

Effectiveness of Aloe Vera Compress on Wound Healing Time in Mice (*Mus Musculus*)

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Abstract: Postpartum infection and hemorrhage are the leading causes of maternal death in Indonesia. One of the causes is skin lacerations or birth canal tissue (10%). A study by Agnes (2009) found out that (57.8%) postpartum women suffered failed episiotomy stitches due to poor wound care. One method of conventional wound care is to apply Aloe vera compress. The objective of the study was to analyze the effectiveness of Aloe vera compress on the wound healing time (trials in mice). This research used Quasi experimental design involving 15 mice as the samples which were divided into 3 groups. The first group was the control group, the second group received Povidone Iodine compress, and the third group received Aloe vera compress. Total sampling technique was applied and REEDA mark observation sheet was administered to collect the data. The results showed that the wound healing time in mice given Povidone Iodine compresses was 6.86 ± 0.900 days. The wound healing time in mice given Aloe vera was 8.13 ± 0.835 days. There is a significant difference in wound healing time in mice given Povidone Iodine compress and Aloe vera compress with a t-count value of 2.831 (p-value = 0.014).

Keywords: Aloe vera Compress, Healing Wounds Time

I. Introduction

Wound healing is a process of replacing dead or damaged tissue with new and healthy tissue by the body through degeneration. Infection can slow wound healing, increase granulation, and form scarring. To prevent postpartum infections, midwives should be responsive to recognize early signs of infection such as "REEDA" marks on skin lesions or the perineal tissue of the mother. A study conducted by Rejeki (2009) discovered that of 53 post partum women signs of REEDA (*Redness, Edema, Echimosis, Discharge and Approximation*) emerged in low educated mothers (47.7%), age 20-35 years (92, 2%), had no history of disease (96%), primipara (72%), episiotomy (64%), spontaneous rupture (36%), wound type ≥ 1 cm (64%), poor wound suture condition (54, 3%), LILA ≤ 23 cm (63.2%), and anemia (77.3%). Perineal wound healing can work well if the postpartum woman is aware of factors affecting wound healing including wound care; nutritional status; knowledge; personal hygiene; circulation; oxygenation; and maternal habits such as smoking, age, community tradition, and etc. Agnes (2009) mentioned that of 14 postpartum women 57.8% suffered perineal stitches breakdown due to less skin care or tissue injuries after defecating, skin wounds or moist tissue, or less care of panties use. In addition, the belief of avoiding foods containing protein such as; eggs, fish, and meat due to the fishy smell of puerperal blood in urine have caused the delayed healing. Therefore, proper wound care is very influential on wound healing.

Skin or tissue treatment can be instituted through modern medicine as well as conventional treatment. Conventional medicine is commonly referred to as herbal or traditional one. Skin or tissue care in the traditional way is more practical, easily accepted, and a lot cheaper to be incurred. The use of Aloe vera is one of the examples of traditional practices which is believed to be able to heal wound. Although still unfamiliar in Indonesia, the use of Aloe vera has been used by the Samaritans in about 1875 BC, the Ancient Egyptians in 1500 BC, and the treatment with Aloe vera is also recorded in historical documents of Arabic, Roman, Grece, Indian and Chinese medicine (Hartawan, 2012: 55).

Several studies have revealed the role of Aloe vera in the medical practices. A study by the University of Maryland Medical Center, as cited in Hartawan (2012: 59), found that treating Aloe vera gel to skin irritation due to scratches, burns, cuts, insect bites and rashes, was nine days faster than without any treatment. A similar study by herbalist Steven Foster, author of National Geograph Desk Reference to Nature's Medicine discovered the properties of Aloe vera gel that can speed healing by reducing pain and inflammation. Another study at Hoshi University Japan found that Aloe vera contained antioxidant compounds which were very useful to get rid of free radicals and protect the two components of natural wound healing in the body, superoxide dismutase (antioxidant enzymes) and glutathione. The later is an amino acid that stimulates the immune system (Hartawan, 2012: 63). Aloe vera contains 72 substances the human body needs. Of the 72 substances needed by the human body Aloe vera contains 18 kinds of amino acids, carbohydrates, fats, water, vitamins, minerals, enzymes, hormones, and drug classes such as antibiotics, antiseptic, antibacterial, anti cancer, antiviral, anti fungi, anti-infective and anti-inflammatory. Anti-bacterial substances and anti-fungal substances can increase blood flow to

the injured area and can stimulate fibroblasts, skin cells responsible for wound healing. Anti-bacterial and anti-fungal substances can be found in clear liquids such as jelly obtained by splitting the Aloe vera rod. Antiseptics and antibiotics contained in the aloin serve as a natural wound cleanser and treat wounds quickly. Aloe vera also contains lignin compounds, and polysaccharides useful as carrier medium nutrients needed by the skin. The characteristics of Aloe vera that has a normal acidity (pH), almost like a human pH can provide the ability to penetrate the skin properly. Amino acids and enzymes in Aloe vera serve to foster the development of new cells at extraordinary speeds and remove dead cells from the epidermis (Jatnika and Saptorningsih, 2010: 56).

In addition to the properties of compounds contained in Aloe vera, wound care using Aloe vera proved more effective than dry gauze or moist gauze to increase the power of wound links. A research by Danu Mahandaru and Ishandono Dachlan (2009), *The Effect of Aloe Vera on Healing Process of Incision Wound*, demonstrated that the group treated with dry gauze had wound link strength ($35 \pm 7 \text{ N / cm}^2$), with moist gauze ($41 \pm 7 \text{ N / cm}^2$), and with Aloe vera ($68 \pm 17 \text{ N / cm}^2$). There were significant differences in the strength of wound closure among the three groups ($p > 0.001$). In addition to providing several benefits, Aloe vera also has several side effects. Some cases have been reported that people experienced abdominal cramps and diarrhea after consuming Aloe vera due to its lactating effects. However, on topical use no significant side effects have been reported. Furthermore, according to the National Toxicology Program (NTP), cited in Hartawan (2012: 64), in general no research has reported the side effects of using Aloe vera.

It can be concluded that Aloe vera acts as a mechanism to promote an active biologic agent that heals wound tissue. In the inflammatory phase, Aloe vera acts as an anti-inflammatory and pain reliever so that the wound inflammatory phase is short. Thus it will accelerate the next phase of the proliferation. In this phase the various content of Aloe vera plays a role to stimulate fibroblasts to produce collagen and proteoglycans. These two components in Aloe vera increase the strength of the inter-wound closure so that the wound healing process runs faster (Plaskett, 2000). Following the steps of the previous studies on the use of Aloe vera in medical practices, this study is meant to discover more on the effectiveness of Aloe vera compress towards the wound healing time.

General Purpose of Research

To analyze the effectiveness of Aloe vera compress towards wound healing time in mice (*Mus musculus*)

Specific Purpose of Research

1. To identify the wound healing time by Iodine Povidone compress in mice (*Mus musculus*).
2. To identify the wound healing time by Aloe vera compress in mice (*Mus musculus*).
3. To analyze the effectiveness of Iodine Povidone compress and Aloe vera compress towards wound healing time in mice (*Mus musculus*).

II. Research Method

Design

The research deployed Quasi-experimental design as part of experimental research using control group. Yet, the control group can not primarily function to control influencing external variables (Nursalam, 2008:50). One group took care of skin or tissue with Aloe vera compress from day 1 of post partum until skin tissue is well recovered. While another group did not (only dry pads used) with the same range of time. The researchers, then, observed the recovery process of the wound to identify the healing process of skin tissue. The design can be described as follows:

Subject	Treatment	Observation	Healing Time
K-A	I	O	IO-A
K-B	-	O	O-B

Note:

- K-A : Subject of Treatment
- K-B : Subject of Control
- I : Compressing with *Aloe Vera*
- : Compressing with *Iodine Povidone*
- O : Observing the healing process of the tissue injury on the subjects of treatment and control subjects
- IO-A : Wound healing time of skin or tissue by Aloe vera compress.
- O-B : Wound healing time of skin or tissue by Iodine Povidone compress

Image (1): Research Design

Technique of Sampling

In the research, the technique of sampling was carried out through random sampling in which sample collection was conducted randomly without considering the strata in the population, in other words, each member or population unit have equal opportunity for selection (Notoadmojo, 2010:120).

Research Variables and Operational Definition

Table (1) Operational Definition of Effectiveness of Aloe vera Compress towards Wound Healing Time

Variables	Operatioal Definition	Indicator	Measurement Tool	Data Scale
Wound healing time by Aloe vera compress	The time required for wound recovery with Aloe vera compress marked with the loss of the REED mark and with the Approximation of both dry and Sub Cutaneous wound.	No sign of REED infection (Redness, Echymosis, Edema, Discharge) and Approximation is well-formed	Observation Sheet Standard Operating Procedure (SOP)	Interval
Wound healing time by Iodine Povidone compress	The time required for wound recovery performed with the morning and afternoon Iodine Povidone compresses marked with the loss of the REED mark and with the good Approximation and dry the Sub Cutaneous wound.	No sign of REED infection (Redness, Echymosis, Edema, Discharge) and Approximation is well formed	Observation Sheet Standard Operating Procedure (SOP)	Interval

Research Hypothesis

H1 : A difference rises in wound healing time between the Povidone Iodine compress and Aloe vera compress.

Experiment Unit and Replication

Mice (*Mus musculus*) taken from the Laboratory of Dentistry Faculty of Jember University were involved as the experimental unit. The selected mice (*Mus musculus*) meet the following requirements:

1. Age of animals treated 8-10 weeks
2. Pregnant
3. Healthy; characterized with active movement

The amount of sample replication is determined by the Cochran formula (1986). The determination is described as follows:

$$(t - 1) (r - 1) \geq 20$$

$$(10 - 1) (r - 1) \geq 20$$

$$9 (r - 1) \geq 20$$

$$9r - 9 \geq 20$$

$$9r \geq 29$$

$$r \geq 3,2$$

Based on the calculation results the numbers of samples (10 mice) were divided into 2 groups each of which consist of 5 experimental animals. The identification of mice (*Mus musculus*) for group sample was taken completely at random.

Data Analysis

In the study, the data analysis was used to test the effectiveness of Aloe vera compress and Iodine Povidone compress towards wound healing time in mice (*Mus musculus*) with independent t-test of 2 samples with 5% error rate. This technique was used to test the average equality of 2 independent populations which means one population is not affected or associated with other populations. In the selection of t-test formula, some considerations were taken to ensure of whether or not the number of samples were the same and the data variants of 2 samples were homogeneous. Thus, it is necessary to test homogeneity in advance.

Research Ethics

This research has been tested and gets recommendation of "Etical Clereance" from Health Research Ethics Commission, State Health Polytechnic of Malang, Republic of Indonesia, with the number of registration: 217/KEPK-POLKESMA/2016.

III. Result

Table (1) Identification of wound healing process based on the day of no redness in mice with Iodine Povidone and Aloe vera compress

			Not Redness				Total
			3	4	5	6	
Treatment	Iodine Povidone	Count	2	1	4	0	7
		% within Treatment	28,6%	14,3%	57,1%	,0%	100,0%
	Aloe vera	Count	1	2	3	2	8
		% within Treatment	12,5%	25,0%	37,5%	25,0%	100,0%
Total		Count	3	3	7	2	15
		% within Treatment	20,0%	20,0%	46,7%	13,3%	100,0%

Based on Table (1) mice given Iodine Povidone compress indicated no redness on the 5th day of wound (57.1%), while mice given Aloe vera compress indicated no redness on the 5th day (37.5%).

Table (2) Identification of wound recovery process based on the day of no Echimosis in mice with Iodine Povidone and Aloe vera compress

			Not Echimosis		Total
			1	2	
Treatment	Iodine Povidone	Count	6	1	7
		% within Treatment	85,7%	14,3%	100,0%
	Aloe vera	Count	4	4	8
		% within Treatment	50,0%	50,0%	100,0%
Total		Count	10	5	15
		% within Treatment	66,7%	33,3%	100,0%

Based on Table (2) mice given Iodine Povidone compress indicated no Echimosis on the 1st day of wound (85,7%), while mice with Aloe vera compress indicated no Echimosis on the 1st and 2nd day (50.0% each).

Table (3) Identification of wound healing process based on the day of no edema in mice with Iodine Povidone and Aloe vera compress

			Not Edema				Total
			3	4	5	6	
Treatment	Iodine Povidone	Count	1	4	2	0	7
		% within Treatment	14,3%	57,1%	28,6%	,0%	100,0%
	Aloe vera	Count	0	3	2	3	8
		% within Treatment	,0%	37,5%	25,0%	37,5%	100,0%
Total		Count	1	7	4	3	15
		% within Treatment	6,7%	46,7%	26,7%	20,0%	100,0%

Based on Table (3) mice given Iodine Povidone compress indicated no edema at 4th day of wound (57,1%), while mice with Aloe vera compress indicated no Edema on Day 4 and 6 (Respectively 37.5%).

Table (4) Identification of wound healing process based on the day of no Discharge in mice with compress Iodine Povidone and Aloe vera

			Not Discharge		Total
			3	4	
Treatment	Iodine Povidone	Count	2	5	7
		% within Treatment	28,6%	71,4%	100,0%
	Aloe vera	Count	3	5	8
		% within Treatment	37,5%	62,5%	100,0%
Total		Count	5	10	15
		% within Treatment	33,3%	66,7%	100,0%

Based on Table (4) mice given Iodine Povidone compress indicated no Discharge at 4th day wound (71,4%), and mice given Aloe vera compress also indicated no Discharge on day 4 (62,5 %).

Table (5) Identification of wound healing process based on the good Approximation day of wound on mice with Iodine Povidone and Aloe vera compresses

			Approximation Good				Total
			6	7	8	9	
Treatment	Iodine Povidone	Count	2	3	2	0	7
		% within Treatment	28,6%	42,9%	28,6%	,0%	100,0%
	Aloe vera	Count	1	1	3	3	8
		% within Treatment	12,5%	12,5%	37,5%	37,5%	100,0%
Total		Count	3	4	5	3	15
		% within Treatment	20,0%	26,7%	33,3%	20,0%	100,0%

Based on Table (5) for mice given Iodine Povidone good Approximation of wound indicated on day 7 (42,9%), while for mice given compress Aloe vera good Approximation indicated on day 8 and 9 (Respectively 37.5%).

Table (6) Identification of wound healing time in mice with Iodine Povidone and Aloe vera compresses

			Wound Healing				Total
			6	7	8	9	
Treatment	Iodine Povidone	Count	3	2	2	0	7
		% within Treatment	42,9%	28,6%	28,6%	,0%	100,0%
	Aloe vera	Count	0	2	3	3	8
		% within Treatment	,0%	25,0%	37,5%	37,5%	100,0%
Total		Count	3	4	5	3	15
		% within Treatment	20,0%	26,7%	33,3%	20,0%	100,0%

Based on table (6) for mice given Iodine Povidone compress wound healing time indicated on day 6 (42,9%), whereas for mice given old Aloe vera compresses wound healing time indicated on day 8 and 9 (Each at 37.5%).

Table (7) Average of wound healing time in mice with Iodine Povidone and Aloe vera compresses

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Wound Healing	Iodine Povidone	7	6,86	,900	,340
	Aloe vera	8	8,13	,835	,295

Table (7) shows that the average of wound healing time in mice given Iodine Povidone compresses is for 6.86 ± 0.900 days, whereas in mice given Aloe vera is 8.13 ± 0.835 days.

Table (8) effectiveness of wound healing time in mice with Iodine Povidone and Aloe vera compresses

		Levene's Test for Equality of Variances		t-test for Equality of Means		
		F	Sig.	t	df	Sig. (2-tailed)
Wound Healing	Equal variances assumed	,119	,736	-2,831	13	,014
	Equal variances not assumed			-2,816	12,407	,015

Based on table (8), the test of the effectiveness of wound healing time in mice given compress Iodine Povidone and Aloe vera compress obtained t-count value of 2.831 with significance of 0.014. The significant value of 0.014 is smaller than α (0.05). This suggests that a difference rises in wound healing time in mice between Betadin compress and Aloe vera compress.

IV. Discussion

The average wound healing time in mice given Iodine Povidone compress was for 6.86 ± 0.900 days, whereas in mice given Aloe vera were 8.13 ± 0.835 days. This indicates that wound healing time in mice belongs to normal wound recovery. According to the theory of Baradora (2008) on the 7th day wound healing appeared good as epithelization process rose in the wound. The epithelization process begins with the formation of granulation tissue in the wound. The granulation tissue consists of a combination of fibroblasts, collagen and macrophages. Fibroblasts originate in undifferentiated mesenchyme cells during epithelization. Fibroblasts multiply and form nets for migrating epithelial cells. In this migration process, the epithelial cells will touch each other and cover the wound entire surface. At this time recovery of epithelial functions the skin as a protector. In addition to this, fibroblasts also produce mucopolysaccharides, aminoglycine acids, and proline as the basic ingredients of collagen fibers that will link the wound edges. The whole process of epithelization is influenced by a substance called growth factor as well as the components present in Aloe vera. Factor of growth in Aloe vera plays a pivotal role in binding to IGF II receptors (Insulin-like growth factor II). IGF II receptors (Insulin-like growth factor II) play a role in stimulating fibroblasts to produce collagen and proteoglycans. Collagen is a major part of the connective tissue whereas proteoglycans are the spot of where collagen fibers get together. In addition to binding to the IGF II (Insulin-like growth factor II) receptor, the growth factor in Aloe vera also binds to the M-6-P receptor (Manosa-6-Phosphate) to form a collagen framework. At first the M-6-P receptor (Manosa-6-Phosphate) stimulates glucose formation in the form of glucosamine in fibroblasts. Glucosamine along with zinc and vitamin C in Aloe vera forms procollagen. Furthermore, Aloe vera encourages fibroblasts to convert procollagen into collagen. Epithelialization process that involves the formation of collagen will not take place properly if the initial stages of wound recovery do not run normally.

According to Morison (2003), the initial stage of wound recovery is vasoconstriction. Blood vessels cut off on the perineal wound will cause bleeding and as a mechanism of the body trying to stop it by vasoconstriction, shrinking the tip of the broken vessel (retraction), and hemostasis reactions. Hemostasis occurs because thromboxane and protombin in the blood stimulate platelet release. Platelets are trapped in nets formed by fibrin yarns and collect. This vasoconstriction and hemostasis process is the earliest mechanism to reduce bleeding rapidly. Shortly after the vasoconstriction process begins, the second wound recovery stage of the inflammation phase has begun. In this phase marked the presence of vascular response and cellular response to remove any foreign substances and dead tissues. The vascular response involves slowing blood flow changes and increased vascular permeability. Slower blood flow occurs due to local artery dilatation. This causes an increase in blood flow (hyperemia) in the perineal wound area so that the wound looks red and hot. Increased vascular permeability occurs due to the release of vasodilator substances such as histamine, serotonin, prostaglandins and cytokines at the time of injury. Increased permeability of blood vessels causes blood plasma fluid to escape from the blood vessels and into the wound area so that Sub-Cutaneous lesions appear edema and swelling occurs. Edema is one of the mechanisms for wound cleansing by neutralizing toxins and bacterial spread. In addition, the inflammatory phase cellular response occurs. Cellular response is the movement of leukocytes through the blood vessel wall (diapedesis) to the wound due to the power kemostasis. Leucocytes secrete hydrolytic enzymes to digest bacteria and wound dirt. Further in the first hour, monocytes will turn into macrophages that ingest debris and bacteria. In the area of the perineal wound will occur discharge or fluid exudate or transudate exudate is an inflamed extravascular fluid that has a high protein concentration, leukocyte-rich, cell debris that has died. From the theory of the inflammatory phase mentioned above, the normal inflammatory

phase can be concluded to have redness, swelling (edema), slightly increased temperature in the local area, the onset of pain, and the discharge in the form of transudate and exudate which appears within ≤ 5 days. To monitor the characteristics of the above inflammatory phases, the presence of REEDA marks can be used to observe the wound. Observation of REEDA marks is to observe the process of wound recovery which has begun since the occurrence of injury to the skin as the front line of resistance to the entry of the organism until signs of wound recovery rises. REEDA marks mean no redness, echimosis, edema, discharge, while the sign of approximation was already good. Based on observation result of REED sign at 30 mice treatment and control provides Aloe vera compress obtained average loss of redness at day 4, mean loss of echimosis on day 1, average loss of edema on day 4, average discharge loss on day 3. This suggests that the end of the inflammatory phase in respondents who provide Aloe vera compress is faster (<5 days).

This is because the effect of Aloe vera is synergistic with salicylic acid that Aloe Vera has an inhibiting system that blocks pain and inflammation. In addition salicylic acid in Aloe vera prevents prostaglandin biosynthesis from arachidonic acid. As a matter of fact, prostaglandin in the inflammatory phase acts as a chemical mediator that increases the permeability of the blood vessels so that the blood vessels dilate to increase blood flow to the wound. This causes the blood plasma fluid out of the blood vessels and into the wound area resulting in vascular effects that appear edema and swelling. Given the salicylic acid content in Aloe vera will prevent the biosynthesis of the prostaglandin thus reducing the immediate vascular effects that arise in the inflammatory phase. In addition to the role of salicylic acid content, Aloe vera also acts as vehicle for steroids. As we have seen one role of steroids in wound recovery is as an anti-inflammatory by blocking acute and chronic inflammation. A study conducted by Davis (2000), mentioned that these steroids can not be absorbed optimally by the stratum corneum because steroids require vehicles to assist them in penetrating into the stratum corneum. Herein lies the role of Aloe vera that can dissolve water-soluble compounds and fat soluble substances including steroids. Thus Aloe vera can help steroids and various other materials to penetrate the layers of the stratum corneum of the skin. Biological activity Aloe vera can synergize with many agents / substances that play a role in wound healing so that Aloe vera is referred to as "biological vehicle." In the above description has been discussed about the sign REEDA Redness, Echimosis, Edema, Discharge and the last is Approximation. According to the latest wound care theory, the best way to accelerate the growth of new cells in wounds is to create a humid environment. This is because the humid environment does not inhibit the flow of oxygen, nitrogen, and other airborne substances. Such a condition is a good environment for the body cells to stay alive and replicate optimally because basically the cells can be in a humid or wet environment. This is where the role of Aloe vera compresses to create a moist wound environment (moist), so that the wound healing process, especially the proliferation phase runs normally.

V. Conclusion

Conclusion

Based on the results of research and discussion about the effectiveness of Aloe vera compress on the wound healing time in mice, it can be concluded that:

1. Wound healing time in mice given Iodine Povidone compresses was for 6.86 ± 0.900 days.
2. Wound healing time in mice given Aloe vera is 8.13 ± 0.835 days.
3. A difference rises on wound healing time in mice given Iodine Povidone compress and Aloe vera compress with t-count value of 2,831 (p -value = 0,014).

Recommendation

Based on the conclusions of the research results, some recommendations can be proposed and are expected to be useful for practical interests and further research interests.

1. For Community
Especially for postpartum women, it is advised that they have to apply Aloe vera compress 3 times/day from the first day of post partum until the wound perineum are well-recovered.
2. For Health Manpower
Midwives are suggested to raise health awareness for personal hygiene to postpartum women, especially on postpartum perineum wounds with Aloe vera treatment and minimize complications

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