.

The physical test results of the gel preparations can be seen in Table 2. It appears that the cassava tuber extract gel preparations in formula II and formula III meet the requirements to assess changes in color, odor, and appearance of the preparation. Observation of the homogeneity test results showed that the gel preparation was physically homogeneous and there was no syneresis phenomenon, which indicated that the ingredients in the gel were completely dissolved and mixed.

Materials	C	v)	
	F1	F2	F3
Extract cassava root	8	8	8
Carbopol 940	0,5	1	2
TEA	Ad pH 6	Ad pH 6	Ad pH 6
Metil paraben	0,25	0,25	0,25
Propilenglikol	10	10	10
Aquadest ad	100	100	100

Table 1. Cassava root extract gel formula

Table	2.	Organoleptic	test	results	of	cassava	tuber	extract	gel	before	and	after
accelerated storage												

Aspects	Formula	Before	After		
Color		Yellow	Yellow		
	П	Yellow	Yellow		
	111	Yellow	Yellow		
Smell	I	Extra typical	Extra typical		
	П	Extra typical	Extra typical		
	III	Extra typical	Extra typical		
Consistency	I	Liquid	Liquid		
	Ш	Gel form	Gel form		
	III	Gel form	Gel form		

pH test results showed that the pH before and after storage under accelerated conditions showed a change in pH. The results obtained indicate that formula III has optimal stability, because when compared to formula I and formula II, the change in pH of formula III is not too significant. The pH test results show that the pH criteria for topical preparations are in the pH range 4.5-6.5 (Maulina and Sugihartini, 2015). In the sense that formula III is still within the normal pH range of the skin so that when used will increase the comfort of the skin affected by burns.

From the results of the observations obtained, further research

was carried out for burns using male rabbits. The gel preparation used was formula III with a concentration of 2% as the most stable gel preparation. The results of the burn healing effect test showed a difference in wound healing time from the first day to the 21st day (Table 3). Activity testing was carried out on rabbits by making 3 burns on the rabbit's back. Based on the observation on the wound, the wound belongs to the group IV degree burns that have reached the muscle layer, tendons, and bone with extensive damage. The damage covers the entire dermis layer, skin organs such as hair follicles, sebaceous glands, and sweat glands.

Day	Control positive			Control negative			Stable gel 2%			
	(Bioplaceton)			(base)			(Formula III)			
	1	2	3	1	2	3	1	2	3	
1	3,00	3,00	3,00	3,00	3,00	3,00	3,00	3,00	3,00	
2	2,50	2,41	2,33	2,51	2,57	2,43	2,49	2,52	2,35	
3	2,38	2,29	2,21	2,48	2,46	2,30	2,40	2,31	2,31	
4	2,27	2,29	2,12	2,48	2,33	2,37	2,31	2,31	2,29	
5	2,21	2,02	2,01	2,43	2,29	2,25	2,27	2,20	2,19	
6	2,17	1,96	1,71	2,42	2,24	2,20	2,23	2,15	1,83	
7	1,74	1,95	1,61	2,40	2,20	2,20	2,11	2,14	1,79	
8	1,63	1,61	1,37	2,29	2,14	1,54	2,04	2,16	1,64	
9	1,63	1,54	1,37	2,22	1,98	1,54	1,93	2,03	1,63	
10	1,43	1,47	1,28	2,19	1,93	1,54	1,93	1,98	1,63	
11	1,31	1,42	1,21	2,14	1,89	1,49	1,93	1,69	1,41	
12	1,16	1,31	1,13	1,63	1,63	1,24	1,44	1,50	1,31	
13	1,13	0,84	1,08	1.32	1,17	1,23	1,30	1,15	1,17	
14	1,05	0,96	0,99	1,22	1,19	1,13	1,20	1,16	1,11	
15	0,93	0,83	0,96	1,19	1,16	1,09	1,10	1,13	0,86	
16	0,80	0,81	0,88	1,07	1,15	1,04	0,99	1,05	0,86	
17	0,63	0,79	0,68	0,89	1,09	1,02	0,73	1,06	0,73	
18	0,30	0,56	0,44	0,64	0,91	0,71	0,44	0,84	0,45	

Table 3. Measurement results diameter (cm) burns on male rabbits

Based on the literature review, it is explained that the healing time for superficial partial thickness burns can heal in approximately 14-21 days (Sentat and Permatasari, 2015). In this study, burns that were given wound care using cassava tuber extract using carbopol formula III gelling agent with a concentration of 2% experienced faster healing than bioplaceton. The acceleration of burn healing in the cassava tuber extract group with formula III occurred because the cassava root extract contained three secondary metabolites, namely flavonoids, tannins and saponins. The anti-inflammatory activity of flavonoids through cyclooxygenase and lipoxygenase inhibition causes a limitation of the number of inflammatory cells migrating to the injured tissue so that the inflammatory reaction will last a shorter period and the proliferative ability of TGF-B is not inhibited, besides that flavonoid as a reduction in inflammatory mediators can accelerate the decrease in the degree of erythema through a mechanism. inhibition of arachidonic acid release. Tannins can affect wound with cellular healing several mechanisms, including increasing the reformation of dermis tissue in wound tissue (fibroplasia) (Negara, 2014). in research (Megawati et al., 2020), showed that the results of statistical tests had a significant effect on wound healing in male rabbits and showed that ethanol extract with a concentration of 60% had the best effect in accelerating the healing process of cuts.

Based on the existing research on cassava leaves, it can be seen the difference in wound healing time cut in male rabbits using a concentration of 60%. The next analysis used a one-way analysis of variance method to determine whether there was a significant difference or not in each treatment group. From the One-Way ANOVA test results which will show a significant difference in the experimental group.

Based on the results using the analysis method used was SPSS 17. Analysis was carried out on the results of changes in wound size from day 0 to day 21. In this study, an analysis of changes in burn size was used to see whether there was a difference between the three treatment groups on one of the parameters of burn healing in rabbits. Based on the results of the data analysis test, it was obtained that the cassava tuber extract gel treatment data was not significantly different from the positive control but very significantly different from the negative control, which means that the cassava tuber extract gel is thought to heal burns the same as the positive control

Conclusion

Based on the results of the research, it can be concluded that the optimal carbopol-based cassava tuber extract gel is Formula III with a concentration of 2% based on the organoleptic test, homogeneity test, pH test, stickiness test, spreadability test, viscosity test, and accelerate test (accelerated storage). The cassava root extract gel provides healing that is not significantly different from the positive control.

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