ISSN (Online): 2455-3662
EPRA International Journal of Multidisciplinary Research (IJMR) - Peer Reviewed Journal
Volume: 9| Issue: 10| October 2023|| Journal DOI: 10.36713/epra2013 || SJIF Impact Factor 2023: 8.224 || ISI Value: 1.188

EFFECTIVENESS TEST OF GARLIC ETHANOL EXTRACT (ALLIUM SATIVUM) IN ACCELERATING WOUND HEALING AFTER TOOTH EXTRACTION

Tang Yaoping
Master of Dental Medicine Program, Faculty of Medicine, Dentistry, Health Sciences, Prima Indonesia University

ABSTRACT

One of the various plants in Indonesia that can be developed into traditional medicine is the garlic plant. Garlic has long been known by the public as a plant that has many benefits such as anti-inflammatory, anticancer, antioxidant, antiulcer, and antibacterial. The purpose of this study was to analyse the effectiveness of garlic extract (Allium sativum) for wound healing after tooth extraction. This laboratory experimental study used a complete randomised design with a post-test only control group design pattern. The experimental animals used in this study were 32 male Wistar rats, physically healthy, 2-3 months old, with body weight between 200-250 grams. The rats will be divided into two groups, namely 16 rats treated with 40% garlic extract (Allium sativum) and 16 rats treated with 80% garlic extract (Allium sativum) to see the comparison of accelerated wound healing after tooth extraction. The results showed a significant relationship between the number of fibroblast tissue per visual field in Wistar rats after tooth extraction with the administration of Garlic Extract (Allium sativum) 40% concentration and Garlic Extract (Allium sativum) 80% concentration, p=0.016 (p<0.05). Based on the results and discussion that has been done in this study, it can be concluded that garlic extract (Allium sativum) is effective in accelerating wound healing time after tooth extraction of Wistar rats.

KEYWORDS: ***Garlic, Wound Healing, Allium Sativum***

I. INTRODUCTION

Based on the results of the Basic Health Research (RISKESDAS) in 2007, the prevalence of tooth extraction in Indonesia is quite high at around 38.5% (1). The results of RISKESDAS in 2013 showed that the DMFT index of the Indonesian people nationally was 4.6 with the largest component being missing teeth at 2.9. This shows that the average Indonesian population has 3 teeth that have been extracted or are indications for extraction. Tooth extraction will cause injury in the form of exposed alveolar bone in the oral cavity (2). The severity of the wound depends on the amount of trauma received by the tissue. Physiologically, the body can repair damage to its own skin tissue (wound), known as wound healing (3). The wound healing process can be divided into three main phases, namely, the inflammatory phase, the proliferation phase and the remodelling phase. These phases continue from the onset of the wound until wound closure. The inflammatory phase is the body's reaction to the wound that starts after a few minutes and lasts about 3 days after the injury (4). The proliferation phase is characterised by the appearance of new blood vessels as a result of reconstruction and occurs within 3-24 days. The maturation phase is the final stage of the wound healing process. This process can take more than 1 year, depending on the depth and extent of the wound. The main cells involved in the wound healing process are fibroblasts. Fibroblasts are stem cells that form and lay down fibres in the matrix, especially collagen fibres. Herbal products have been used for a long time in the medical world. Nowadays, herbs are widely used for various treatments. The trend of back to nature lifestyle causes people to use natural medicines that are believed to have no side effects like chemical drugs and are more affordable than synthetic drugs (5). Of the many varieties of plants in Indonesia that can be developed into traditional medicine (6), one of them is the garlic plant. Garlic has long been known by the public as a plant that has many benefits such as anti-inflammatory (7), anticancer (8), antioxidant, antiulcer, and antibacterial (9). The purpose of this study was to analyse the effectiveness of Garlic (Allium sativum) extract for wound healing after tooth extraction.

II. RESEARCH METHODS

This experimental laboratory study uses a randomized controlled design with a post-test only control group design pattern. The experimental animals used in this study are Wistar rats, 32 males, physically healthy, 2-3 months old, with a body weight between 200-250 grams. The rats will be divided into two groups, namely, 16 treated with 40% Garlic extract (Allium sativum) and 16 treated with 80% Garlic extract (Allium sativum) to see the comparison of accelerated wound healing after tooth extraction. The sample size was determined by the Federer formula, namely: (t - 1) (r - 1) ≥ 15. Where t = several treatments; (2 treatments) r = several replications. Thus, the minimum sample size for each treatment was 16 rats.

$= (t-1)\ (r-1) \geq 15$
$= (2-1)\ (r-1) \geq 15$
$= (r-1) \geq 15$
$= (r-1) \geq 15$
$= r \geq 15 + 1$
$= r \geq 16$

ISSN (Online): 2455-3662
EPRA International Journal of Multidisciplinary Research (IJMR) - Peer Reviewed Journal
Volume: 9| Issue: 10| October 2023|| Journal DOI: 10.36713/epra2013 || SJIF Impact Factor 2023: 8.224 || ISI Value: 1.188

Tools

Tools used in research :

1. Number-coded experimental animal cages.
2. Diagnostic set (mouth glass, sonde, tweezers).
3. Nierbeken.
4. Dental extraction forceps (in this case a needle holder is used) under sterile conditions.
5. Syringe.
6. Gloves.
7. Mask.
8. Petri dish of jaw preparation.
9. A set of tools for making histology preparations.
10. Microscope.

Material

Materials used in the study:

1. Garlic extract (Allium sativum) Extract 50%
2. Garlic extract (Allium sativum) Extract 100%
3. Ketamine.
4. Formalin 10%.
5. Histology preparation material with Hematoxylin Eosin (HE) staining.
6. 70% alcohol as sterilization material.
7. Cotton pellet.

Data Type

The type of data collected in this study is primary data obtained from the results of measurements (scoring) on the histological picture of the process of accelerating wound healing after tooth extraction by administering Garlic extract (Allium sativum) 40% and Garlic extract (Allium sativum) 80%.

Extraction on Garlic extract (Allium sativum)

Collecting 3 kg of Garlic extract (Allium sativum), the Garlic extract (Allium sativum) was washed and divided into two parts to take the inner meat to obtain the gel. After washing, the flesh of the Garlic extract (Allium sativum) was dried in an incubator at 500 ℃ for 72 hours. The dried flesh of the Garlic extract (Allium sativum) was then pulverized using a blender until it became powder. Garlic extract (Allium sativum) meat that had become powder was then extracted by maceration while stirring. The extraction process uses a water solvent. The powder was put into a maceration vessel or container with a watertight lid and then filtered using filter paper; the pulp was macerated up to 2 times. The obtained maceration results were collected and evaporated using a rotary vacuum evaporator at a temperature of 500C until there was no more solvent condensation on the condenser. After the solvent was evaporated using a rotary vacuum evaporator, the evaporation was continued using a 70°C water bath to obtain a pure extract. The Garlic extract (Allium sativum) extract was then diluted with water to get 50% and 100% extract concentrations.

Treatment of Wistar Rats

1. Before treatment, 32 rats were divided into 40% Garlic extract (Allium sativum) extract and 80% Garlic extract (Allium sativum) extract. After that, all rats were adapted for one week. Then, animals were put into cages, with five rats in each cell in the same environmental conditions, given the same food, and monitored for health.
2. Rat tooth extraction will be performed using a modified needle holder under the anesthetic effect of ketamine 1000 mg/10 ml at a dose of 20 mg/kg bw intraperitoneally.
3. One incisor tooth will be extracted from every five rats daily.
4. After tooth extraction, observe the extraction wound and apply a tampon (cotton pellet) to stop bleeding in the wound for 5 minutes.
5. Dropped Garlic extract (Allium sativum) 40% in treatment group I and dropped Garlic extract (Allium sativum) 80% in treatment group II shortly after tooth extraction as much as 0.05 ml every day.
6. After extraction and treatment, the test animals (rats) were fed fine porridge with attention to the health of the test animals.
7. On the 5th day after tooth extraction, rats from each group were physically sacrificed by neck dislocation. The rat's tail was held and then placed on a surface it could reach. The rat will stretch its body; when the rat's body extends, a holder held by the left hand is placed on the nape of the neck. The right hand pulls the tail hard so the rat's neck will be dislocated. Then the jaw of the rat is taken out.
8. Then the tissue was fixed with 10% formalin for 24 hours at room temperature, then the decalcification process was carried out using Ethylene Diamine Tetra Acetic Acid (EDTA 10%) solution at room temperature.
9. Tissue dehydration was then performed using alcohol. First, the specimen was put into toluol alcohol solution (1:1) using pure toluol, then into a paraffin-saturated toluol solution.
10. The following process is infiltration in the oven by inserting the specimen into liquid paraffin.
11. The embedding process is carried out (inserting the tissue into paraffin) and then labeled/coded. After the embedding stage, the tissue is sliced in series with a thickness of approximately 6 microns using a microtome.
12. Evaluating fibroblast cell response using Hematoxylin Eosin (HE) staining. The procedure that must be done is deparaffinization using xylol and alcohol solution, then continued with the rehydration process with alcohol. After that, it is washed with running water, rinsed with distilled water, and then wiped. The glass slide was then placed in Meyer's hematoxylin solution, washed with running water, and then rinsed with distilled water, after which the staining was assessed under a light microscope. If the staining has been considered good, proceed to the next step, namely the dehydration process with alcohol in stages, and then wipe.
13. The next step, put it into xylol solution, and the object glass was covered with deck glass and observed using a light microscope.
14. Fibroblast density was assessed by counting the fibroblasts in 5 fields of view.

Histopathology Scoring Parameters for Fibroblast Counts

Histopathology scoring parameters to determine the distribution of fibroblast tissue is done based on the field of

ISSN (Online): 2455-3662
EPRA International Journal of Multidisciplinary Research (IJMR) - Peer Reviewed Journal
Volume: 9| Issue: 10| October 2023|| Journal DOI: 10.36713/epra2013 || SJIF Impact Factor 2023: 8.224 || ISI Value: 1.188

view is:

1. (-) = no fibroblast tissue found
2. (+) = small number of fibroblasts (less than 10% per field of view)
3. (++) = moderate amount of fibroblast tissue (10%-50% per field of view)
4. (+++) = large amount of fibroblast tissue (50%-100% per field of view) 4.

Data analysis using the SPSS 16 program. Research using a pure experiment with a non-parametric Chi-Square Test, after testing, showed that (p <0.05) means there is a significant difference between groups.

III. RESULTS AND DISCUSSION

Data distribution and frequency of the number of fibroblast tissue per field of view in Wistar rats after tooth extraction in groups given 40% and 80% garlic extract (Allium sativum) can be seen as follows:

Table 1. Distribution and Frequency Data of Fibroblast Tissue Counts Per Field of View After Tooth Extraction

Number of Fibroblasts	Garlic (Allium sativum)			
	Concentration 40%		Concentration 80%	
	n	%	n	%
No fibroblast tissue found	0	0	0	0
Small number of fibroblasts (less than 10% per field of view)	6	19	2	6
Moderate amount of fibroblast tissue (10%-50% per field of view)	6	19	5	16
Large amount of fibroblast tissue (50%-100% per field of view).	4	13	9	28

Table 1. it can be seen that all samples found fibroblast tissue in the administration of 40% and 80% garlic extract (Allium sativum) after tooth extraction of Wistar rats. The number of fibroblasts found in the small category (less than 10% per field of view) in the administration of 40% garlic (Allium sativum) extract after tooth extraction of Wistar rats as many as 6 (19%) and in the administration of 80% garlic (Allium sativum) extract as many as 2 (6%). The number of fibroblasts found in the moderate category (10%-40% per field of view) in the administration of garlic extract (Allium sativum) 40% after tooth extraction of Wistar rats as many as 6 (19%) heads and in the administration of garlic extract (Allium sativum) 80% as many as 5 (16%) heads. The number of fibroblasts found in the large category (40% - 80% per field of view) in the administration of garlic extract (Allium sativum) 40% after tooth extraction of Wistar rats as many as 4 (13%) heads and in the administration of garlic extract (Allium sativum) 80% as many as 9 (28%) heads.

Table 2: Relationship between the number of tissue fibroblasts per field of view in Wistar rats after tooth extraction with Garlic (Allium sativum) extract concentrations of 50% and 100%.

Jumlah Fibroblas	Garlic (Allium sativum)		p-value
	Concentration 40%	Concentration 80%	
1. No fibroblast tissue found	0	0	0,016*
2. Small number of fibroblasts (less than 10% per field of view) 3.	6	2	
3. Moderate amount of fibroblast tissue (10%-50% per field of view)	6	5	
4. Large amount of fibroblast tissue (50%-100% per field of view).	4	9	

Signifikan p<0,05. Uji Chi Square

From Table 2, it can be seen that there is a significant relationship between the number of fibroblast tissue per field of view in Wistar rats after tooth extraction by giving Garlic Extract (Allium sativum) with a concentration of 40% and Garlic Extract (Allium sativum) with a concentration of 80%, p = 0.016 (p <0.05).

Tooth extraction is the process of removing teeth, both whole and the remaining roots, from the alveolar because it cannot be treated anymore (Lande R et al., 2015). Tooth extraction will cause injury by exposing the alveolar bone in the oral cavity. The wound is anatomical damage or destruction of part of the tissue due to trauma (Sorongan et al., 2015). The body will repair tissue damage (harm), known as the wound healing process, and begins from the time of injury until wound closure (Novyana and Susianti, 2016). The primary cells involved in the wound-healing process are fibroblasts. The proliferation of fibroblasts determines the outcome of wound healing. This is because fibroblasts will produce collagen that will link the wound and affect the revitalization process that will close the wound (Masir et al., 2012).

This study aims to compare the effectiveness of 50% garlic

ISSN (Online): 2455-3662
EPRA International Journal of Multidisciplinary Research (IJMR) - Peer Reviewed Journal
Volume: 9| Issue: 10| October 2023|| Journal DOI: 10.36713/epra2013 || SJIF Impact Factor 2023: 8.224 || ISI Value: 1.188

extract (Allium sativum) and 100% garlic extract (Allium sativum) in accelerating wound healing time after tooth extraction of Wistar rats. The samples used in this study were Wistar rats. Wistar rats are known to have a physiological body similar to human physiology and have a short average age of 1-2 years, so it is appropriate to use it as an experimental object (Lailani et al., 2013). The number of research samples taken was 32 Wistar rats that were physically healthy and 2-3 months old with body weight between 200-250 grams. The samples were divided into two groups, namely 16 (50%) for the group treated with 50% garlic extract (Allium sativum) and 16 (50%) for the group treated with 100% garlic extract (Allium sativum). The results of this study are supported by Hendri Poernomo (2020), which states that the higher the concentration of garlic extract, the better the quality of the section and the higher the levels of active substances in the guinea pig gingival wound healing process (10). The results of Mufimah's research (2018) stated that the Kruskal Wallis and Anova one-way tests showed concentrations of 20%, 40%, and 80% sig values <0.05, namely 0.00. Therefore, it is concluded that 20%, 40%, and 80% garlic extract gel is effective in healing inflammatory wounds. Therefore, garlic extract gel is more effective in healing inflammatory injuries (6). Also supported by research by Barus and Lestari (2018), stating that garlic bulb extracts with a concentration of 15% is more effective in healing burns in rabbits than 5% and 10% concentrations, garlic bulb extract with a concentration of 15% is also more effective than shallot bulb extract with concentrations of 5%, 10%, and 15% (7). Likewise, Handayani's research (2019) states that the study results show that the provision of garlic extract affects the percentage of wound healing and survival of tilapia fish seeds (11). Garlic contains various vitamins, minerals, and other essential trace elements. Essential oils in garlic in organic sulfur are diallyl disulfide, diallyl trisulphide, and methyl allyl tri-sulfate. Allicin is formed from the amino acid allin, released when garlic is crushed, giving it a characteristic odor. It is believed that this compound is responsible for the pharmacological activity of garlic (12). This good immune state can improve the immune system's function, increasing proliferation (Putra, et al., 2013; Mohammed et al., 2015). Saponins are steroids or triterpenoid glycosides that are essential to human and animal health. Saponins can trigger vascular endothelial growth factor (VEGF) and increase the number of macrophages migrating to the wound area, thus increasing the production of cytokines that will activate fibroblasts in wound tissue. In addition, saponins will increase the action of TGF-β on fibroblast receptors, and TGF-β will stimulate fibroblast migration and proliferation (Putra, dkk., 2013).

IV. CONCLUSION

Based on the results and discussion that has been done in this study, it can be concluded that garlic extract (Allium sativum) are effective in accelerating wound healing time after tooth extraction of Wistar rats.

REFRENCES

1. *Adelia Dp. Efektivitas Gel Ekstrak Buah Andaliman (Zanthoxylum Acanthopodium Dc .) 10 % Terhadap Penyembuhan Ulkus Traumatikus Dina Priscila Adelia Fakultas Kedokteran Gigi Universitas Sumatera Utara. Fak Kedokt Gigi Univ Sumatera Utara Medan 2021. 2021;*
2. *Jane W, Bernat H, Wellsy L. Pengaruh Pemberian Ekstrak Biji Pinang (Areca Catechu L.) Terhadap Waktu Perdarahan Pasca Ekstraksi Gigi Pada Tikus Jantan Wistar (Rattus Norvegicus L.). J Ilm Sains. 2015;15(2):25–8.*
3. *Putri GA. Pada Penyembuhan Luka Soket Pasca Pencabutan Gigi Tikus Putih Galur Wistar (Rattus Novergicus) Secara Hematoxilin Eosin (He) Universitas Sumatera Utara Medan 2020. 2020;*
4. *Fitria V, Arifin RF, Kurniasih N. Uji aktivitas gel ekstrak daun pohpohan (Pilea trinervia W.) terhadap penyembuhan luka bakar pada kelinci (Oryctolagus cuniculus). Kartika J Ilm Farm. 2017;5(2):75.*
5. *Poernomo H, Ma'ruf MT. Pengaruh Gel Ekstrak Bawang Putih (Allium Sativum L.) Terhadap Jumlah Sel Makrofag Pada Penyembuhan Luka Insisi Gingiva Marmut (Cavia Porcellus). Interdental J Kedokt Gigi. 2020;16(2):34–9.*
6. *Mufimah M, Rusdian Hidayat U, Budiharto I. Efektivitas Gel Ekstrak Bawang Putih terhadap Proses Penyembuhan Luka Fase Inflamasi. J Vokasi Kesehat. 2018;4(2):109.*
7. *Barus BR, Lestari I. Pengaruh Ekstrak Umbi Bawang Putih Dan Umbi Bawang Merah Terhadap Luka Bakar Pada Kelinci. J Farm [Internet]. 2018;1(1):1–5. Available from: https://ejournal.medistra.ac.id/index.php/JFM/article/view/86*
8. *Prehananto H. Sitotoksisitas Ekstrak Etanol Bawang Putih Terhadap Sel Fibroblas Bhk-21. :117–23.*
9. *Puspa Dewi I, Verawaty, Taslim T. Efektivitas Gel Ekstrak Air Umbi Bawang Putih terhadap Penyembuhan Luka Bakar dan Luka Sayat. J Ilm Manuntung. 2020;6(2):215–22.*
10. *Hendri Poernomo. Pengaruh Gel Ekstrak Bawang Putih (Allium Sativum L.) Terhadap Jumlah Sel Makrofag Pada Penyembuhan Luka Insisi Gingiva Marmut (Cavia Porcellus). Interdental J Kedokt Gigi. 2020;16(2):34–9.*
11. *Handayani L, Siswanto S. Penggunaan ekstrak bawang putih untuk menanggulangi bakteri Aeromonas hydrophyla yang menyerang benih ikan Nila (Oreochromis niloticus). J Ilmu Hewani Trop … [Internet]. 2019;8(2):93–7. Available from: https://www.unkripjournal.com/index.php/JIHT/article/view/158*
12. *Yoga K. Fakultas kedokteran universitas wijaya kusuma surabaya tahun akademik 2020. 2020;(L).*