American Scientific Research Journal for Engineering, Technology, and Sciences (ASRJETS)
ISSN (Print) 2313-4410, ISSN (Online) 2313-4402

http://asrjetsjournal.org/

Effectivity of Gel Ethanolic Extract of Senggani Leaves (*Melastoma candidum* D. Don) in Increasing the Number of Fibroblast Cells and Thickness of Collagen Fibers Against Socket Wound after Tooth Extraction on Male White Rats

Susanna Halim[a]*, Ermi Girsang[b], I Nyoman Ehrich Lister[c], Ali Napiah Nasution[d]

[a,b,c,d] *Facutly of Medicine, University of Prima Indonesia, Medan, North Sumatera, Indonesia*
[a]*Email: susanna_halim@yahoo.com*

Abstract

The important cells in wound healing process are the fibroblast cells and the density of collagen fibers. The Senggani leaves (Melastoma candidum D.Don) contains some active compound of flavonoids / phenols, triterpenoid / steroid, saponins, tannins and glycosides. This research aims to determine the effectiveness of gel ethanolic of Senggani leaves (Melastoma candidum D.Don) towards increased number of fibroblast cells and collagen density score in the process of wound healing after tooth extraction on male white rats (Rattus noevegicus). The research design was purely experimental in vivo, design post test only control group and non – randomly or non – probability sampling using the sample of 25 Wistar rats. Male white rats anesthetized intra-peritoneal (IP) using ketamine 100 mg / 1 ml at a dose of 20-40 mg/kg body weight of the rat, then extraction of the mandibular left incisor using a sharp excavator and pulling the pliers off, then Dental sockets are irrigated with a sterile was given Aloclair Plus gel ®, the treatment group was given 1% EDS, 5% EDS and 10% EDS twice a day with a frequency of 12 times the treatment.

---

* Corresponding author

On the seventh day, euthanasia was carried out, the rats were put into a closed box containing ether cotton until suffocation, followed by histological tissue making. Histology preparations were seen with the Boeco Binocular microscope BM-180 SP model, with the help of MDCE-5A digital microscope camera connected to a binocular microscope through the eyepiece entry hole, and using a 10 x objective lens and analyzed using Adobe Photoshop CS 6.0 software to view a number of fibroblast cells and density of collagen fibers. Calculations were carried out with 5 fields of view then calculated how many fibroblasts and collagen density scores in each field of view so that they were clearly visible. From the field of view 1 to 5 it is added up, taken the average. The results of descriptive analysis of the number of fibroblast cells and collagen fiber density scores showed that the average number of fibroblast cells and the highest collagen fiber density score in the 10% EDS treatment group were 83,133 and 3,933, followed by the positive control group of 82,667 and 3,667, the 5% EDS treatment group is 41,200 and 3,067, the EDS treatment group is 1% which is 33,800 and 3,000, and the lowest result is negative control which is 16,533 and 1,200. Data analysis was using Shapiro Wilk normality test, Levene homogeneity test, One way Anova test and the test of the comparative effect (LSD = Least Significant Difference and Tukey's HSD = Honestly Significant Difference). The results showed that has normal distribution of data ($p > 0,05$) and homogeneity of data ($p > 0,05$). One way annova test results the significant number of fibroblast cells and collagen fiber density score on the fifth group (given Melastoma candidum D. Don rind extract gel concentration of 10 %). The conclusion showed that Melastoma candidum D. Don rind extract gel concentration of 10 % is effective towards increased the number of fibroblast cells and thickness of collagen fibers score against socket wounds after tooth extraction in male white rats.

Keywords: Gel ethanolic extract of Senggani Leaves; Rattus norvegicus; number of fibroblast Cell; Collagen density score; tooth extraction and wound healing.

1. Introduction

Tooth extraction is a common practice in dental practice. In Indonesia the utilization of dental and oral health services for tooth extraction is very high, reaching 79.6%. Tooth extraction is a surgical procedure that involves mobile tissue and soft tissue from the oral cavity, access which is limited by the lips and cheeks, and then connected or united by tongue and jaw movements, using pliers, elevators, or emphasis on alveolar trans[1]. Definition ideal tooth extraction is painless extraction of a single tooth or tooth root with minimal trauma to the supporting tissues of the tooth so that the extraction marks can heal completely and there are no prosthetic problems after tooth extraction in the future[2]. The process of wound healing is a complex and physiological process in the body. Wound healing consists of several phases, namely inflammation, proliferation, and maturation. Wound healing is needed to regain intact body tissue. The sequence in the healing of an alveolar socket after tooth extraction was: Reference [1] blood clot formation; [2] replacement of the blood clot by granulation tissue; [3] substitution of granulation tissue by connective tissue ; [4] complete epithelization, [5] appearance of osteoid tissue at the base of the socket ; and [6] filling in of the apical two thirds of the socket by trabecular bone. Several factors play a role in accelerating healing, namely internal factors, and external factors. External factors that can accelerate wound healing by wound irrigation using physiological solutions (0.9% NaCl) and the use of synthetic and natural medicines [5]. A drug commonly used for healing post-extraction wounds is povidone iodine which acts as a bacteriostatic for all germs. Excessive use of povidone-iodine can

cause side effects in the form of itching, pain that is very close to the area of the wound, swelling, and dermatitis. Nowadays many herbal medicines have been developed which have health benefits with low side effects [6]. Therefore, many people now use Aloclair plus gel ®. Aloclair is a medicine to help reduce pain due to canker sores while preventing further sores. Aloclair plus gel ® contains aloe vera extract (Figure 1) which functions to heal wounds, reduce pain, and prevent infection, sodium hyaluronate to moisturize and help heal wounds, glychyrrhetinic acid to reduce swelling and pain, polyvinylpyrrolidone for the formation of protective layers, and cinnamon anti-infectious [7], in this study Aloclair Plus Gel ® was used in the positive control group.

Figure 1: Aloe vera

Indonesia is a tropical region that has a variety of plants. In Indonesia, many plants are used as herbal medicines, one of which is senggani leaves (Melastoma candidum D. Don). According to Starr and his colleagues (2003), the name Melastoma comes from Greek. "Melas" means black and "stoma" means mouth. This naming is based on the appearance of black on the edge of the mouth when someone eats it. This plant can be found in Madagascar, India to Australia, but can also be easily found in Southeast Asia, including Indonesia. This Senggani habitat grows wild in places that get enough sunlight, such as on the slopes, shrubs, fields that are not too arid, or in tourist areas as ornamental plants. This plant can usually be found at an altitude of 1,650 meters above sea level [8]. Morphology Senggani plants in the form of shrubs or small trees. The stem is woody, brown in color, erect as high as 1.5-5 m with sympodial branches. The leaves are single, stemmed, they are cross-faced. The leaves are green, ovoid 2-20 cm long and 1-8 cm wide, have pointed ends and base of leaves, flat edges of leaves, rarely short-haired surfaces and are stiff so they are roughly rough with 3 curved leaf bones, with petiolus length 5-12 mm [8.9]. Senggani plants can be seen in Figure 2.

Figure 2: Senggani (Melastoma candidum D.Don)

Senggani works to overcome food indigestion (dyspepsia), bacillary dysentery, diarrhea, hepatitis, leucorrhoea (leukorrhea) and canker sores. In addition Senggani is also often used to treat excessive menstrual blood, uterine bleeding outside of menstruation, nosebleeds, defecation (melena), bleeding hemorrhoids, inflammation of the walls, blood vessels accompanied by blood clots in the ducts (thromboangitis), breast milk (ASI) non-fluency, cassava poisoning, intoxication, drinking water, scabies and boils [8,10]. Senggani has various chemical ingredients, especially in the leaves. The chemical content of Senggani leaves includes the content of flavonoids / phenols which are powerful antioxidants and anti-inflammatory, triterpenoid/steroid which are anti-inflammatory which can prevent stiffness and pain, saponins have an antibacterial function and are precipitating and coagulating cells red blood, glycosides that are analgesic and reduce surface tension and hydrolyzed tannins, commonly called Nobotanin B, function to accelerate the process of wound healing through cellular mechanisms and are antimicrobial by increasing epithelialization [8.10]. Senggani flower contains kaempferol, anthocyanin, tannin, fatty acids, and sterols. Senggani fruit is reddish purple and is thought to contain anthocyanin. Senggani fruit can be used as a source of natural dyes. In general, in measuring antioxidants, the most frequently used ethanol solvents are [10]. Based on this, senggani leaves have the potential to be formulated as topical preparations. One effective dosage form for topical therapy is gel [11]. Gel (from Latin gelu - freezing, cold, ice or gelatus - freezes) is a colloidal mixture between two different phase substances: solid and liquid [12]. A gel is usually applied to mucous membranes injured or burnt tissue. The gel is preferred because its use leaves a transparent, elastic layer, good drug release and attractive dosage appearance [13]. In addition, gel preparations are chosen because of the high water content which provides a cooling effect for the mucosa or skin and reduces irritation [14]. Some studies show senggani leaf extract (EDS) has benefits in the process of wound healing especially to increase the number of fibroblasts and collagen fibers. Fibroblast cells (L. fiber, fiber: Greece. Blancos, seed: Latin) are the cells most commonly found in connective tissue and synthesize several components of the extracellular matrix (collagen, elastin, reticular), several anionic macromolecules (glycosaminoglycans, proteoglycans) and glycoproteins multi adhesive, laminin, and fibronectin which can encourage cell attachment to the substrate. In addition, fibroblast cells secrete cytokines and several growth factors (growth factors) which can stimulate cell proliferation and inhibit differentiation. The proliferation phase is a phase where fibroblasts lay the basic substance and new collagen fibers and blood vessels will infiltrate wounds Phase is called phase fibrillation because in this period the role of fibroblasts is very prominent. Collagen fibers that are formed cause the strength to link the edges of the wound. In this phase, granulation begins, wound contractions and epithelial [3,15]. Based on the description above, this study aims to determine the effectiveness of ethanol extract of senggani leaves in increasing the number of fibroblast cells and thickness of collagen fibers against socket wounds after tooth extraction in male white rats. (Use 10 point **font,** times new roman) Here introduce the paper. The paragraphs continue from here and are only separated by headings, subheadings, images and formulae. The section headings are arranged by numbers, bold and 10 pt. Here follows further instructions for authors.

2. Materials and Methods

The research design was purely experimental *in vivo*, which is a study by conducting experiments which aims to

determine the symptoms or effects that arise as a result of the existence of certain treatments or experiments, to determine the influence or relationship of independent variables with the dependent variable. The research design used is *The Post Test Only Control Group Design*, which is taking measurements after the treatment is given, then the results are compared with the control group [12]. Research conducted at Pharmacology and toxicology laboratory, Faculty of Pharmacy, Universitas Sumatera Utara. Independent variables are Aloclair plus gel, Without treatment, Gel ethanolic extract of senggani leaf 1 %, Gel ethanolic extract of senggani leaf 5 %, and Gel ethanolic extract of senggani leaf 10 %. Dependent variables are histopathology description based on total fibroblast cell and histopathology description based on density of collagen fibers. Dependent controlled are place (breeding place, temperature, and light), maintanance of rats (food and water), white male rat wistar train (age and body wwight), way of make ekstract and gel excract senggani leaf, way of gicing, time and dose of gel extract senggani leaf given, way of extracted the tooth of rat, way of make tissue preparation, way of count fibroblast cell, density score of collagen fibers, and diameter of wound. Sampling is done by non-random (non-probability) sampling or sampling is not random, ie sampling that is not based on possibilities that can be calculated, but solely based solely on mere aspects of practicality. With a purposive sampling technique or purposive sample that is without comparing with other regions taken in Punden Rejo Village, Deli Serdang Regency, North Sumatra Province [12]. Purposive sampling is based on certain considerations made by the researcher myself, based on the characteristics or characteristics of the population that were previously known. The animals used in this study were male white Wistar rats (Rattus norvegicus) [13]. Before this study began, the test animals were acclimatized for one week at room temperature conditions (22-25 ° C), under a 12 hour light / dark cycle, fed pellets and drinking water ad libitum tap. This is to obtain uniformity before conducting research to control experimental animals. Plant identification is carried out at Herbarium Medanense (MEDA), Faculty of Mathematics and Natural Sciences University of North Sumatra Medan, with No.: 4014/ MEDA / 2019. Sample of fresh senggani leaves were washed until clean, then drained and weighed. Next the leaves are dried in a cabinet dryer with a temperature of 40 o C for + 24 hours untilthe leaves dry. The next stage, the samples that have been dried and pounded coarsly in a mortal and stamfer, as well as aerated again until dry + 8 hours, and then pulverized with a blender to become powder and kept in tightly closed containers at room temperature. Preparation of extracts was carried out by maceration using 80% ethanol solvents because polyphenols from senggani leaves were polar and relatively stable under acidic conditions so that the polyphenols in senggani leaves were more soluble in polar solvents such as methanol and ethanol. As much as 1 kg of simplicia powder is put into a vessel, poured with 75 parts 80% ethanol, closed, left for 5 days protected from light while occasionally stirring, shaking, squeezing, then through enough pulp to obtain 100 parts with 80% ethanol. Transfer the macerate to a closed vessel, leave it in a cool place protected from light for 2 days, settle and pour. The concentration of the extract was carried out by rotary evaporator at a temperature of ± 50 oC until thick extract was obtained, then freeze-drying was carried out with freeze dryer at -40 oC for ± 24 hours [14]. Quantitative examination of ethanol extracts including the determination of moisture content aims to provide a minimum or range limitation about the amount of water content in simplicia and extracts, the determination of water-soluble juice content aims to determine the level of water-soluble (polar) extracts, determination of soluble juice content ethanol to find out the compounds dissolved in ethanol, both polar and non-polar, determination of total ash content to provide an overview of internal and external mineral content from the initial process to the formation of extracts, and determination of ash content insoluble in acid to

evaluate simplicia and extract against contamination of silica-containing materials, heavy metals such as lead / plumbum (Pb) [15]. Phytochemical screening of the ethanol extract of senggani leaves included: an examination of alkaloid compounds, saponins, flavonoids, tannins, triterpenoids / steroids, and glycosides. The process of making ethanol extract of senggani leaves using maceration techniques. Starting from carbopol, it is developed with part of heated distilled water. Let stand in a mortar for 15 minutes, where the mortar is inserted in a container containing hot water to help the process of forming gel viscosity. Carbopol was added with glycerin, crushed, then added water and crushed again (mass 1). Mass 2 is formed from propylene glycol which is added to the remaining water, then add Nipagin, after which it is heated. Mix mass 1 with mass 2, then Triethanolamine (TEA) is put drop by drop into the mixture until a gel mass is formed. Stir until homogeneous and add the remaining aquades to form a homogeneous gel mass. Add a little base gel to the mortar, then the ethanol extract of senggani leaves is added and crushed until homogeneous [16]. Preparations are made in 4 formulas with a composition of 200 g each shown in Table 1 below. How to make EDS gel preparations:

a. into the mortar the EDS is entered each with a concentration of 1%, 5%, and 10%,
b. dropped with a few drops of ethanol,
c. added little by little the base of the gel then crushed until homogeneous.

Table 1: Composition formula of gel EDS

No.	Bahan Gel	EDS 1 %	EDS 5 %	EDS10 %
1.	Carbopol	2 g	2 g	2 g
2.	Nipagin	0,2 g	0,2 g	0,2 g
3.	Gliserin	1 g	1 g	1 g
4.	TEA	2 g	2 g	2 g
5.	Propilen Glikol	6 g	6 g	6 g
6.	Aquadest	87,8	83,8	78,8
7.	Ekstrak Etanol Daun Senggani	1 g	5 g	10 g

The organoleptic examination was carried out to see the physical appearance of the preparation by observing the shape, color, and smell of the preparations made. The gel preparation was applied to a piece of glass and then observed whether there were parts that were not properly mixed. A good gel shows a homogeneous arrangement. The pH measurement of the dosage is done using a pH meter. Preparations were taken and placed at the pH meter sample, then wait until the pH meter indicator was stable and showed a constant pH value, the pH results obtained compared to the pH range of the skin between 4.50 - 6.50. The viscosity test is done by a mechanical method using a viscometer, speed (speed) 12, Spindle number 4 or 64, a correction factor (spindle factor) 500, the scale obtained from the spindle playback is multiplied by the spindle factors. After gel 1%, 5% and 10% were available, the treatment was 25 mice adapted for a week, then divided into 5 groups, namely the negative group (without treatment), positive group (given Aloclair Plus gel ®), gel group EDS 1%, 5% EDS gel group, and 10% EDS gel group. Male white rats anesthetized intra-peritoneal (IP) using ketamine 100 mg / 1 ml at a dose of 20-40 mg/kg body weight of the rat, then extraction of the mandibular left incisor using a sharp excavator and pulling the pliers off, then Dental sockets are irrigated with a sterile aquades solution. Furthermore, the negative group was not given treatment, the positive group was given Aloclair Plus gel ®, the treatment group was given 1% EDS, 5% EDS and 10% EDS twice a day with a frequency of 12 times the

treatment. In male white rats diameter measurements were carried out using calipers and wound depth using UNC 15 probes on days I, III, V, and VII. Then a descriptive analysis, normality test with Shapiro Wilk was made because the amount of data was only 25 samples (less than 50 samples) and homogeneity test with Levene-Test, parametric test with one way ANOVA and followed by Post Hoc Test which was a test of the effect of comparability (LSD = Least Significant Difference and Tukey's HSD = Honestly Significant Difference).

3. Results and Discussion

3.1 Determination of senggani leaf

The plants used have been identified at Herbarium Medanense (MEDA), University of North Sumatra with no. 4014 / MEDA / 2019 is senggani leaf plant (Melastoma candidum D.Don).

Kingdom : Plantae (kerajaan Tumbuhan),

Divisi : Spermatophyta (tumbuhan berbiji),

Kelas : Dicotyledoneae

Ordo : Mytales,

Famili : Melastomaceae,

Genus : Melastoma,

Spesies : Melastoma candidum D. Don

Local name : Senggani

3.2 Quantification examination on simplicia and ethanolic extract of senggani leaf

Quantification examination on simplicia and ethanolic extract of senggani leaf according to material [15, 17].

Table 2: Result of characterization of simplicia and extract of senggani leaf

No	Parameter	Result (%)	
		Simplicia	Extract
1	Water content	6,66	11,31
2	Water soluble extract content	27,68	39,68
3	Ethanol soluble extract content	33,83	66,58
4	Total ash content	3,61	2,18
5	Acid insoluble ash content	0,64	0,25

The results of phytochemical screening of senggani leaf Simplicia powder in the Pharmacy-Biology laboratory,

USU Faculty of Pharmacy, Medan with No. 2091 / UN5.2.1.11 / PSS / 2019 shows the presence of tannins, saponins, triterpenoids/steroids, , and glycosides.

Table 3: Result of phytochemical content

No	Screening	Reagents	Observation	Result	
			Before	After	
1	Tanin	$FeCl_3$ 1 %	Green	Blacking green	+
2	Saponin	Distilled water	Green	Bublly / Froth Foam	+
3	Flavonoid	HCl p, Mg	Green	Red orange	+
4	Steroid/ Triterpen	cloroform, acetic acid anhidrat, H_2SO_4 p	Green	Ring formation	+
5	Glikosida	Ethanol 96% aetic acid anhidrat, H_2SO_4 p	Green	Purple ring formation	+
6	Alkoloid	HCl 2N, distilled water, Dragendroff	Green	There is no reddish brown precipitate	-
		HCl 2N, Distilled water, Wagner	Green	There is no reddish brown precipitate	-
		HCl 2N, Distilled water, Meyer	Green	White sediment is not formed	-

Information : + = contains compound, - = does not contain compound

3.3 Result of test gel ethanolic extract of senggani leaf

The results of the gel test of the ethanol extract of senggani leaves aimed to provide a general description of the physicochemical properties of the gel preparations of the ethanol extract of senggani leaves so that they can be known for their physicochemical properties and safety before use. The gel test was carried out at the Physics Pharmacy laboratory, Faculty of Pharmacy, USU with the number: 2092 / UN5.2.1.11 / PSS / 2019 with the following results:

a. Result of organoleptic test

Table 4: Organoleptic test

No.	Formula	Color	Savor	consistency
1	F1 (EDS1%)	Thick brown	distinctive smell of senggani	viscous
2	F2 (EDS5%)	Thick brown	distinctive smell of senggani	viscous
3	F3 (EDS10%)	Thick brown	distinctive smell of senggani	viscous

The color of the gel produced from the ethanol extract of senggani leaves from all variations of concentration

was different. The color difference of all gel preparations was due to differences in the concentration of active substances that followed the color of the ethanol extract of senggani leaves which were blackish green in color, while the K + (positive control) of the active substance used was very different so the resulting color was also different. Color differences can be seen in Figure 2. The odor produced from F1 (1%), F2 (5%), and F3 (10%) are caused by the ethanol extract of senggani leaves which have a distinctive aroma. The results of the organoleptic examination of gel preparations can be seen in Table 4.

b. Result of homogeneity test

The homogeneity test results showed that all gel preparations were F1 (1%), F2 (5%), and F3 (10%), giving homogeneous results which were characterized by the absence of coarse grains when the preparation was applied to transparent glass. This shows that the preparations made have a homogeneous arrangement. The results of homogeneity checks can be seen in Figure 3.

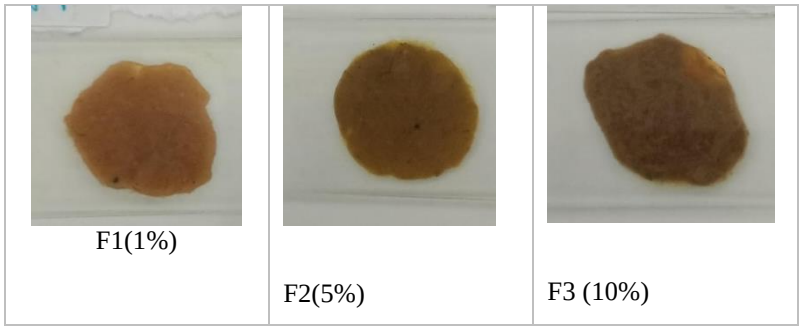

Figure 3: Result of homogeneity

c. Result of pH test

The pH test results show that all gels meet the pH criteria for skin or gum, which are intervals of 4.5-6.5, so that the pH of preparations in F1 (1%), F2 (5%), and F3 (10%), can be accepted by the skin or gum. The difference in the pH value of each gel was caused by the ethanol extract of senggani leaves in the gel, the greater the concentration of the ethanol extract of senggani leaves, the pH value in the gel will be smaller, and vice versa the smaller the concentration of ethanol extract the leaves will increase. (table 5.)

Table 5: Result of pH test

No.	Formula	pH
1	F1 (1%)	6,5
2	F2 (5%)	6,2
3	F3 (10%)	6,0

d. Result of viscosity gel ethanolic extract of senggani leaf

The consistency test is done by mechanical means using a viscometer, speed (speed) 12, Spindle number 4 or

64, a correction factor (spindle factor) 500, the scale obtained from the spindle playback is multiplied by the spindle factor. For F1 (1%) the scale obtained is 10.5 multiplied by 500 obtained by the viscosity of 5250 cp. For F2 (5%) the scale obtained is 13.5 multiplied by 500, the viscosity is 6750 cp. For F3 (10%) the scale obtained is 16.5 multiplied by 500, the viscosity is 8250 cp. (table 6.)

Table 6: Result of viscosity test gel ethanolic extract senggani leaf

No.	Formula	Viscosity
1	F1 (EDS 1%)	5250 cp
2	F2 (EDS 5%)	6750 cp
3	F3 (EDS 10%)	8250 cp

3.4 The effectiveness of the ethanol extract gel senggani extract (Melastoma candidum D. Don) on socket wound healing after tooth extraction on rat

Table 7: The average value of fibroblasts and collagen density scores

No.	Group	Mean Fibroblast	Mean collagen
1	K (-)	16,53	1,20
2	K (+)	82,6	3.67
3	EDS 1%	33,8	3,00
4	EDS 5%	41,2	3,07
5	EDS 10%	83,13	3,93

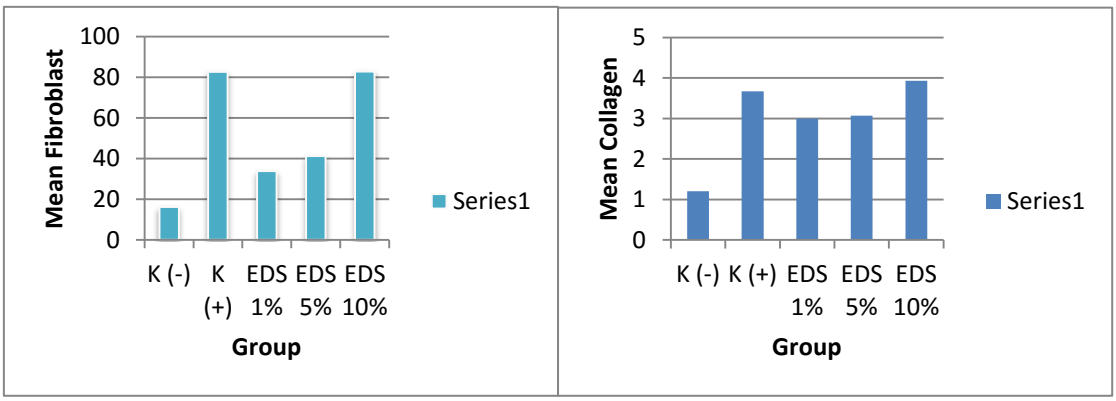

Figure 4: The average value of fibroblast and collagen density scores

The descriptive analysis of the number of fibroblast cells and collagen fiber density scores showed that the average number of fibroblast cells and the highest collagen fiber density score in the 10% EDS treatment group were 83,133 and 3,933, followed by the positive control group of 82,667 and 3,667. the 5% EDS treatment group is 41,200 and 3,067, the EDS treatment group is 1% which is 33,800 and 3,000, and the lowest result is

negative control which is 16,533 and 1,200. The normality test with Shapiro-Wilk showed that the average data on fibroblast cell count and collagen fiber density score in each group were normally distributed with a value of p> 0.05. Levene Test Homogeneity Test obtained a significant value of p> 0.05 which indicates the data has the same variant (homogeneous). The One Way ANOVA test results shown in the SPSS table show a significant value of 0,000 on day 7 or p <0.05 on days 3 and 5, indicating that there is healing in socket wounds with increased of fibroblast cell count and collagen fiber density score in each treatment group. The One Way Annova test can only show whether there is a difference in effectiveness between treatment groups towards positive controls and negative controls. So the ethanol extract of senggani leaves (Melastoma candidum D. Don) is effective against socket wound healing after tooth extraction in male white Wistar rats. To find out the difference in the effectiveness of each treatment group, a further test with Post Hoc Test was conducted, namely the test of the comparative effect (LSD = Least Significant Difference and Tukey's HSD = Honestly Significant Difference). The results of this follow-up test showed that the highest Mean Difference in the EDS group was 10% compared to the positive control group, followed by the 5% EDS group, 1% EDS group and the lowest Mean Difference in the negative control group. So the most effective ethanol extract of senggani leaves (Melastoma candidum D. Don) against socket wound healing after tooth extraction in male white Wistar rats was 10% EDS compared to positive controls (aloclair plus gel ®) followed by 5% EDS, EDS 1 % and negative controls. The wound healing process is a complex and dynamic process in the recovery of cell structures and damaged tissue layer to return to normal [21]. After injury, healing process and cell regeneration occurs automatically as a physiological response of the body [22,23]. The process of normal wound healing that involves a process that is interconnected to one another namely hemostasis, inflammation, proliferation and remodeling [23,24,25,26]. On the phase of hemostatic, platelets as the cells will close the injured blood vessels. Blood vessels vasoconstriction, while the platelets will facilitate forming blood clots in the blood vessels breaking [26,27]. Next on the inflammatory phase, neutrophils and macrophages as cells that are dominant in this phase will migrate to the wound area and phagocytic microorganisms and dead cells [28,29]. In the proliferative phase would have seen an increase in the number of cells and the factors wound healing, one of the fibroblasts. The number of fibroblasts can be regarded as parameters of wound healing [2]. Fibroblasts are the main elements in the phase of proliferation that appears the first time on the third day and reached a peak on the seventh day [30]. Fibroblasts will produce collagen which will link the wound, affect the process of reepithelization, migrate and proliferate to form connective tissue and new collagen synthesis that affects the tensile strength and strength on the place of wound healing [22,23,24,26,31]. The proliferation of fibroblasts determine the final outcome of wound healing [2]. Although the process of wound healing is a process that is natural and naturally possessed of living creatures, but to accelerate the process of wound healing is needed certain conditions that support the sustainability of the process of wound healing, one of which is nutrition [2,26]. In the treatment group, an increase in the number of fibroblasts due to the substances contained in the extracts of the leaves senggani (*Melastoma candidum* D. Don) have an influence in the process of wound healing [21,22,32]. As for polyphenolic compounds and flavonoids are known to have the ability as an antioxidant and antitiinfammatory [31]. The mechanism of flavonoids in inhibitiung the process of inflammatory through two ways, namely by inhibiting capillary permeability and inhibiting arachidonic acid metabolism and secretion of lysosomal enzymes from cells neutrophils and cells of the endothelial, inhibit the activity of the enzyme cyclooxygenase and lipooxygenase, inhibit the release of histamine and stabilize Reactive

Oxygen Species (ROS) [33,34]. Flavonoids also play an important role in maintaining the permeability and increase the resistance of capillary blood vessels. Therefore, the flavonoids used in a pathological state such as occurence of interference permeability the walls of blood vessels [33]. Flavonoids serve to increase the expression of receptors Insulinlike Growth Factor-1 (IGF-1) as a mediator of proliferation of fibroblasts and synthesis of collagen [35]. Saponins are compounds that can be used for healing wounds and stop the bleeding. Saponins have properties of precipitating and coagulating red blood cells [36]. Antibacterial effects of saponins plays a role in optimizing the formation of collagen the treatment group, by preventing tissue damage due to bacteria and their products. It can also stimulate the inflammatory response [37]. Tannins play a role in improving the attractiveness of the wound on the wound healing process. Tannins serve as an astringent that can cause shrinkage of skin pores, strengthen skin, stop exudates and bleeding that is light, so as to cover the wound and prevent the bleeding wich usually arises on wounds and accelerates ephithelization [21,22]. Steroids have properties astrigen and antimicrobial and has the activity of anti inflammatory, analgesic effects, which play a role in the wound healing process.The activity of the antiinflammatory can prevent the occurance of inflammatory prolonged, thereby accelerating the process of wound healing [38,39].

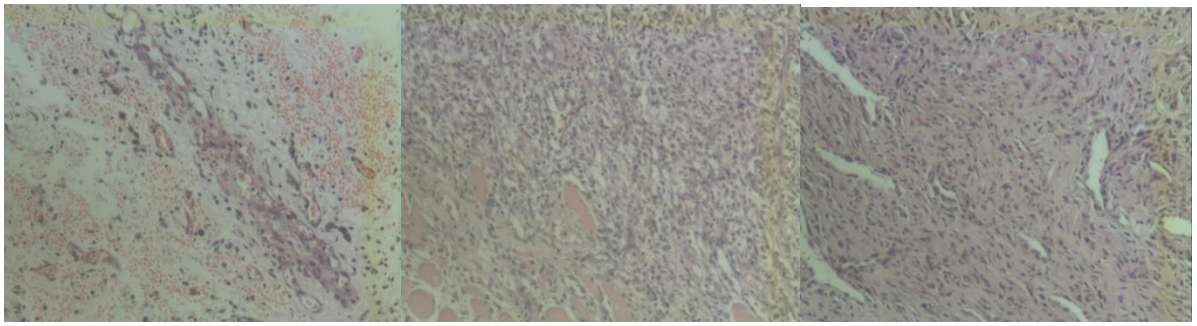

Figure 5.1: Negative control **Figure 5.2:** Positif control **Figure 5.3:** Gel EDS 1% group

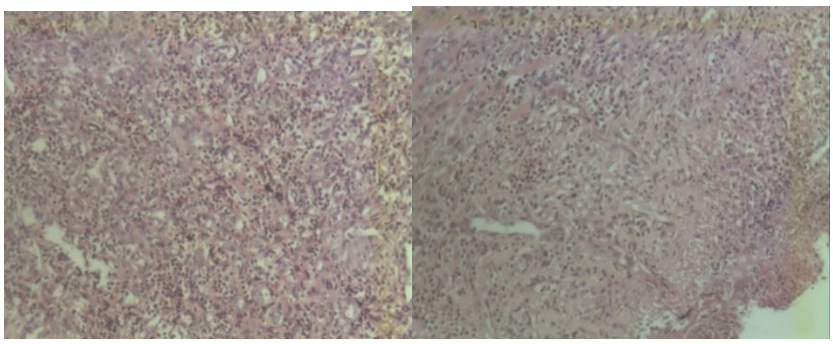

Figure 5.4: Gel EDS 5% group **Figure 5.5:** Gel EDS 10 % group

5. Conclusion

In the ethanol extract of senggani leaves (Melastoma candidum D.Don) contains chemical compounds such as glycosides, flavonoids/phenols, triterpenoids/steroids, tannins and saponins which have a function to accelerate

wound healing, namely antioxidant, antimicrobial, anti-inflammatory, analgesic and astringent. The ethanol extract of senggani leaves can be formulated in the form of gel preparations to accelerate socket wound healing after tooth extraction in male white rats and the most effective dose is 10% EDS compared to Aloclair Plus Gel ®, From the One Way ANOVA test results show that EDS Gel has effective significant for socket wound healing such as increased number of fibroblast cells and collagen fiber density scores on the 7th day. So the ethanol extract of senggani leaves (Melastoma candidum D. Don) has effectiveness on socket wound healing after tooth extraction in male white rats, with the most effective dose is 10% EDS gel, compared to positive controls (Aloclair Plus Gel ®), gel EDS 5% and EDS gel 1%, while negative controls (without treatment) showed slow healing.

6. Recommendation

1. Need to research more about the percentage of each component active substance contained in the extract of leaves senggani (Melastoma candidum D.Don).
2. Need to research more about the toxicity of gel of ethanol extract of leaves senggani (Melastoma candidum D.Don) to evaluate the limit of safety of use.

References

[1]. Nadia Farhatika, 2018.Efektifitas Gel Ekstrak Biji Kakao (Theoborma cacao L.) Terhadap Intensitas Kolagen Pada Penyembuhan Lika Jaringan Lunak Soket Pasca Pencabutan Gigi Tikus Wistar. Fakultas Kedokteran Gigi, Universitas Jember.

[2]. Oroh CG, Pangemanan DHC, Mintjelungan CN. Efektivitas lendir cekicot (Achatina fulica) terhadap jumlah sel fibroblas pada luka pasca pencabutan gigi tikus Wistar. J E-Gigi. 2015;3(2):515–20.

[3]. Nyoman Sidi Wisesa, 2017. Kombinasi Pasta Ekstrak Daun Jambu Biji (Psidium guajava linn.) 5% dan Povidon Iodine 10% Efektif Meningkatkan Jumlah Fibroblas dan Ketebalan Kolagen Pasca Pencabutan Gigi Marmut (Cavia cobaya), Program Magister. Program Studi Ilmu Biomedik, Fakultas Kedokteran Universitas Udayana, Denpasar.

[4]. Sjamsuhidajat R., Warko K., Theddeus OHP, Reno B. Buku Ajar Ilmu Bedah (3 rd ed.) Jakarta: EGC. 2012. 95-101.

[5]. Rahayu, Fitri., Wiwit Ade FW., Wiwik Rahayu. 2013. Pengaruh Pemberian Topikal Gel Lidah Buaya (Aloe cinensis Baker) Terhadap Reepitelisasi Epidermis pada Luka Sayat Kulit Mencit (Mus musculus).Universitas Riau. Pekanbaru.

[6]. ITD Kusumowati, Rosita Melannisa, Angga Prasetyawan. Daya Antibakteri Ekstrak Etanol Daun Senggani (Melastoma Affine D.Don) Fakultas Farmasi Universitas Muhammadiyah Surakarta, Agustus 2014: Vol 6(2): 20-25

[7]. Olivia Avriyanti Hanafiah, 2019. Perbedaan Ekspresi TGF B1 dan PDGF – BB sebagai Penanda Proliferasi Sel Fibroblas dengan Pemberian Gel Ekstrak Daun Benahong (Anredera cordifolia (Ten.) Steenis) dan Gel Aloclair R Pada Penyembuhan Luka di Mukosa Palatal Tikus Wistar, Program Doktor (S-3) Ilmu Kedokteran Gigi, Fakultas Kedokteran Gigi, Universitas Sumatera Utara, Medan.

[8]. Andi Irdam Hidayat. Uji Aktifitas Antimikroba Fraksi Ekstrak Daun Senggani (Melastoma affine

D.Don) Terhadap Mikroba Patogen. Fakultas Kedokteran dan Ilmu Kesehatan, Universitas Islam Negeri Alauddin. 2017; 7-9.

[9]. Backer, C.A. and Bakhuizen V.d. Brink, Jr. 1968. Flora of Java Volume 3. Netherlands: Netherlands Organisation for the Advancement of Research

[10]. Sentra Informasi IPTEK, 2009. Senggani (Melastoma Candidum D. Don) http://www.iptek.net.id/ind/pd_tanobat/view.php/mnv=2&id=156 [09April 2009]

[11]. Robertus Wandi, 2015. Efektifitas Gel Combustio Derajat II Ekstrak Etanol Daun Senggani (Melastoma malabathricum L.) Pada Tikus Jantan (Rattus nurvegicus), Program Studi Farmasi, Fakultas Kedokteran Universitas Tanjung Pura, Pontianak.

[12]. Sry Wardiyah, 2015. Perbandingan Sifat Fisik Sediaan Krim,Gel, dab Salep yang Mengandung Etil P – Metoksisinamat dari Ekstrak Rimpang Kencur (Kaempferia galanga Linn.). Fakultas Kedokteran dan Ilme Kesehatan, Program Studi Farmasi. UIN Syarif Hidayatullah, Jakarta.

[13]. Ulfa Zara Izzati, 2015. Efektifitas Penyembuhan Luka Bakar Salep Ekstrak Etanol Daun Senggani (Melastoma malabathricum L.) Pada Tikus (Rattus norvegicus) Jantan Galur Wistar. Program Studi Farmasi Fakultas Kedokteran, Universitas Tanjungpura, Pontianak.

[14]. Mappa T, Edy, J.H., Kojong, N. Formulasi Gel Ekstrak Daun Sasaladahan (Peperomia pellucida (L.) H.B.K) dan Uji Efektifitasnya Terhadap Luka Bakar pada Kelinci (Oryctolagus cuniculus). Jurnal Ilmiah Farmasi. 2013; 2(2); Hal 49-55.

[15]. Zulfah Al-Fa'izah, 2018. Efektifitas Gel Ekstrak Biji Kakao(Theobroma cacao L.) Terhadap Jumlah Sel Fibroblas Pada Soket Pasca Pencabutan Gigi Tikus Wistar Jantan. Fakultas Kedokteran Gigi, Universitas Jember.

[16]. Soekidjo Notoatmodjo, S.K.M., M.Com.H., 2018, Metodologi Penelitian Kesehatan, Jakarta. Rhineka Cipta. Cetakan ketiga.

[17]. Hafidhah, N., R.F.Hakim, dan Fakhurranzi, 2017. Pengaruh ekstrak biji kakao (Theobroma cacao L.) terhadap penumbuhan Enterococcus faecalis pada berbagai konsentrasi. Journal Canalis Dentistry. 2(2): 341 -343.

[18]. Isye Matiani, et.al., 2013. Laporan Praktikum Farmakognosi Penetapan Kadar Sari Dalam Pelarut Tertentu, Jurusan Farmasi Fakulktas Matematika dan Ilmu Pengetahuan Alam, Universitas Garut.

[19]. Fuadi, M.I., E. Ulfa dan Misnawi, 2015. Jumlah Fibroblas Pada Luka Bakar Derajat II pada Tikus dengan Pemberian Gel Ekstrak Etanol Biji Kakao dan Silver Sulfadiazine. E- Jurnal Pustaka Kesehatan. 3(2): 247-248.

[20]. Barua, N., Absar, N., Paul, S., Barua, A., Gazi, M.Y., Saha, M., Islam, M.S., Belaly, J.M., 2014. In Vitro Phytochemical, Cytoxicity and Mineral Composition Analyses of Micania cordata (Bumr.f.) B.L. Robinson Leaves. International Journal of Biosciences. **5(8)**: 154-160.

[21]. Dewi IALP, Damriyasa IM, Dada IKA. Bioaktivitas Ekstrak Daun Tapak Dara (Catharanthus roseus) terhadap periode epitelisasi dalam proses penyembuhan luka pada tikus wistar. Indonesia Medicus Veterinus 2013; 2(1): 58-75.

[22]. Ferdinandez MK, Dada IKA, Damriyasa IM. Bioaktivitas Ekstrak Daun Tapak Dara (Catharantus roseus) Terhadap Kecepatan Angiogenesis Dalam Proses Penyembuhan Luka Pada Tikus Wistar. Indonesia Medicus Veterinus 2013; 2(2): 180-190.

[23]. Nurcahaya MI. Pengaruh ekstrak etanol lidah buaya (Aloe vera) terhadap peningkatan jumlah fibroblas pada proses penyembuhan luka mukosa rongga mulut tikus (Rattus norvegicus) strain wistar.Skripsi. Surakarta: 2015. P. 35.

[24]. Guy Masir O, Manjas M,Putra AE, Agus S. Pengaruh cairan Cultur Filtrate Fibroblast (CFF) terhadap penyembuhan luka: penelitian eksperimental pada Rattus novergicus galur Wistar. Jurnal Kesehatan Andalas. 2012; 1(3): 112-117.

[25]. Glim JE, Egmond MV, Niessen FB,Everts V, Beelen RH. Detrimental dermal wound healing: What can we learn from the oral mucosa. International Jounal of tissue repair and regeration. 2013; 21(5): 648-660.

[26]. Guo S. Dipietro LA. Factors Affecting Wound Healing. Journal of Dental Research. 2010; 89(3): 219-229.

[27]. Orsted HL, Keast D, Lalande LF, Megie MF. Basic Principles of Wound Healing: An Understanding of the basic physiology of wound healing provides the clinician with the framework necessary to implement the basic principles of chronic wound care. Wound Care Canada 2011, 9(2): 4-12.

[28]. Velnar T, T Bailey, V Smrkolj. The wound healing process: an overview of the cellular and molecular mechanism. The Journal of International Medical Research 2009; 37(5): 1528-1542.

[29]. Suryadi IA, Asmarajaya AAGN, Maliawan S. Proses penyembuhan dan penanganan luka. Denpasar. P3-4.

[30]. Triyono B. Program perbedaan tampilan kolagen di sekitar luka insisi pada tikus wistar yang diberi infiltrasi penghilang nyeri levobupivakain dan yang tidak diberi levobupivakain. Tesis 2005. p. 25

[31]. Chairunnisa A. Pengaruh aplikasi ekstrak daun ceremai (Phyllanthus acidus (L.)) terhadap jumlah fibroblas pada hari ke-7. Skripsi. Banda Aceh: 2015. P. 29.

[32]. Nayak BS, Pereira LMP. Catharantus roseus flower extracthas wound-healing activity in Sprague dawley rats. BMC Complementary and Alternative Medicine 2006; 6(41): 1-6.

[33]. Fitriyani A, Winarti L, Muslichah S dan Nuri. Uji antiinflamasi ekstrak metanol daun sirih merah (Piper crocatum Ruiz & Pav) pada tikus putih. Majalah Obat tradisional 2011; 16(1): 34-42.

[34]. Hidayati NA, Listyawati S, Setyawan AD. Kandungan kimia dan uji antiinflamasi ekstrak etanol Lantara camara L. Pada tikus putih (Rattus novergicus L.) jantan. Bioteknologi 2008; 5(1): 10-17.

[35]. Priosoeryanto B, Huminto H, Wientarsih I, Estuningsih S. Aktivitas Getah Batang Pohon Pisang dalam Proses Persembuhan Luka dan Efek Kosmetiknya pada Hewan. Lembaga Penelitian dan Pemberdayaan Masyarakat. Institut Pertanian Bogor, 2006: 11(2): 70-73.

[36]. Harisaranraj R, Suresh K, Saravanababu S. Evaluation of the Chemical Composition Rauwolfia serpentine and Ephedra vulgaris. Advances in Biological Research, 2009; 3(5): 174-178.

[37]. Hasanoglu. Efficacy of Micronized Flavonoid Fraction in Healing of Clean and Infected Wounds. Medicine Oral, 2001; 10 (1): 41-44.

[38]. Dian NI. Isolasi dan elusidasi struktur senyawa turunan terpenoid dari kulit batang slatri (Calophyllum soulattri Burm f.) Skripsi. 2011. p. 13-14.

[39]. Simanjuntak MR. Ekstraksi dan fraksinasi komponen ekstrak daun tumbuhan senduduk (Melastoma malabathricum L.) serta pengujian efek sediaan krim terhadap penyembuhan luka bakar. Skripsi. Medam: 2008. p.6.