

Nasution Sri Lestari Ramadhani, Putri Monica, Hulu Welpin, Girsang Ermi, Nasution Ali Napihah. Healing Potential of *Senna Alata* Leaves Extract in Rats. Journal of Education, Health and Sport. 2019;9(5):127-136. eISSN 2391-8306. DOI <http://dx.doi.org/10.5281/zenodo.2707605> <http://ojs.ukw.edu.pl/index.php/johs/article/view/6889> <https://pbn.nauka.gov.pl/sedno-webapp/works/913160>

The journal has had 7 points in Ministry of Science and Higher Education parametric evaluation, Part B item 1223 (26/01/2017).
1223 Journal of Education, Health and Sport eISSN 2391-8306 7

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The authors declare that there is no conflict of interests regarding the publication of this paper.

Received: 28.04.2019. Revised: 28.04.2019. Accepted: 09.05.2019.

HEALING POTENTIAL OF *SENNA ALATA* LEAVES EXTRACT IN RATS

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Abstract

Burns known as damaged tissue which caused by contact with hot sources, such as water, fire, chemicals, electricity, and radiation. It has impact not only to skin damage but also to the whole body system. *Senna Alata* (Cassia alata L) is considered as herbal medicine with spicy and warm

taste which believed could heal itching, skin damage and others. This study aimed to analyze the optimization of *Senna Alata* leaves extract in healing burn. Thirty Wistar rats or *Rattus norvegicus* were used as the sample. Those Wistar rats were divided into 6 groups with 5 rats in each group which treated differently. Group 1 was negative control which given no treatment at all, group 2 was treated with bioplacenton cream, while the rest of the groups were treated using *Senna Alata* leaves extraction cream with different concentration; 25% for group 3, 50% for group 4, 60% for group 5, and 95% of extraction cream for group 6. This study used experimental research as the methodology using ANOVA and Tukey test. The result showed that healing potential of *Senna Alata*'s leaves extract had significant effect in healing burn.

Keywords: healing potential, *Senna Alata* leaves extract, burn, ethanol extract

Introduction

Burn is known as damaged tissue due to contact with agents, thermal, chemical, or electricity. Burn loses its tissue since its interaction with heat sources, such as; water, fire, chemical materials, electricity, and radiation which could inflicts to skin damage and the whole body systems⁽¹⁾. Burn's healing process could be divided into three phases, including; inflammation, proliferation, and (c) respectively. The inflammation phase happens since the early occurrence of burn to the seventh days of burn. Then, continued to the proliferation phase which happens in the end of the inflammation phase until the end of the third week. While the respectively phase happens for several months and could be declared healed after the whole inflammation signs are gone⁽²⁾. Another way in healing burn is by giving plant's extract to the injured area. Plant is one of the biodiversity which varies and has potential to utilize and develop as a traditional medicine's basic ingredients⁽³⁾. *Senna Alata* (*Cassia alata* L) or known as emperor's candlesticks is recognized as one of the medical plants used for curing many kinds of diseases. It is warm and has spicy taste. It also contains some components, such as *tannin*, *rein*

aloe-emodina, *rein aloe-emodinadiantron*, *chrysophanic acid*, *alkaloid*, *saponin*, *flavonoid*, and *anthraquinone glycosides*. Basically, the pharmacology effects on *Senna Alata* (*Cassia alata L*) could be used as laxative, anthelmintic, anti-itch cream, and drug for skin disorder caused by skin parasite⁽⁴⁾.

Materials and Methods

Tools and Materials

The tools used in this study were metal with 2x2 mm diameter, analytical balance, blender, mortar porcelain, filter cloth, spatula, container, razor, brush, digital caliper, maceration tank, rotary evaporator, aluminum foil, tissue paper, plaster, gauze bandage, hand gloves, and electric oven. While the materials consisted of ketamine, *Senna Alata* (*Cassia Alata L*) leaves extract, 96% ethanol and Vaseline.

Experimental Design and Animal Grouping

This study was using thirty Wistar rats (*Rattusnorvegicus*) aged around 2-3 months with average weight of 200-250g which taken from the Pharmacology Laboratory of North Sumatera University. Those Wistar rats were divided in 6 groups, each group consisted of 5 with classification as below;

- F1 : no treatment
- F2 : treated with bioplacenton cream or ointment for burn
- F3 : given 25% concentration of *Senna Alata* leaves extract ethanol preparation cream
- F4 : given 50% concentration of *Senna Alata* leaves extract ethanol preparation cream
- F5 : given 60% concentration of *Senna Alata* leaves extract ethanol preparation cream
- F6 : given 95% concentration of *Senna Alata* leaves extract ethanol preparation cream

Wistar rats in group 1 was given no treatment at all, while in group 2 bioplacenton cream was given. The rest of the groups were given *Senna Alata* leaves extract ethanol preparation cream, ranged from 25% for group 3, 50% for group 4, 60% for group 5 and 95% for group 6.

Phytochemical screening

Senna Alata (*Cassia Alata L*) consists of *alkaloid*, *saponin*, *tannin*, *flavonoid*, and *polyphenol* compounds.

Preparation of ethanol extract

Senna Alata leaves were dried in a room temperature for 1 - 5 days. After that *Senna Alata* leaves were cut into small pieces and dried again using oven for an hour with temperature of 50 – 60°C. The dried leaves then blended using blender until it had smooth powder texture and measured for 1000g. The measured leaves then placed into a container and dissolved in 96% ethanol until the whole leaves were sunken. Then covered with cap in order to protect from light and rested for 5 days long with regular stirring for each days. After that, filtered into an empty container and the dregs were macerated back with 96% ethanol. The maceration process was done until 3 times filtering. Thus, the extract was collected and concentrated with rotary evaporator, and the ethanol was removed from the extract.

Table 1. The Making of Cream and *Senna Alata* Leaves Extract Preparation

Ingredients	Sample Preparation in Cream Base			
	Concentration 25% w/w	Concentration 50% w/w	Concentration 60% w/w	Concentration 95% w/w
Senna Alata	25 gr	50 gr	60 gr	95 gr
Leaves Extract				
Vaseline	75 gr	50 gr	40 gr	1. Gr

The Making of Cream and *Senna Alata* Leaves Extract Preparation

The making of cream using *Senna Alata* leaves extract was started with the defined measurement which put into mortar and stirred until it homogeny. The extract then, added into each concentrations. Therefore, the preparation cream had concentration of *Senna Alata* leaves extract for 25%, 50%, 60%, and 95%.

Making Wound in Wistar rats

Thirty Wistar rats aged for 2-3 months, weighing 200-250gr were used for experiment. Those were placed in laboratory for a week for adaptation before the experiment was started. The rats were given enough food and nutrition due to keep their weight stable. After that, the rats were anesthetized using 0.2 – 0.3 cc doses of ketamine to make them unconscious. Then the hair on their back were shaved with diameter of 2cm and disinfected using 70% of alcohol. The burn then made using metal heated with Bunsen fire for 30 minutes. The hot metal was attached to the

rats' skin for about 2 – 5 seconds until it formed the burn injury, which marked with red color on the skin. The burns area in the back of Wistar rats were treated based on the control groups, including negative control (without giving any treatment), positive control (giving burn ointment or bioplacenton), and given the extract of *Senna Alata* leaves with the concentration ranged from 25%, 50%, 60%, and 95%. The treatment were done by treating the rats two times a day started from day one until day twenty two. The wound were left open until it heal which indicated with the closed wound.

Observation on Wistar rats Burns

This study used quasi experimental design by observing the Wistar rats three times a day in 21 days long. The wounded area was measured in day 3, day 6, day 9, day 12, day 15, day 18, and day 21 from various directions.

$$I_x = \frac{I_x(1) + I_x(2)}{2}$$

Information:

I_x : wound area in day x (cm)

$I_x(1)$ and $I_x(2)$: wound area measured from various directions (cm)

Result

Extraction Results and Phytochemical Screening

The percentage yield of the extract was 1200gr of *Senna Alata* leaves which macerated in 96% ethanol. The weight after immersion was 6000 gr and rotated until it became caramel preparation cream weighted for 250 gr. The phytochemical screening of *Senna Alata* leaves' extract and ethanol could be seen on tables below:

1. Alkaloid Identification

Sample	Reactor			
	Bouchardart	Dragondoff	Maeyer	Wagner
Dried <i>Senna Alata</i>	++	++	++	++

2. Steroid and Triterpenoid Identification

Sample	Reactor
	Salkowsky
Dried <i>Senna Alata</i>	-

3. Saponin Identification

Sample	Reactor		
	Aquadest	Aquadest + Alcohol 96%	Aquadest + Alcohol 96% + HCL 2N
Dried <i>Senna Alata</i>	+	+	-

4. Flavonoid Identification

Sample	Reactor			
	FeCL3 5%	NaOH 10%	H2SO4 (p)	Mg(s) + HCL(p)
Dried <i>Senna Alata</i>	++	-	-	+

5. Tannin Identification

Sample	Reactor
	FeCL3 5%
Dried <i>Senna Alata</i>	++

6. Polyphenol Identification

Sample	Reactor
	FeCL3 5%
Dried <i>Senna Alata</i>	++

Based on the tables above, it showed that *Senna Alata* leaves extract contained with *tannin, saponin, flavonoid, alkaloid* and *polyphenol*.

Optimization Result based on Treatment Concentration.

The optimization of *Senna Alata* leaves extract in healing burn based on treatment concentration (negative control, positive control, 25%, 50%, 60% and 95%) it was conducted by ANOVA test and the results as follows:

Table 2. ANOVA test

Wound Area					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	65.528	5	13.106	49.796	.021
Within Groups	3357.838	204	16.460		
Total	3423.366	209			

Table 2 above showed that value of sig-p = 0.021 was smaller than 0.05. It meant that *Senna Alata* leaves' extract significantly affected the wound area. The application of *Senna Alata* leaves extraction based on concentration successfully narrowed wound area significantly. In other words, *Senna Alata* leaves' extract was effective to regenerate burn on Wistar rats' skin. Moreover, Tukey test was conducted to identify the how much *Senna Alata* leaves' extract concentration that effectively regenerated burn. The results as follow:

Table 3. Results of Tukey test between wound area and treatment concentration

Treatment	N	Subset for Alpha = .05
		1
60%	35	16.5486
50%	35	17.0529
K -	35	17.4843
95%	35	15.5014
25%	35	17.5743
K +	35	18.3914
Sig.	35	.405

Means for groups in homogeneous subsets are displayed

Uses harmonic mean sample size = 35.000

Table 3 showed that the subset of group 1 had the largest wound area obtained on K+ treatment for 18.38. Meanwhile, the smallest area for 15.50 was obtained by 95% treatment. It meant that 95% concentration treatment was more optimally healing the burn. However, there was no significant difference between 6 treatments. It was identified by significant value for p = 0.405 which bigger than p = 0.05.

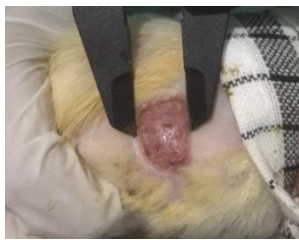


Fig. 1. F1 Negative Control



Fig. 2. F2 Positive Control



Fig. 3. F3 25% Concentration

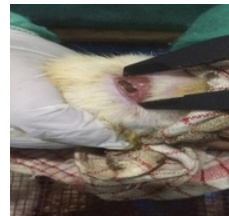


Fig. 4. F4 50% Concentration



Fig. 5. F5 60% Concentration



Fig. 6. F6 95% Concentration

Discussion

Based on the treatment concentration, ANOVA test result showed the value of sig-p = 0.021 was smaller than 0.05. It meant that *Senna Alata* leaves' extract was significantly affected the wound area depend on the treatment concentration. The most optimal effect was obtained by 95% treatment concentration which identified by significant value p = 0.405 bigger than 0.05. This experiment was in accordance with Noor Hujitusnaini's experiment entitled, "Uji Potensi Ekstrak Daun Ketepeng Cina (*Cassia Alata* L) terhadap Penghambatan Pertumbuhan *Trichophyton* sp". The study claimed that *Senna Alata* leaves' extract was effectively inhibited the growth of *Trichophyton* sp that caused skin disease such as phlegm and ringworm, with clinical figure in the form of sphere skin surface – a small circle with red and scaly edges and a slippery center without hair in the area of infection⁽⁵⁾.

It also similar to the study written by Mahmudah et al, entitled, “Uji Efektivitas Ekstrak Etanol pada Daun Ketepeng Cina (*Cassia Alata L*) terhadap Mikroba Penyebab Sariawan (Stomatitis Aphthosa)”. This study showed that flavonoid in *Senna Alata* leaves could inhibit the bacterial growth that caused sprue identified by the decrease of bacteria colony. The higher the extract’s concentration, the lower the amount of the bacteria colony⁽⁶⁾. Some chemical substance in *Senna Alata* leaves extract that could inhibit bacteria or fungus growth were; *tannin*, *rein aloe-emodine*, *rein aloe-emodina-diantron*, *chrysophanic acid*, *alkaloid*, *saponin*, *flavonoid* and *gulcoside antraxion*. Generally, pharmacologic effect of *Senna Alata* leaves was functioned as laxative, worm medicine, itching remover, and skin abnormalities caused by skin parasites⁽⁴⁾.

Conclusion

Based on the experiment result, it could be concluded that *Senna Alata* leaves’ extract (*Cassia alata L*) had significant effect toward burn healing in Wistar rats (*Rattus norvegicus*), either based on the treatment time or treatment concentration. Moreover, the most optimal concentration treatment in healing burn was using 95% concentration of *Senna Alata* leaves’ extract with the wound area for 15.50, smallest from the other concentrations.

Conflict of Interest

Authors declares there is no conflict of interest

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