

Effectiveness Test of Turmeric Ethanol Extract (*Curcuma Longa*) in Accelerating Wound Healing after Tooth Extraction in Wistar Rats

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Abstract

Tooth extraction will cause a wound in the form of exposed alveolar bone in the oral cavity. The wound healing process can be divided into three main phases, namely, the inflammatory phase, the proliferation phase, and the remodeling phase. Curcumin is a bright yellow spice derived from the rhizome of *Curcuma longa* Linn. Scientific studies have shown the beneficial pharmacological effects of curcumin. This study aims to analyze the impact of administering turmeric (*Curcuma longa*) extract 35% with 65% in accelerating wound healing time after tooth extraction in Wistar rats. This experimental laboratory study uses a randomized controlled design with a post-test-only control group design pattern, March 2023. The experimental animals used in this study were Wistar rats, 32 males, physically healthy, 2-3 months old, with a body weight between 200-250 grams. There was a significant relationship between the number of fibroblast tissue per field of view in Wistar rats after tooth extraction by administering Turmeric Extract (*Curcuma Longa*) with a concentration of 35%, and Turmeric Extract (*Curcuma Longa*) with a concentration of 65%, $p = 0.002$ ($p < 0.05$). Conclusion Based on the results and discussions in this study, it can be concluded that turmeric extract (*Curcuma Longa*) effectively accelerates wound healing time after tooth extraction of Wistar rats. The more sections given increase the effectiveness of healing in the wounds of experimental animals.

Keywords: *Curcuma longa* Linn, Wound healing, Tooth Extraction

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I. INTRODUCTION

In dentistry, tooth extraction is one of the most common surgical procedures worldwide (1). The ideal tooth extraction is a painless extraction of one whole tooth or tooth root with minimal trauma to the supporting tissues of the tooth so that the extraction site can heal completely and there are no postoperative prosthetic problems in the future (2). Tooth extraction will cause a wound in the form of exposed alveolar bone in the oral cavity (3). Wounds are anatomical damage or destruction of some tissues due to trauma (4). Routine wound healing is a complex and dynamic process (5). The wound healing process can be divided into three main phases, namely, the inflammatory phase, the proliferation phase, and the remodeling phase (6).

The World Health Organization (WHO) recommends using traditional medicine, including herbs, to maintain public health and prevent and treat diseases, especially chronic diseases, degenerative diseases, and cancer (7). Herbal products have been used since ancient times in the medical world. Nowadays, herbs are starting to be widely used for various treatments. Modern research results also show that herbal medicines are effective for health and cause fewer side effects than chemical drugs (2); (8). The roots of *Curcuma longa* L. have been used as medicine for thousands of years. The plant has several pharmacological properties, including anti-inflammatory actions (9); (10). Curcumin is a bright yellow spice derived from the rhizome of *Curcuma longa* Linn. It has been shown that curcumin is a highly pleiotropic molecule that can be a modulator of intracellular signaling pathways controlling cell growth, inflammation, and apoptosis (11). Scientific studies have shown the beneficial pharmacological effects of curcumin. This study aims to analyze the impact of administering turmeric (*Curcuma longa*) extract 35% with 65% in accelerating wound healing time after tooth extraction in Wistar rats.

II. RESEARCH METHODS

This experimental laboratory study uses a randomized controlled design with a post test only control group design pattern, March 2023. 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 35% turmeric extract (*Curcumin Longa*) and 16 treated with 65% turmeric

extract (Curcumin Longa) to see the comparison of accelerated wound healing after tooth extraction. The sample size was determined by the Federer formula, namely: $(t - 1) (r - 1) \geq 15$. Where t = several treatments; (2 treatments) r = several replications. Thus, the minimum sample size for each treatment was 16 rats.

$$\begin{aligned} &= (t-1) (r-1) \geq 15 \\ &= (2-1) (r-1) \geq 15 \\ &= (r-1) \geq 15 \\ &= (r-1) \geq 15 \\ &= r \geq 15 + 1 \\ &= r \geq 16 \end{aligned}$$

Materials used in the study:

1. Turmeric (Curcumin Longa) Extract 30%
2. Turmeric Extract (Curcumin Longa) 60%
3. Ketamine.
4. Formalin 10%.
5. Histology preparation material with Hematoxylin Eosin (HE) staining.
6. 70% alcohol as sterilization material.
7. Cotton pellet.

Treatment of Wistar Rats

1. Before treatment, 32 rats were divided into 35% turmeric extract and 65% turmeric 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 turmeric extract (Curcumin Longa) 35% in treatment group I and dropped turmeric extract (Curcumin Longa) 65% 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 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%-35% per field of view)
4. (+++) = large amount of fibroblast tissue (35%-65% per field of view) 4.

Research Variables

The variables in this study consist of:

1. Independent variable: Turmeric Extract (Curcumin Longa) 35% and 65%.
2. Dependent variable: Wound healing process after tooth extraction.

Data Analysis Method

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

III. RESULTS AND DISCUSSION

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

NO	Number of Fibroblasts	Turmeric (Curcuma Longa)			
		Concentration 35%		Concentration 65%	
		n	%	n	%
1	No fibroblast tissue found	0	0	0	0
2	A small number of fibroblasts (less than 10% per field of view)	9	28	2	9
3	Moderate amount of fibroblast tissue (10%-35% per field of view)	4	16	7	19
4	A large amount of fibroblast tissue (35%-65% per field of view).	3	6	7	22

From Table 1. it can be seen that all samples found fibroblast tissue in the administration of turmeric extract (Curcuma Longa) 35% and 65% 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 turmeric extract (Curcuma Longa) 35% after tooth extraction of Wistar rats was 9 (28%) and in the administration of turmeric extract (Curcuma Longa) 65% was 3 (9%). The number of fibroblasts found in the moderate category (10%-35% per field of view) in the administration of turmeric extract (Curcuma Longa) 35% after tooth extraction of Wistar rats as many as 5 (16%) heads and in the administration of turmeric extract (Curcuma Longa) 65% as many as 6 (19%) heads. The number of fibroblasts found in the large category (35% - 65% per field of view) in the administration of turmeric extract (Curcuma Longa) 35% after tooth extraction of Wistar rats as many as 2 (6%) heads and in the administration of turmeric extract (Curcuma Longa) 65% as many as 7 (22%) heads.

Table 2. Relationship between the number of tissue fibroblasts per field of view in Wistar rats after tooth extraction with turmeric extract concentrations of 35% and 65%

Number of Fibroblasts	Turmeric (Curcuma Longa)		p
	Concentration 35%	Concentration 65%	
1. No fibroblast tissue was found	0	0	0,002*
2. A small number of fibroblasts (less than 10% per field of view)	9	2	
3. Moderate amount of fibroblast tissue (10%-35% per field of view)	4	7	
4. A Large fibroblast tissue (35%-65% per field of view).	3	7	

Significant $p < 0.05$. Chi Square Test

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 administering Turmeric Extract (Curcuma Longa) with a concentration of 35% and Turmeric Extract (Curcuma Longa) with a concentration of 65%, $p = 0.002$ ($p < 0.05$).

IV. DISCUSSION

TheThis study's results obtained a P-value of $0.002 \leq 0.05$, which states that there is an effect of giving turmeric extract with an accelerated healing process after extraction in experimental animals. The results of this study are supported by research by Budiman et al. in 2015 on the effect of Turmeric (Curcuma Longa) on incision wound closure time in mice (9). This study showed that Turmeric (Curcuma Longa) influences the closing time of incision wounds on the oral mucosa of Wistar rats. Wounds in Wistar rats given Turmeric (Curcuma Longa) closed faster than Wistar rats that were not given Turmeric (Curcuma Longa). Active substances such as mannose, glucomannan, chrysophane acid, acemannan, flavonoids, saponins, tannins, vitamin A, vitamin C, vitamin E, and enzymes contained in Turmeric (Curcuma Longa) are beneficial in the wound healing process (12); (13); (2).

Tannin contains astringents to stop bleeding, accelerate wound healing, reduce mucous membrane inflammation, and regenerate new tissue. In addition, tannin content has antibacterial ability (2). The tannin content accelerates wound healing by several cellular mechanisms, namely scavenging free radicals and reactive oxygen, enhancing wound closure, and increasing the formation of capillary blood vessels and fibroblasts (14). Flavonoids in Turmeric (*Curcuma Longa*) function as antioxidants, antimicrobials, and anti-inflammatories in wounds (15). Flavonoids can help wound healing by increasing collagen formation, reducing tissue edema, and increasing the number of fibroblasts (16). The examination results showed that the total flavonoid content in 65% Turmeric (*Curcuma Longa*) extract was 2.39%, and the entire flavonoid content in 35% Turmeric (*Curcuma Longa*) extract was 1.19%, so it was found that 65% Turmeric (*Curcuma Longa*) extract was more effective in accelerating wound healing.

From the results of this study, it can be seen that 65% Turmeric (*Curcuma Longa*) extract is more effective in the wound healing process than 35% Turmeric (*Curcuma Longa*) extract because the higher the concentration of the section, the content in the Turmeric (*Curcuma Longa*) extract is also higher so that the wound healing process is faster.

V. CONCLUSION

Based on the results and discussion that has been done in this study, it can be concluded that turmeric extract (*Curcuma longa*) is effective in accelerating wound healing time after tooth extraction of Wistar rats. The more sections given increase the effectiveness of healing in the wounds of experimental animals.

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