



Exploration of the ability of turmeric cream with ethanol extract in healing incision wounds in wistar rats

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Abstract

The shape of the wound is different depending on the cause; some are open and closed, and the healing consists of several phases, namely the inflammatory phase, proliferative phase, and maturation phase. Several sources state that turmeric rhizome extract is effective in wound healing. This study aimed to test the potential of turmeric rhizome extract to be effective in wound healing. This type of research is experimental with a pre-test and post-test group-control design approach, and it was conducted in January 2024. The samples used were turmeric rhizomes (*Curcuma Longa*) and male white rats. Determination of sample size according to Frederer's formula, so the number of pieces was 25 rats, the division of 4 treatment groups and one control group. The data will be analyzed for normality and continued with the ANOVA test. The results of turmeric extract contain chemical compounds of alkaloids, flavonoids, saponins, and tannins. Based on the research, turmeric ethanol extract (*Curcuma Longa*) with a 9% concentration showed optimal efficacy in healing incision wounds in white rats, reaching a comparable healing percentage of 93% on day 15 compared to the positive control, Bioplacenton®. The extract contains bioactive compounds like alkaloids, flavonoids, saponins, and tannins, contributing to its wound-healing properties. These results support the potential use of turmeric ethanol extract as an effective alternative in supporting the wound healing process.

Keywords: Wound healing phases, turmeric rhizome extract, experimental research, ANOVA test, bioactive compounds

Introduction

The forms of wounds vary depending on their causes, from open to closed injuries. An example of an open wound is an incision or cut impairment characterized by a linear tear in the skin and underlying tissues. Wounds, as common injuries experienced by every human being, involve the loss or damage of a part of the body tissues due to factors disrupting the body's protective system (Suarni and Prameswarie, 2015) [33]. Contributing factors include bites, accidents, sharp objects, gunshot wounds, and metallic objects (Afandi, 2017) [3]. The wound healing process is divided into several phases: inflammatory, proliferative, and maturation. The inflammatory phase is marked by hemostasis, chemotaxis, and increased blood vessel permeability that limits further damage, closes the wound, removes cellular debris and bacteria, and promotes cell migration. The duration of the inflammatory phase typically lasts for several days (Wissen *et al.*, 2020) [37]. The proliferative phase is characterized by forming granulation tissue, reepithelialization, and neovascularization, lasting several weeks. In the maturation and remodeling phase, the wound reaches maximum strength at maturity (Kartika *et al.*, 2015; Hernawati, 2015; Hardhani, Lastianny, and Herawati, 2014) [13, 10, 9]. Wound healing is the body's effort to restore its structural integrity and normal function after tissue disruption (Johnston, 2017) [12]. The healing process can be divided into three main phases: the inflammatory phase, the proliferative phase, and the remodeling phase (Novyana and Susanti, 2016) [19].

Turmeric (*Curcuma longa* Linn or *Curcuma domestica* Val), belonging to the Zingiberaceae family, has long been recognized by society for its numerous benefits, such as anti-inflammatory, anticancer, antioxidant, anti-ulcer, and

antibacterial properties (Purwaningsih, 2016) [24]. According to the study by Wientarsih *et al.* (2012) [36], turmeric rhizome extract is effective in wound healing (Wientarsih, Winarsih, and Sutardi, 2012) [36]. Supported by Yuniarto *et al.*'s research (2017) [39], their findings on the activity of ointment with turmeric as an active ingredient demonstrate its antimicrobial properties, capable of killing and inhibiting the growth of various fungi, bacteria, and viruses both *in vitro* and *in vivo* (Yuniarto, Lestari, and Winarso, 2017) [39]. This study aims to assess the potential effectiveness of turmeric rhizome extract in wound healing.

Research Methodology

This experimental research, with a Pre-test and Post-test group-only control design approach, was conducted in January 2024. The sample comprises turmeric rhizomes (*Curcuma Longa*) and male white rats. The sample size was determined using the Frederer formula, resulting in 25 rats divided into four treatment groups and one control group. The materials used include alcohol, aluminum foil, distilled water, Turmeric (*Curcuma Longa*), 96% ethanol, test animals (mus musculus rats), sterile gauze, Whatman filter paper, methylparaben, petroleum ether, plaster, propylene glycol, gloves, and triethanolamine. The equipment used includes glassware (pyrex®), an autoclave, a maceration container, a blender (Maspion®), a porcelain dish, calipers (Tricle brand®), oven, forceps, rotary evaporator (Heidolf®), iron spoon, analytical balance (Precisa®), and a water bath.

Turmeric (*Curcuma Longa*), identified and cleaned with running water, was then drained and spread on a matting paper until dry. The turmeric samples were weighed, dried, ground into powder, and formed into a simplicia (Kosasih *et*

al., 2019) [15]. A total of 25 rats were divided into five groups, each consisting of Group I, was given standard feed and applied with a 3% ethanol turmeric (*Curcuma Longa*) extract cream at 1g once every 24 hours. Group II was given a standard meal and applied with a 5% ethanol turmeric (*Curcuma Longa*) extract cream at 1g once every 24 hours. Group III was given standard feed and applied with a 7% ethanol turmeric (*Curcuma Longa*) extract cream at 1g once every 24 hours. Group IV was given a standard meal and applied with a 9% ethanol turmeric (*Curcuma Longa*) extract cream at 1g once every 24 hours. Group V, the positive control, was given standard feed and applied 1g of Bioplacenton® once every 24 hours. Inclusion criteria include white rats, male gender, aged 6-8 weeks, weighing 150-200g, and being healthy throughout the incision wound on the back, measuring 2 cm. Exclusion criteria included rats sick during the adaptation period, sick during the treatment, and dead during the treatment. Data processing techniques involved observing changes in the wounds and measuring the size of the injuries in the treated area. The data were then analyzed for normality, and ANOVA testing was performed.

Results and Discussion

a. Phytochemical Screening

Table 1: Turmeric Phytochemical Screening (*Curcuma Longa*)

Test	Results	Description
Alkaloids	Brown, red precipitate	(+)
	White precipitate	(+)
	Brown precipitate	(+)
Flavonoids	Red color on the layer of amyl alcohol	(+)
Saponins	Permanent foam	(+)
Tannins	Blackish-green color	(+)

Table 1 shows that turmeric extract (*Curcuma Longa*) contains chemical compounds of alkaloids, flavonoids, saponins, and tannins (Baud, Sangi, and Koleangan, 2014). Tannin compounds can act as astringents in wounds, while saponins work to increase the speed of epithelialization. Flavonoid compounds also play a role in wound healing by stopping bleeding through vasoconstriction mechanisms in blood vessels, free radical antidotes, hydrolysis and enzyme oxidation inhibitors, and anti-inflammatory (Soni and Singhai, 2012).

Table 2: Data on the percentage of inhibition of turmeric extract (*Curcuma longa*) against DPPH

Konsentrasi Ekstrak (ppm)	Absorbansi Ekstrak	Absorbansi Kontrol	Inhibisi (%)
3	0.236	0,343	36.31
5	0.228	0,343	37.68
7	0.219	0,343	37.74
9	0.178	0,343	63.44

Based on the table above, it can be seen that the extract activity inhibition test data showed variations in response to concentration, with four conditions tested at extract concentrations of 3 ppm, seven ppm, and nine ppm. The results demonstrated that the higher the attention of the extract, the higher the percentage of activity inhibition observed. At an extract concentration of 9 ppm, the highest rate of inhibition was 63.44%, indicating the potential of the

extract to inhibit certain activities. These data suggest that quotes at certain concentration levels can have significant inhibitory effects, and these results may form the basis for further research into the potential of compounds in such extracts as activity inhibition agents tested.

Table 3 provides insights into the varied responses of wound length changes over several days at different concentrations (3%, 5%, 7%, and 9%) and the use of Bioplacenton as a control. On the first day, all concentrations and the Bioplacenton control exhibited a 3 cm change in wound length. However, by the third day, each concentration showed a reduction in wound length, with the 9% concentration indicating the most significant change (1.5 cm). Throughout day 7, the 9% concentration maintained the most substantial change (1.2 cm), while other concentrations also displayed a decrease in wound length. This trend continued until day 15, with the 9% concentration consistently demonstrating the best results (0.5 cm). Bioplacenton, serving as the positive control, consistently exhibited lower changes in wound length. The data highlights the notably positive impact of the 9% concentration in expediting wound healing, presenting a potential area for further investigation compared to the Bioplacenton control. As a positive control, Bioplacenton® accelerated wound healing, evident in reduced wound length by day three and the highest percentage of recovery observed on day 15. This efficacy is attributed to Bioplacenton®'s composition containing active elements like placental extract and neomycin sulfate, stimulating new tissue formation and preventing infections in the wound area (Fitria, Arifin, and Kurniasih, 2017) [8]. Turmeric (*Curcuma Longa*) possesses wound-healing capabilities, albeit slower than Bioplacenton®, considering the day-to-day reduction in wound length. The healing potential may be linked to compounds within the extract, such as flavonoids, alkaloids, saponins, and tannins. Bioplacenton achieved the highest healing percentage at 93%, with a remaining wound length of 0.2 cm on day 15 from an initial 3 cm. It surpassed the 9% v/v concentration of turmeric extract (*Curcuma Longa*), followed by the 7% v/v concentration and subsequent concentrations. Following Indah's (2019) [11] study, an 8% dose of turmeric rhizome extract (*Curcuma domestica* Val.) ointment proved effective for incised wounds but fell slightly short compared to povidone-iodine cream (Indah and Br, 2019) [11]. The inflammatory phase spans from the wound's onset to approximately day 3 (Purnama, Sriwidodo, and Ratnawulan, 2017) [23], marked by the activation of platelets and hemostasis reactions in response to damaged blood vessels (Pebri, Rinidar, and Amiruddin, 2017) [21].

Table 3: Changes in wound length with varying concentrations of extracts

Day To	Change in Wound Length (cm)				Bioplac enton
	Concentra tion 3%	Concentrati on 5%	Concentra tion 7%	Concentra tion 9%	
1	3	3	3	3	3
3	1.5	1.5	1.5	1.5	1.5
5	1.6	1.5	1.5	1.5	1.3
7	1.3	1.2	1.2	1.2	0.7
9	1.2	0.8	0.8	1	0.7
11	1	0.3	0.3	0.5	0.6
14	0.7	0.7	0.6	0.5	0.2

Table 4: Results of the Test on the Influence of Extract Administration on Incision Wound Healing

ANOVA						
		Sum of Squares	df	Mean Square	F	Sig.
Turmeric Extract	Between Groups	6.634	3	3.245	7.112	.004
	Within Groups	37.232	136	.367		
	Total	52.856	139			

Table 4. The analysis of variance (ANOVA) results indicates a significant difference in response to turmeric extract at a significance level of 0.05. In the ANOVA table, "Between Groups" refers to the variation between groups (turmeric extract with different concentrations), while "Within Groups" reflects the variation within the groups. The F value of 7.112 indicates that the variability between groups is more remarkable than within groups, signifying a significant difference in the impact of turmeric extract at different concentrations on the observed response. The p-value (Sig.) of 0.004 is less than 0.05, thus concluding that the difference between groups is statistically significant. Therefore, it can be concluded that turmeric extract at different concentration levels has a different impact on the measured response, and this difference is statistically significant.

The above table shows the calculated F value of 7.112. To find the value in the F Table for $df = 3/136$ with a probability (α) of 0.03, the F-table value is 2.67. Hence, the calculated F value > F-table indicates a significant overall influence of turmeric extract (*Curcuma Longa*) on wound healing incisions. To strengthen this hypothesis test, observe the calculated Sig. Value of 0.004, whereas the Sig (α) value is 0.05, meaning the calculated Sig. < Sig (α). This implies a significant influence of turmeric extract (*Curcuma Longa*) on incision wound healing in rats (Nuryadi *et al.*, 2017).

Table 5: Test Results of Bioplacenton® (Positive Control) Effect on Wound Length

ANOVA					
	Sum of Squares	Df	Mean Square	F	Sig.
Between Groups	11.726	3	1.987	12.223	.001
Within Groups	1.702	136	.061		
Total	22.428	139			

Based on Table 4.5, it can be observed that the results of the analysis of variance (ANOVA) indicate a significant difference between groups in the measured response, as indicated by the considerable p-value (0.001). In the ANOVA table, "Between Groups" describes the variation between groups (in this context, possibly treatment groups or groups with different conditions), while "Within Groups" reflects the variation within those groups. The F value of 12.223 indicates that the variability between groups is more remarkable than within groups, signifying that the difference between groups is statistically significant. Therefore, it can be concluded that there is an influence from the tested factor (possibly treatment or different conditions) on the observed response. At a significance level of 0.05, these results support the existence of a significant difference between groups in the measured response.

From this data, it can be concluded that the administration of Bioplacenton® significantly affects wound healing incisions. The wound-healing process requires proper management and treatment to prevent the wound area from becoming infected and ultimately causing chronic wounds

(Aminuddin *et al.*, 2020) [4]. Wound healing is a complex biological process resulting in the restoration of integrated tissue. Physiologically, the wound healing process can be divided into four sequential stages: hemostasis, inflammation, proliferation, and tissue remodeling. Many factors slow down wound healing, such as poor nutrition, hypoxia, immunosuppression, chronic diseases, and post-surgical conditions. Surgeons must understand the physiological processes involved in wound healing to minimize patient morbidity from delayed wound-healing procedures (Phillips, 2000) [22]; (Young and McNaught, 2011) [38].

Wound healing is a natural repair process for tissue injuries involving inflammatory mediators, blood cells, extracellular matrix, and parenchymal cells. The process consists of three phases: hemostasis and inflammation, proliferation, maturation, and tissue remodeling. Any trauma causing vascular injury will initiate cellular responses to start the hemostasis phase. This phase occurs from the onset of the wound until approximately the fifth day. The main contributions are vasoconstriction, platelet aggregation, and fibrin deposition, leading to blood clot formation. Meanwhile, inflammation reactions such as erythema, edema, pain, and heat occur to bring inflammatory cells to the wound area (Phillips, 2000) [22]; (Young and McNaught, 2011) [38].

Conclusion

Based on the results of research and data analysis regarding the effectiveness of turmeric ethanol extract (*Curcuma Longa*) and Bioplacenton® on wound healing in white rats, it can be concluded that turmeric ethanol extract contains bioactive compounds such as alkaloids, flavonoids, saponins, and tannins that play a role in the wound healing process. The optimum concentration of turmeric ethanol extract for healing incision wounds in white rats was 9%. The highest percentage of cure on day 15 was recorded in the positive control group (Bioplacenton®) at 93%, followed by turmeric extract at a concentration of 9% v / v. Turmeric ethanol extract cream preparations showed healing abilities close to Bioplacenton® in the context of wound healing in rats. These findings support the potential use of turmeric ethanol extract as an effective alternative in supporting the wound healing process.

Suggestions for future research include quantitative phytochemical analysis to obtain in-depth information about the content of bioactive compounds in turmeric ethanol extract (*Curcuma longa*) and determine their relative concentrations. Quantitative analysis can provide an accurate picture of the amount of certain compounds in the section, which can be linked to the effectiveness of wound healing. It is also recommended to involve histopathologic testing to understand tissue structure and composition during the wound healing process, specifically changes in fibrin and collagen. Integrating phytochemical and histopathological analysis can provide a comprehensive

understanding of the wound-healing mechanisms of turmeric extract.

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