

Usage of Asian African Leaf (*Vernonia amygdalina*) As Analgesic during Post Tooth Extraction in Rabbit (*Rattus norvegicus*) In Vivo Gel Spray Form

Harun Achmad¹, Ali Yusran², Andi Nadwa Tenri³, Andi Esty Wijayanti⁴, Reski Amalia Rosa⁵

¹Department of Pediatric Dentistry, Faculty of Dentistry, Hasanuddin University, Indonesia

²Department of Oral Medicine Dentistry, Faculty of Dentistry, Hasanuddin University, Indonesia

³Clinical Dental Student, Faculty of Dentistry, Hasanuddin University, Indonesia

⁴Clinical Dental Student, Faculty of Dentistry, Hasanuddin University, Indonesia

⁵Pharmacy Student, Faculty of Pharmacy, Hasanuddin University, Indonesia

E-mail Correspondence: harunachmader@gmail.com

ABSTRACT

Background: Tooth extraction has a detrimental effects, namely disturbance in the masticatory function, unpleasant aesthetics, disturbed phonetics, temporomandibular dysfunction, pain, psychological problem and decreased level of trust and self confident these are the complications of the aforementioned problem. **Aim:** In treating the complication that is marked by pain in the patient, there needs to be analgesic after tooth extraction, however some people dislike the idea of drug consumption, especially if it has to be ingested orally. With that in mind, *Vernonia amygdalina* in gel spray form is made to help treat post tooth extraction pain. **Methods:** Four formulas were used, F1 (5% NaCMC), F2 (3% NaCMC), F3 (1% NaCMC) and F4 (carbopol) to evaluate the suitable formula for gel spray making, the evaluations are organoleptic testing, purity, viscosity, pH and spraying model. **Result:** Based on the evaluating data we conclude that F3 is the suitable formula. Then, it mixes with the 5% extract and we use rabbit that has been tooth extraction as the trial animal to see the analgesic effect of the extract and it considers with mefenamic acid as the positive controlled. **Conclusion:** The results of this study showed that Asian African leaf ethanol extract (*Vernonia amygdalina*) possesses an analgesic effect or relieved post-rabbit dental extraction given topically by spray gel preparation. Due to the presence of flavonoids, tannins, and saponins.

Keywords: Analgesic; Gel spray; Oral Surgery; *Vernonia amygdalina*

INTRODUCTION

Dental health problems in Indonesia are caused by poor oral hygiene, age-related periodontal problems, bone atrophy, tooth wear and smoking habits, resulting in damage to the dental supporting tissues. The result of the study shows that the most common tooth extraction is the first upper premolar teeth of 9.4% due to lack of prophylactic such as fissure sealants to protect teeth from caries, poor eating habits that consume candy overly, bad brushing teeth technic and lack of visits to the dentist [1].

Tooth extraction is a process of removing the tooth from the alveolar bone, the purpose is because of the tooth cannot be restored [2]. Because of the removal of tooth will cause injury to the surrounding tissue. Therefore, causing a reaction that is known as inflammation as the initial stage of wound healing. Inflammation due to tissue damage has several signs, that: tumor (swelling), rubor (redness), dolor (pain), heat (heat) and functio laesa (impaired function) [3]. In order to overcome the inflammatory reaction, especially the pain that arises doctor often prescribe NSAID medicine to overcome the pain. Mefenamic acid is the one of NSAID drug that is often used.

The drugs that are often prescribed is mefenamic acid, aspirin, paracetamol followed by antibiotics as anti-bacterial. Mefenamic acid is also one of the most common analgesics used at Dental Hospital post tooth extraction for clinical students [4]. These non-steroidal drugs (NSAID) have the ability to suppress signs and symptoms of inflammation that are the trigger factors of pain quickly, but often it has the harmful effects such as gastrointestinal, nephrotoxic, and hepatotoxic [5].

Based on empirical studies, the author found that the leaves of Asia African (*Vernonia amygdalina*) is one of the plants often used by people to recover the pain in the teeth, as well as other diseases. This has also been verified by a study which states that the Asian African leaf extract (*Vernonia amygdalina*) can accelerate wound healing [6]. This plant contains antioxidant that can accelerate wound healing and anti-inflammatory and anti-

bacterial effects. Therefore, a study of the analgesic effect of the leaves on dental pain that is caused by tooth extraction. In addition, the Asian African leaf extract will be formulated in gel spray form because by using of spray gel preparation will reduce contamination or infection and trauma of the patient during the use of dosage, then this also can increase the potential penetration of active ingredients into the tissue injured, the medication contact time of the wound is longer, and because it works locally it will not experience a first pass effect or through a GI solution. Because of these advantages, this study was done by formulating the Asian Africa leaf ethanol extract (*Vernonia amygdalina*) tested on in vivo experimental by using animals (rabbit) with post-extraction conditions to see the analgesic effect of the extract.

Botanical profile of Asian-African leaves (*Vernonia amygdalina*)

Taxonomi

Kingdom:	Plantae
Phylum:	Angiosperm
Class:	Eudicots
Order:	Asterales
Family:	Asteraceae
Genius:	Vernonia
Species:	<i>Vernonia amygdalina</i>



Fig 1. Asian-Africa leaf (*Vernonia amygdalina*) Adedappo, Adeolu Alex; et.al. (2014). *Anti-Oxidant, Anti-Inflammatory and Antinociceptive Properties of the Acetone Leaf Extract of Vernonia amygdalina*. Advanced Pharmaceutical Bulletin. Pp. 595.

Description:

The Asian African leaf in English is called the bitter leaf, in Malaysia it is called the South African Leaf, and in the local language the Nigerians call it *ewuro* (Yoruba), *editot* (Ibibo), *onugbu* (Igbo), *ityuna* (Tiv), *oriwo* (Edo) and *chusardoki* (Hausa) (Egedigwe, 2010).

Etnomedical

These Asian-African roots and leaves are used to treat fever, hiccups, and kidney problems (Burkill, 1985; Hamowia and Saffaf, 1994). Asian-African leaves are often used to treat tooth pain.

Gel Spray Form

Spray gel is a gel with two terms, namely gel or hydrogel which means an aqueous phase which has at least 10% to 90% of the weight of the preparation. The term spray consists of small or large liquid droplets applied through an aerosol applicator or spray pump (Kamishita, Takzo., Et.al., 1992).

This gel spray is applied using a spray technique which has the advantage of allowing the preparation to be delivered to the wound without contact with a cotton swab or swab, thereby minimizing waste, reducing contamination or infection and trauma to the patient. This gel spray preparation is given topically, so that it will not go through the first pass effect's process and can minimize the potential toxicity. In addition, the drug delivery system from the spray can increase the penetration of the polymer into the wound area, thereby increasing the potential for more efficient delivery of active substances. The delivery mechanism, under stress, is caused by a mechanical mechanism which will cause a decrease in the viscosity of the formulation when sprayed. After spraying, the viscosity of the gel will return to its original consistency because it is free from stress (Suyudi, 2014).

Material and Methods

Plant Collection and Extract Preparation

The Asia African Leaf (*Vernonia amygdalina*) were harvested from Dg. Tata region, Makassar on May 2018. These leaves are made using the simplisia process through wet sorting, weighing, washing, slicing, drying in simplisia oven, and dried sorting. Extraction process is done by using 1L 96% ethanol as solvent. The simplisia is macerated for 5 days. After 5 days, the extract is filtrated by using filtrate paper. Filtrate is evaporated by using rotary evaporator 50°C and obtained viscous extract which will be formulated.

The extraction of Asia Afrika leaf (*Vernonia Amygdalina*) were done on an are in Dg Tata region, the sorting process is the done with the purpose to eliminate dirt and any other contaminants or foreign bodies during the harvesting process. The aforementioned foreign bodies are moldy or broken leaves.

The washing process is done using flowing water. This process is odne in order to clean the contaminants such as dust on the leaves. However, before the washing process is done, a weighing process is done which resulted in a sample weight of 3 kg.

c



Fig. 2. Plant collection (*Dokumentasi pribadi*)

The slicing process is a process in which the samples are sliced into small parts In order to facillitate an easier drying process by reducing the surface area.

The drying process is done using 2 methods which uses sunlight with slow airing and using an oven with temperature of 45-50°C.



Fig.3. Drying using an oven

After the sample is dried in the oven for several hours in the simplisia cabinet and the received water content of 8,67%.

On figure 4.a is dired simplisia weighing 260 grams. On figure 4.b 50 gr where weighed using sieve shaker with the purpose of knowing the particle size of the simplisia itsel.



Fig.4. *Simplisia* weighing

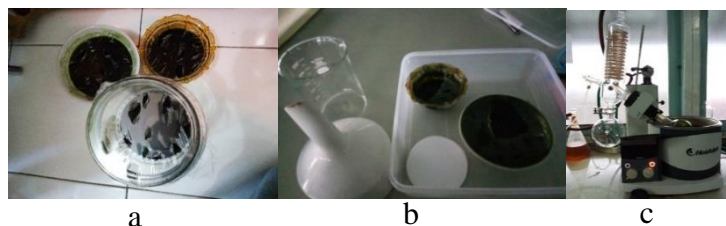
Figure 5.a shows that the used substances, which is 96% Ethanol and the simplisia which will be extracted using maceration procedure. Figure 5.b shows that an extraction process will be done. Moreover, the comparison between simplisia and the solvent being used is 110 mg for 1000mL of solvent and 100mg for 800mL.



Fig.5. Extraction process (*dokumentasi pribadi*)

Extraction and filtering process of the extract is done using filtering paper that has been shaped for the intents and purpose and also a funnel. After the filtration, a 1500mL of extract is received. Afterwards, the next step is evaporation. Evaporation is the next step after the extract has been filtered. This phase is done to evaporate the solvent from the extract. Moreover, the types of evaporation is by letting it sit and by giving it a gentle airing process.

Fig.6. Evaporation Process



Filtration is done again for the 2nd time when during the evaporation process is found to be inadequate. Therefore a re-filtration is done using filtering paper (fig.6.b). Evaporation is done using *rotary evaporator*. The principle of usage of the object is to reduce the pressure on the beaker and rotation of the beaker is done so that solvent can evaporate quicker below its boiling point (Fig.6.c)

Extract (Fig.7.a) is the result of manual evaporation, however it is still in the form of liquid extract, therefore a further evaporation is needed. Extract (Fig.7.b) is the liquid result of the rotary evaporator, therefore needs further evaporation.

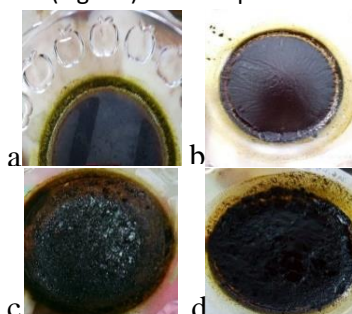
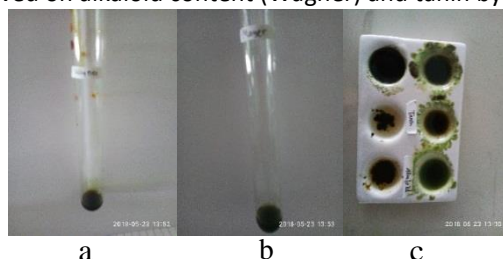


Fig.7. Extract result

A dried extract is received and then weighed in order to know the amount of extract received. On the 7.c container is the amount of extract which is : 355,4-346,2 gr = 9,2 gr. On the 7.d container the received extract is : yaitu : 355,4-346,2 gr = 9,2 gr.

Qualitative testing of Ethanol extract of *Vernonia amygdalina* using the tube method (Fig 8. A and b) and droplet tests (Fig.8.c). Fig.8.a is an alkaloid content test using Wagner Reagent and a positive result is achieved by the formation of a brown deposit or brick red colour ; On figure 8.c is a testing procedure with the droplet test and a positive result is achieved on alkaloid content (Wagner) and tanin by a change to a blackish colour.



Sieve shaker is done to know the size of the aforementioned simplisia. Sive Shaker consists of 2 sifters for the simplisia, which is number 14 and 18 sifters. Shaker is done for 10 minutes.



Fig.9. Shieve shaker (dokumentasi pribadi)

Formulation

The base formation using the F1 formula using 5% concentration of NaCMC. It can be perceived that the F1 base is incapable to be used as a gel spray because of the high viscosity. Base formation using the F2 and F3 of 3% and 1% respectively, can be seen that theres a difference in clarity between F2 and F3, in which F2 is slightly more feculent or cloudy in comparison with F3, this is due to the concentration difference between the NaCMC.

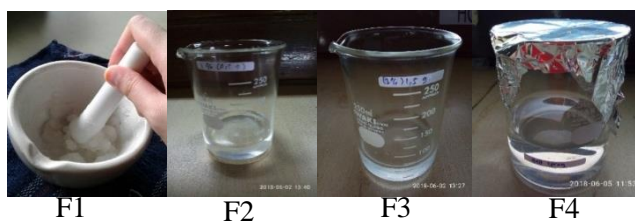


Fig.10. Formulation

The F4 fabrication use a base that consists of 0,4% carbopol for 17,5 g or each 100mL of water. The formation of gel is done using a homogenizer Ultra-Turrax with a working principle, of dispersion of 2 liquids is achieved by passing the mix through a small access hole with high pressure. In comparison between F1, F2, and F3 a clearer base is achieved by using carbopol.

A pH testing is done using a pH indicator. On figure 11.a is F2 NaCMC 3% has a pH of 6 ; on Figure 11.b is F3 NaCMC 1% has pH of 7, on Figure 11.c uses F4 Carbopol has a pH of 8 ; and figure 11.d is F1 NaCMC5% with a pH of 5. F2 and F1 has an acidic pH, therefore if its too acidic or it can cause pain and discomofort. Therefore F1 and F2 is not reccommended for usage in the creation of gel spray.

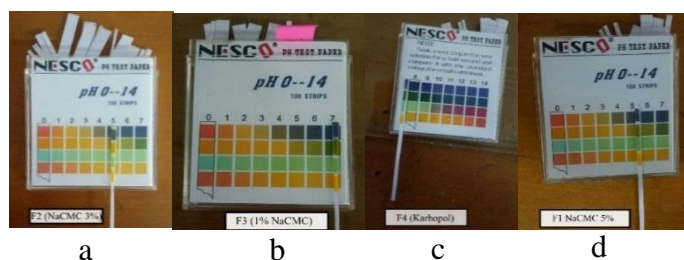


Fig.11.pH Testing.

A reagent making of 25 mL of positive control (+) by usign mefenamic acid that is dissolved on 1% NaCMC.



Fig.12. Making of positive control

Formulation is started by base making, firstly by using the 1st, 2nd and 3rd formula with a various NaCMC concentration as the polymer. The formula of gel spray can be seen in table 1.

Table 1: Formulation

Materials	F1	F2	F3	F4	F5	F6
Extract	-	-	-	5%	5%	5%
NaCMC	5%	3%	1%	5%	3%	1%
Glycerin	10%	10%	10%	10%	10%	10%
Propylene glycol	5%	5%	5%	5%	5%	5%
Methyl paraben	0.25%	0.25%	0.25%	0.25%	0.25%	0.25%
Aquadest ad	100%	100%	100%	100%	100%	100%

Formulation of gel spray base is done firstly, NaCMC is added in to aquadest on the mortar and it is crushed by using pestle until it is in a gel form. Propylene glycol that has been mixed with glycerin is added into the mortar and crushed until it reached homogenous form. Methyl paraben is added and crushed until it is homogenous. Each formula is evaluated including pH, viscosity, and spray model.

Physical Evaluation Base of Gel Spray

Physical evaluation includes organoleptic, pH, viscosity and spray model from base of gel spray that has been made.

1. Organoleptic
Organoleptic evaluation consists of smell, colour, and purity of the F1 – F3 base.
2. pH Value
pH of gel spray base is measured by using pH paper testing or pH meter.
3. Viscosity
Viscosity is measured by using Brookfield Viscometer in Pharmaceutics Laboratory, Hasanuddin University
4. Spraying Models
Spraying models is measured by seeing the spraying of every base by using sprayer.

Formulation of Gel Spray

Gel spray contains 5% extract of Asia Africa leaf. In the fabrication process, NaCMC (based on the concentration that want to make) and 100 mL water is added in to the mortar and it is mixed by using pestle until homogenous. 5g of Extract that has been dissolved in 5 g propylene glycol and 10 g glycerin are added in to the mixing and mix it until homogenous. Lastly, menthol 0.2 g and 0.25 g methyl paraben are added and crushed until homogenous.

Formula fabrication of the preparations starting from F1 to F4 added with 5% extract, afterwards an evaluation is done which consists of organoleptic, pH, Viscosity and spraying intensity.

Ethanol qualitative testing of Asian African leaf by using several specific reagents. Moreover the testing and reagents being used consists of : 1. Saponin = Several mL of Asian African leaf extract placed within the test tube and then 10 mL of aquadest is added and mixed for 1 minute, and then 2 drops of HCl is added. If the formed foam is stable for approximately 7 minutes, then the extract tested positive for saponin. 2. Flavonoid = Several ml of extract is inserted into the test tube, and a hot methanol is added, 0,059 of Mg powder and 1ml of concentrated HCl and then shaken vigorously. A positive test result is showed by the formation of red, yellow or orange colouration. 3. Alkaloids are analyzed using the Dragendorff reagent. 4-5 drops of Dragndorff reagents added on liquid extracts of Asian African leaf on the test tube containing 1ml of extract. A positive result is shown by the formation of red/orange deposit. 4. Flavonoid testing is done using FeCl₃ and the droplet plates.



Fig.13. Qualitative testing result

Treatment

The weighed rabbit weighs approximately 2 Kg each animal. Treatment groups are divided into 3 groups namely, the 1st treated using mefenamic acid as the positive control by oral administration (doses 50 mg/Kg BB); the 2nd treatment 5% extract Asian African leaf in gel spray form with 3 times of spray (based on the wound); the 3rd uses base of gel spray as negative control with 3 times of spraying (based on the wound). Treatment is applied for 2 days, while the observation is done for 3 days.

Tooth extraction is done on 3 rabbits, where the extracted tooth is a molar, premolar or anterior tooth. The extraction process is done on a Clinic ini Nuri Street, Makassar.



Fig.14. Tooth Extraction Process

On figure 15.a, the treatment is a post tooth extraction process on rabbits, which in this case is the negative control using a base of 1% concentration. On Figure 15.b the treatment post-extraction treatment uses 1% bases + 5% extract. Furthermore, on figure 15.c a post tooth extraction procedure using mefenamic acid which is dissolved on 1% of NaCMC. The application is done 1 hour after to extraction procedure.



Fig. 15. Treatment on test animals.

Result

Extraction

Total obtained extract is 18.7 g. Organoleptic of Asian African Leaf resulted in a dark green colouration with a distinctive odor.

Physical Evaluation Base of Gel Spray

The preferred spray gel base is the F3 sample because it fulfills the requirements of gel spray base compared with other formulas (Table 2). The difference lies in the viscosity and pH of each base formula due to the concentration of NaCMC that used.

Table 2: The Result of F1 – F3 Gel Spray Base

Evaluation	F1	F2	F3
Odor	-	-	-
Color	-	-	-
Clarity	+++++	+++	++
Viscosity	+80000 cPs	20000 cPs	1267 cPs
pH	5	5	7
Spraying models	No	Yes	Yes

Information:

Odor : (-) No odor

Color : (-) No color

Clarity : (+) Most clarity (++) Enough Clarity , (+++) not enough clarity, (+++++) No clarity, (+++++) No clarity.

Based on the evaluation results that have been done on F1, F2 and F3 have no odor and color. In the evaluation of clarity, the NaCMC-based formula is not very clear (cloudy), since NaCMC forms a colloidal solution in water that resulted in the gel to be less clear because it produces a colloidal dispersion in water characterized by the presence of gel spots (Rowe, et al., 2006). In F1, F2 and F3 on the NaCMC base have different clarity because of the various NaCMC concentration used. 5% NaCMC that is used in F1, 3% NaCMC that is used in F2, and 1% NaCMC that is used in F3. In the pH testing, pH 5 was obtained on F1 and F2, while in pH 7 was obtained on F3. In the evaluation of spraying models, only on F1 and F2 that can be sprayed. On the viscosity, F1 has the viscosity +80000 cPs, F2 has the viscosity 20000 cPs, and F3 has the viscosity 1267 cPs. Based on the requirement viscosity of gel spray base that 800 – 3000 cPs (Kamishita, Takuzo, 1992) and the F3 that has the viscosity which is accordance with the viscosity requirement. It was concluded that formula 3 with 3% NaCMC concentration with neutral pH according to the target organ, so the F3 is selected as the optimum formula as the base of gel spray.

Treatment

The results of treatment activity of gel spray asia afrika leaf extract as a post-extraction pain reliever compared with mefenamic acid (dose 50 mg / KgBB) as the positive control used (Table 3).

Table 3: The Result of Treatment Activity

Observation	1 st Day			2 nd Day			3 rd Day		
	Control (+)	Extract	Control (-)	Control (+)	Extract	Control (-)	Control (+)	Extract	Control (-)
Edema (swallen)	-	-	-	-	-	+	-	-	+
Rubor (Red)	++	++	++	+	+	+++	-	-	++
Fungtio laesa (appetite)	+++	+++	-	+++	+++	++	+++	+++	+++
Respon Nyeri (stretching response)	+++	+++	+++	+	++	+++	-	+	++

Information:

- Edema : (-) Not swollen; (+) slightly swollen; (++) swollen; (+++) big swollen
- Rubor : (-) not red; (+) slightly red; (++) red; (+++) very red
- Fungtio laesa : (1) does not have appetite; (2) lack of appetite; (3) good appetite
- Stretching : (-) no respond; (+) respond; (++) little response; (+++) very response.

The comparing of each treatment can see in the figure 16.

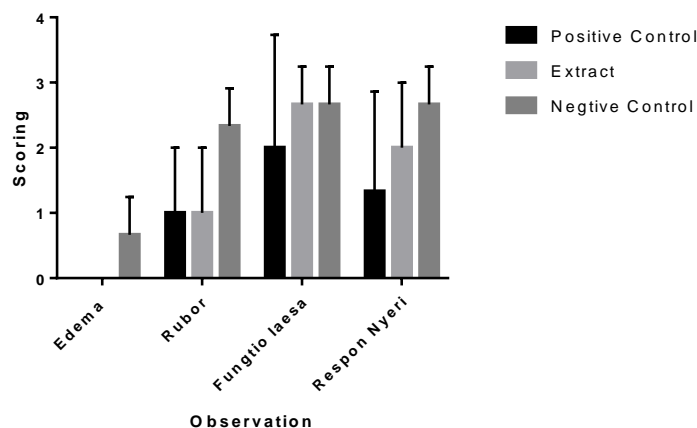


Figure 16: The graphic of each treatment

Result

Based on the result of observation of the treated animals observations of the edema, rubor (red), fungtio laesa (with parameters of appetite) and movement that occurs when pressure is given on the gums or the injured part.

On observation of edema, edema does not occur for animal given mefenamic acid (positive control) on day 1, to-2 and to-3. There were no differences with animals applied in the extract in treatment gel spray. However, in animals fed a base (negative control) are experiencing edema (swelling) on the 2nd day. The data shows that the leaf extract of Asian African have the potential to eliminate edema. It is supported by the results of a study that concluded that the Asian-African leaf extract to reduce the time of edema induced by xylene significantly ($p \leq 0.05$) (P.C. Adiukwu & dkk, 2014). As for the content of the Asian-African leaf extract that is used for the reduction of edema contains saponins. The occurrence of edema is dependent on the role of kinin and polimorponuclear leukocytes by a factor of its pro-inflammation including prostaglandins. This species of *Vernonia amygdalina* has that show inhibitory effects of prostaglandin E2 (PGE2) and prostaglandin D2 (PGD2) biosynthesis [10].

On rubor (red) observation for animal that is given mefenamic acid as the positive control on the 1st, 2nd, 3rd day changes of redness from day to day in accordance with healing conditions. This is the same case with what occurred in Asian African leaf extract, the same result is not shown animals that is fed a negative control of a base, where on the 2nd day of rubor scoring ,redness has increased. Rubor (redness) is one of the signs of inflammation, where blood is collected on the area due to the release of inflammatory chemical mediators (kinin and prostaglandins, histamine,) which resulted in a pinkish colouration. Based on the literature found that extracts Asian African leaf this has the effect of inhibiting the biosynthesis of prostaglandins [10].

Fungtio laesa is one sign of the onset of inflammatory, the observation fungtio laesa seen from the appetite of any animal attempting to correspond to the test and the data received that in animals treated with mefenamic acid as positive controls have a normal activity until the 3rd day. Whereas, in animals given the extract treatment have normal eating activities until the 3rd day. In animals applied negative control as base treatment on the 1st day shows no appetite or its eating activity is given a score of 1, 2nd day an improvement is shown and on the 3rd day appetite returend to an almost normal level. Loss of appetite is due to one of the tools of mastication has been removed so it will be pain during the mastication process which will interfere with the process of feeding.

On observation of when given the pressure on the sockets shows a response to pain. On the pain response scale in the grade of – 1 shows movement as a responses, but on the 2nd – 3rd day still shows a response. For animal that is the positive control they still shows a response but the response unlike in animals fed on the base. This is because the presence of compounds that can reduce pain for example the flavonoids compounds may inhibit enzymes that play a role in cyclooxygenase biosynthesis with prostaglandins as mediators of the formation of the pain, so it will increase the incidence of pain inhibition [11]

On Figure 16.a, it shows a rabbit tooth socket that was treated using mefenamic acid. On figure 16.b shows a rabbit socket that were treated with F3+extract dan on figure 16.c, shows the socket that were treated using the base (F3,1%)

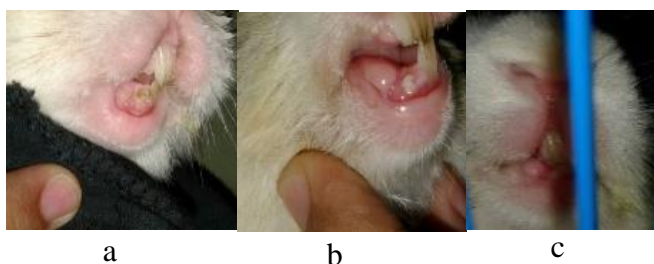


Fig.17. View of the socket on the 3rd day after the preparation application.

Histopathology

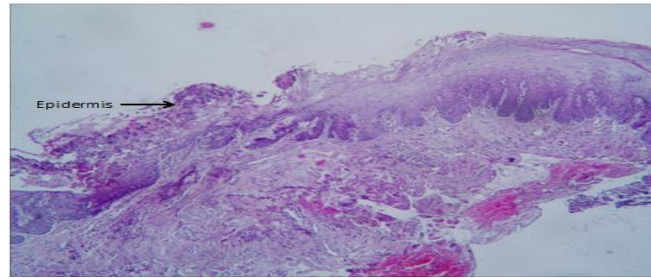


Fig.18. Application vernonia amygdalina, HE 40x. Epedermis contained infiltrated solid inflammation cells, the dermis and subcutaneous region contains solid inflammatory cells

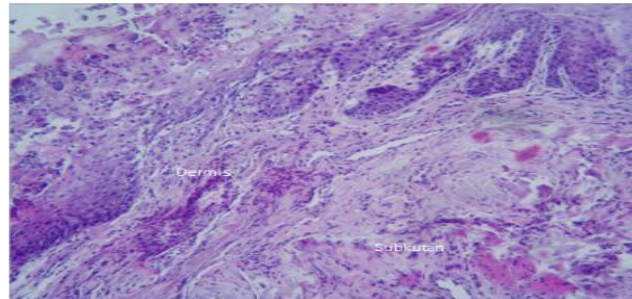


Fig.19. Application of vernonia amygdalina gel extract, HE 200x. Infiltration of inflammatory cells which is really solid dermis and subcutaneous level

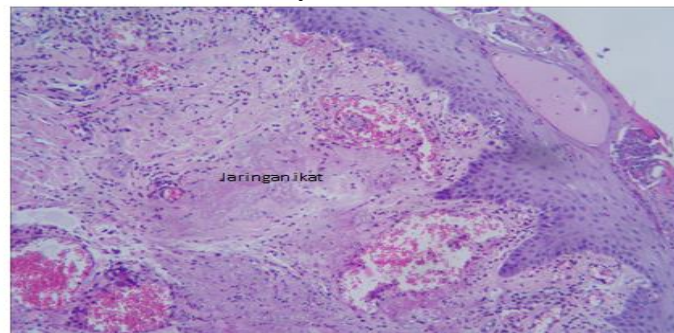
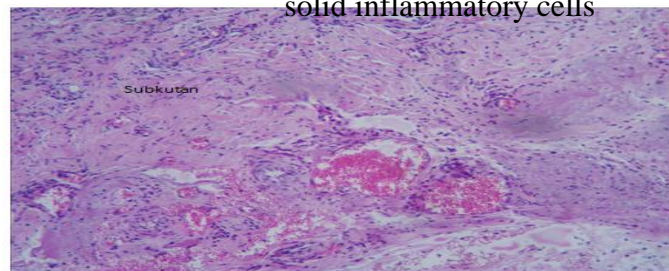


Fig.20. Without treatment, HE 200x. Fibrohyaline tissue is loose on the dermis containing solid inflammatory cells



- **Samples (Application of VA extract)**

Microscopic : Tissue preparation is layered with epidermis shows infiltrating lymphocytes and solid PMN Leukocytes. Superficial dermis reaching the subcutaneous muscle contains cells infiltrating lymphocyte

Fig.21. Without treatment, HE 200x. Subcutaneous region contains infiltrated solid inflammatory cells

cells and solid PMN leukocytes cells.

- **Samples (No treatment)**

Microscopic : Tissue preparation layered with hyperplastic epidermis and containing PMN Leukocyte infiltrators, on superficial dermis to the profunda dermis shows a loose fibrous connective tissue. Subcutaneous tissues contains inflammatory lymphocytes and solid PMN Leukocytes.

Discussion

The results of the study in positive control group, negative control and African leaf extract obtained comparison of pain parameters and socket condition on the 2nd day of rabbits shown in Figures 1, 2 and 3. In Figure 1 is a comparison between positive control (mefenamic acid) and African leaf extract shows that the effect shown is almost the same as on the second day as there is no visible edema, rubor (red) with a score of 1 states that there is still redness in the socket, *fungtio laesa* (disturbed function) with score 3 for both means that appetite of the animal shown to be normal, and stretching that occurs when the animal gums try to be pressed still respond with score 1 which means that there is still pain response in figure 2 is a comparison between negative control and extract of African leaf, obtained data differ significantly based on scoring of each parameter, on negative controls occur edema in the different sashes, red (rubor) on a negative control with a score of 3 which means redness on an animal given Asian african leaf extract, *fungtio laesa* (disturbed function) with a score of 2 on a negative control which means the animal is not so active in eating due to pain in the gums, and wincing or moving due to pain with a score of 2, which is higher than the African leaf extract which means there are still pain in the socket, so when given pressure they will respond in accordance with the feeling of pain they felt. Figure 3 shows a comparison of treatment results observed for up to 3 days and found that between positive control and African leaf extract gave almost the same effect in relieving pain after dental extraction.

Day-to-day pain decreases are marked by a decrease in response to the gums where tooth extraction and other signs of inflammation include rubor (red), edema (swelling) begins to decrease as shown in positive control data and Asian African Leaf extract. Based on the literature study which stated that it is due to the content of antioxidant compounds, anti-inflammatory antibacterial on Asian African Leaf extract. One of the antioxidants contained in Asian African leaf extract is the secondary metabolite of the flavonoid group [6]. Flavonoid compounds have similar activity to aspirin, which inhibits the formation of inflammatory mediators through inhibition of cyclooxygenase enzyme [7]. In addition, Asian African leaves have tannin content with analgesic and anti-inflammatory property through COX-1 inhibitors [8]. The content of saponins has a protective role for the treatment of post-extraction of teeth when the tooth is removed or removed from the socket will damage to the peripheral blood vessels, causing the blood to fill the deprivation area. Where, the compound is thought to accelerate the wound healing process by accelerating the precipitation and coagulation of red blood cells [9]

Conclusion

The results of this study showed that Asin African leaf ethanol extract (*Vernonia amygdalina*) has an analgesic effect or relieved post-rabbit dental extraction pain applied topically by spray gel preparation. Due to the presence of flavonoids, tannins, and saponins.

Reference

- [1].Sadiq D, Masound H, Ali D.dkk. (2015)Pattern of Thooth Extraction in Relation to Age and Sex in Patients Attended School of Dentistry of Sulaimani. *IOSR.JDMS*. vol 14(9), pp.101-2, 2015
- [2].Fithri, Zulfa. (2017, Jnuary). "Distribution of Tooth Extraction Based on Sociodemographic Characteristic of Dental Hospital of University of Jember Patients on January-December 2014". E-Journal Pustaka Kesehatan. Vol 5 (1). pp. 177-178.
- [3]. Bach T, Le Dds, and Woo I. Management of Complications of Dental Extractions. Penwell. 2008
- [4].Febriana, Posangi J, Bernart SP, Hutagalung. (2015). Test The Effect of Mefenamic Acid Administration Prior to Tooth Extraction on The Duration of Pain Threshold After Tooth Extraction. *Jurnal e-Gigi*. vol 3(2), pp.562.
- [5]. Achmad H, Djais AJ, Petrenko EG, Larisa V, Putra AP. 3-d printing as a tool for applying biotechnologies in modern medicine. *IJPR* 2020. 12(4), pp. 3454-3463.
- [6].Semiawan F, Ahmad I, Masruhim M.(2015). Inflammation Activity of Kareharu Leaf Extract. *Jurnal Sains dan Kesehatan*. Vol.1(1), pp.1-2.
- [7].Ruslim, A. K., Silvia & dkk.(2017). Effect of African Extract (*Vernonia amygdalina*) on Wound Healing Velocity Afrter Tooth Extraction in *Rattus norvegicus*. *Jurnal Sains dan Kesehatan*. Vol.1, pp. 409. (translate)
- [8].Middleton E, a. K. C.(1992). Effects of Flavonoids on Immune and Inflammatory Cell Function. *Biochemical Pharmacology*, pp. 167 - 1179.
- [9].Aremu, A. a.(2014). Anti-Inflammatory, Analgesic And Antioxidant Activities of Aqueous Leaf Extract of *Vernonia amygdalina* in Some Laboratory Animals. *Academic Journal of Science*, pp. 253 - 265.2014.
- [10]. Health Department of Republic Indonesia. Herbal pharmacopoeia Indonesia.2008. (translate)

- [11]. Adedappo, Adeolu Alex; et.al. (2014). Anti-Oxidant, Anti-Inflammatory and Antinociceptive Properties of the Acetone Leaf Extract of *Vernonia amygdalina*. *Advanced Pharmaceutical Bulletin*. Pp. 595.
- [12]. Afrianti, Ria, et al. (2014). Analgesic Activity Test of Leaf Ethanol Extract Papaya (*Carica papaya* L.) In Male White Mice. *Journal of Pharmaceutical & Clinical Science*.pp. 59.
- [13]. Djais Al, Achmad H, Dewiayu D, Sukmana BI, Huldani. 2020. Effect of Combination of Demineralization Freeze Dentin Matrix (DFDDM/0 and Moringa oleifera lam osteoprotegerin (OPG) and receptor activator of nuclear factor kappa Bligand (RANKL) as a marker of bone remodeling. *Sys Rev Pharm*. 11(6), pp.771-779.
- [14]. Adedapo, A. A., Aremu, O. J. & Oyagbemi, A. A., 2014. Anti-Oxidant, Anti-Inflammatory and Antinociceptive Properties of the Acetone Leaf Extract of *Vernonia amygdalina* in Some Laboratory Animals. *Advanced Pharmaceutical Bulletin*, Volume 4, p. 595.
- [15]. Achmad H, Djais Al, Syahrir S, Fitri A, Ramadhany YF. 2020. A literature us regarding the use of herbal medicines in pediatric dentistry. *IJPR*. 12,PP. 881-897.
- [16]. Ketno, Prichatin, A. & et al., 2008. Effect of Drying Time on Tannin Levels of Dutch Teak Leaves (*Guazuma ulmifolia* Lamk.). *Journal of Indonesian Medical Plant*, 1 (1), p. 41.
- [17]. P.C. Adiukwu, F. K. & dkk, 2014. Anti-Inflammatory and Anti-Pyretic Activity of the Leaf, Root and Saponin. *British Journal of Pharmacology and Toxicology*, Volume 4, p. 5.
- [18]. Price, S. A. & Wilsom, L. M., 2006. *Patofisiologi*. Indonesia: EGC.
- [19]. Sean C Sweetman, d., 2009. *Martindale: The Complete of drug Reference*. London: Pharmaceutical Press.
- [20]. Achmad H, Djais Al, Jannah M, Huldani, Putra AP.2020. Antibacterial chitosan of milkfish scales (*Chanos chanos*) on bacteria porphyromonas gingivalis and agregatibacter actinomycetescommitans. *Sys Rev Pharm*, 11(6), pp. 836-841.
- [21]. Shargel, L., 2005. *Biofarmasetika&Farmakokinetika Ed. 5*. surabaya: Aiirlangga University Press.
- [22]. Su'aida, N. & Destria, d., 2017. Optimization of Katsuri Fruit (*Magnifera casturi*) Ethyl Acetate Fraction Gel with NaCMC Base Combination and Kabopol Using the Simplex Design Method. *Pharmaceutical Science Today*, 1 (1).
- [23]. Suyudi, S. D., 2014. Spray Gel Formulation Using a Combination of Carbopol 940 and Hydroxypropyl Methylcellulose (HPMC) as a Gel Former. Jakarta: UIN Syarif Hidayatullah Jakarta.
- [24]. T. Le, D. M. a. I. W. M. D. M., n.d. *Management of Complications of Dental Extractions*. s.l.:ineedce..
- [25]. Udeme O Gergewill, O. A. G., 2009. Evaluation of Anti - Inflammatory Activity of Extract of *Vernonia Amygdalina*.
- [26]. Achmad H, Djais Al, Syahrir S, Fitria A, Ramadhany YF. 2020. Impact Covid-19 in pediatric dentistry: A literature review.*IJPR*,12,p.830-840.