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Research Paper

Therapeutic Efficacy of Brotowali Ointment for Diabetic Burn Wound Healing in Streptozotocin-nicotinamide-induced Rats

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ABSTRACT

Introduction: Diabetes mellitus (DM) is a metabolic disorder characterized by hyperglycemia and is caused by factors such as genetics, obesity, improper diet and sleep patterns, stress, and lack of physical activity. One of the most common complications of DM is diabetic wound. The Dayak Ngaju people in Central Kalimantan use brotowali (*Tinospora crispa* L.) to treat skin diseases. The aim of this study is to pharmacologically assess the capacity of brotowali stem ointment in treating diabetic burn wound.

Materials & Methods: Diabetic rats were divided into six groups, each containing five rats, and wounds were created on their dorsal regions. The first group received oral brotowali extract and no topical treatment. The second group received oral brotowali extract and topical Betadine ointment. The third group received oral brotowali extract and topical base ointment. The fourth group received oral brotowali extract and topical 20% brotowali ointment. The fifth group received oral brotowali extract and topical 35% brotowali ointment. The sixth group received oral brotowali extract and topical 50% brotowali ointment. Diabetic burn wound healing was assessed based on wound scores, which were evaluated macroscopically by observing the color and the amount of scab detachment.

Results: The average wound scores at the end of the observation period indicated that all groups have a lower average wound score. Moreover, group K6 had a lowest average wound score ($P<0.05$).

Conclusion: The 20% brotowali extract ointment demonstrated a healing capacity for diabetic burn wound comparable to that of Betadine ointment. Furthermore, the 35% and 50% brotowali ointments exhibit superior healing capabilities.

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1. Introduction

Diabetes mellitus (DM) is a metabolic disorder characterized by hyperglycemia [1] and is caused by factors such as genetics, obesity, improper diet and sleep patterns, stress, and lack of physical activity [2]. DM has long-term health impacts and, if not properly managed, can lead to chronic complications [3]. One of the most common complications of DM is diabetic gangrene [4]. Diabetic gangrene is a chronic wound resulting from excessive infection and inflammation in patients with DM [4], often occurring on weight-bearing areas such as the lower extremities [3]. Infection arises due to delayed wound management caused by diabetic angiopathy and neuropathy [3, 4], which can even extend to deeper tissues, including subcutaneous tissue, tendons, muscles, bones, and joints [3]. Many cases of diabetic gangrene can lead to amputation [3], but this risk can be minimized with appropriate and intensive wound care [4].

The evolving principle of wound care is moisturizing dressing, which involves maintaining moisture around the wound, often through the use of topical ointments. Ointments create a moist environment at the wound site, thereby enhancing epithelial tissue development and migration, which accelerates wound healing [5]. Concurrently, advancements in the healthcare field have led to numerous efforts to develop the most effective medications for wound management [5]. At the same time, the development of chemical-based drugs and traditional plant-based remedies has proven beneficial in wound healing [3]. The active compounds in plants have motivated researchers to develop plant-based medications, including ointments [3].

Brotowali (*Tinospora crispa* L.) is a plant from the family Menispermaceae, native to Asia [6], including Indonesia [7]. Traditionally, brotowali has been used by various communities to treat a range of ailments [6]. The Dayak Ngaju people in Central Kalimantan use brotowali to treat skin diseases, rheumatism, and jaundice [6], while people in Bali use it for diabetic ulcers [8]. Scientific studies have shown that brotowali leaves can cleanse the digestive tract [9], while the stems can treat diabetes [1] and malaria [10]. Phytochemical studies reveal that brotowali leaves contain alkaloids, saponins, tannins, an d flavonoids [9]. Additionally, literature studies indicate that brotowali stems possess anti-inflammatory [11] and antioxidant effects [7].

Traditionally, various plants have been used to treat diabetic wound, including soursop (*Annona reticulata*), pegagan (*Centella asiatica*) [7], guava (*Psidium guajava*), bitter melon (*Momordica charantia* L.), and combinations of brotowali (*T. crispa* L.) with ginger (*Zingiber officinale)*, black pepper (*Piper nigrum*), and calamus (*Acorus calamus*) [8]. Scientifically, plants known to treat diabetic wound include bidara upas (*Merremia mammosa* [Lour.]) [12]. The healing efficacy for diabetic wound is supported by flavonoid compounds that act as antibacterial agents [12], antioxidants [12], and anti-inflammatory agents by stimulating macrophage growth [12], modulating cytokines [13], and increasing lymphocyte count [12]. Additionally, in treating diabetic wound, phenolic compounds serve as antimicrobial agents, while alkaloids, saponins, tannins, and steroids play roles in stimulating cell proliferation, migration, and skin fibroblasts [13]. Thus, the aim of this study is to pharmacologically assess the capacity of brotowali stem ointment in treating diabetic burn wound in streptozotocin-nicotinamide induced rats.

2. Material and Methods

2.1. Materials

The study utilized brotowali stem, 96% ethanol, vaseline album, adeps lanae, streptozotocin (Cayman, USA), nicotinamide (Nacalai, Kyoto), 30 male white rats (200-250 g), and Betadine ointment (PT. Mahakam Beta Farma).

2.2. Extraction

The brotowali stems were obtained from Sanggau and Mempawah, West Kalimantan, Indonesia. The extraction process was performed using the maceration method as described by Panjaitan et al. [14] Fresh brotowali stems, weighing 13.5 kg, were cut, cleaned, and dried. Extraction was carried out with 96% ethanol for 24 hours and was repeated three times. The resulting macerate was concentrated, yielding 172.52 grams of thick extract with a yield of 1.28%. The yield calculation using the following formula:

2.3. Qualitative phytochemical analysis

Qualitative phytochemical testing of brotowali extract was conducted at the Chemistry Laboratory, Faculty of Mathematics and Natural Sciences, Tanjungpura University, Pontianak, Indonesia. The secondary metabolites tested qualitatively included alkaloids, flavonoids, saponins, terpenoids, steroids, and tannins. The results were reported in a certificate No. 022/LABKIM/XI/2023.

2.4. Experimental animals and ethical statement

A total of 30 male white rats (*Rattus norvegicus*), two months old and weighing 200-250 grams, were obtained from the Food and Nutrition Study Center, Gadjah Mada University, Yogyakarta, Indonesia. Prior to testing, the rats were acclimated for seven days and provided with standard food and water ad libitum. The use of experimental animals and the study procedures were approved by the Faculty of Health Sciences, Universitas Respati, Yogyakarta, Indonesia.

2.5. Formulation and preparation of ointment

The formulation of brotowali extract ointment follows the method described by Panjaitan et al. [14]. The ointment was prepared in three concentrations: 20%, 35%, and 50%. The ointment base used consisted of 15 g of adeps lanae and 85 g of vaseline album. The preparation process adhered to the method outlined by Maru and Lahoti [15]. The ointment was made using a heated mortar and pestle pre-warmed in an oven at 70 ˚C. After removal from the oven, the mortar and pestle were used to mix the adeps lanae, vaseline album, and brotowali extract until a homogeneous formulation was obtained.

Brotowali ointment (*T. crispa* L.) 20% concentration

Brotowali stem extract: 1 g

Ointment base: 4 g

Ointment formula: 5 g

Brotowali ointment (*T. crispa* L.) 35% concentration

Brotowali stem extract: 1, 75 g

Ointment base: 3, 25 g

Ointment formula: 5 g

Brotowali ointment (*T. crispa* L.) 50% concentration

Brotowali stem extract: 2, 5 g

Ointment base: 2, 5 g

Ointment formula: 5 g

2.6. Evaluation of the healing capacity of brotowali ointment for diabetic burn wound (*T. crispa* L.)

The rats were fasted for 8-12 hours, after which their initial blood glucose levels were measured. They were then induced with streptozotocin-nicotinamide (STZ-NA) at doses of 45 mg/kg and 110 mg/kg body weight, respectively, via intraperitoneal injection [16]. Nicotinamide induction was administered 15 minutes before streptozotocin induction. The STZ-NA induction was designated as day 0. After 72 hours (day 3), blood glucose levels were checked again; rats with glucose levels >200 mg/dL were considered diabetic [16]. Once diabetes was confirmed, wounds were created on day 4. The wound creation procedure followed the method described by Tumigolung et al. [17]. First, a 3×3 cm area on the rats' backs was shaved and disinfected with 70% alcohol. A metal with a diameter of 2 cm was then heated for 3 minutes and applied to the rats' backs for 10 seconds. The wounds were left for 4 days (up to day 8), and wounds observations began on day 8 as an initial wound assessment.

Subsequent observations were conducted until day 22. On day 8, the diabetic burn wound rats were divided into six groups, each consisting of five rats. The first group (normal control) received brotowali extract orally at a dose of 450 mg/kg without topical treatment. The second group (positive control) received brotowali extract orally at a dose of 450 mg/kg and was treated with Betadine ointment. The third group (negative control) received brotowali extract orally at a dose of 450 mg/kg and was treated with the ointment base. The fourth, fifth, and sixth groups received brotowali extract orally at a dose of 450 mg/kg and were treated with brotowali stem ethanol extract ointment at concentrations of 20%, 35%, and 50%, respectively. The oral extract dose followed the method described by Ashari et al [1]. Ointment application was performed twice daily at 08:00 AM and 02:00 PM by applying thin, even layer to the wound surface [14]. Wound healing was assessed based on scores obtained from macroscopic observations over 15 days (from day 8 to day 22). A lower score indicated better wound healing. On day 23, the rats' blood glucose levels were measured again to determine the final glucose levels.

2.7. Assessment of diabetic burn wound healing

Diabetic burn wound healing was evaluated using a macroscopic observation scoring system adapted from the study by Priamsari and Yuniawati [18] (Table 1).

Table 1. Diabetic burn wound healing score in rats through macroscopic observation

Score	Wound Description
1	White and clean, all crusts have fallen off
2	White with brownish, all crusts have fallen off
3	White with brownish, most crusts have fallen off
4	White with brownish, half of the crusts have fallen off
5	White with brownish, nearly half of the crusts have fallen off
6	White with brownish, a small amount of crusts have fallen off
7	White with brownish, crusts present and not fallen off
8	Dark brown, all crusts have fallen off
9	Dark brown, most crusts have fallen off
10	Dark brown, half of the crusts have fallen off
11	Dark brown, nearly half of the crusts have fallen off
12	Dark brown, a small amount of crusts have fallen off
13	Dark brown, crusts present and not fallen off
14	Brown, all crusts have fallen off
15	Brown, most crusts have fallen off
16	Brown, half of the crusts have fallen off
17	Brown, nearly half of the crusts have fallen off
18	Brown, a small amount of crusts have fallen off
19	Brown, crusts present and not fallen off
20	Reddish brown, all crusts have fallen off
21	Reddish brown, most crusts have fallen off
22	Reddish brown, half of the crusts have fallen off
23	Reddish brown, nearly half of the crusts have fallen off
24	Reddish brown, a small amount of crusts have fallen off
25	Reddish brown, crusts present and not fallen off
26	Reddish pale, crusts present
27	Reddish pale, no crusts present
28	Pale wound bed, no scab, no crusts present

2.8. Statistical analysis

Wound scores recorded during the observation period (days 8 to 22) were analyzed statistically using SPSS software, version 27. The statistical analysis involved the Kruskal-Wallis test, followed by the Mann-Whitney test.

3. Results

3.1. Phytochemical content of brotowali extract *(T. crispa L.)*

Qualitative phytochemical testing was conducted to identify secondary metabolites in brotowali stem extract that contribute to the healing of diabetic burn wound. Based on the results of the qualitative phytochemical test, brotowali extract was found to contain secondary metabolites, including flavonoids, saponins, steroids, and tannins.

Healing capacity of Brotowali ointment (*T. crispa* L.) for diabetic burn wounds diabetic burn wound was observed macroscopically from day 8 to day 22. The macroscopic characteristics of the wound were then matched with the descriptions in the wound scoring table. A lower wound score indicates better healing capacity. After assigning scores to each wound, a more specific analysis of the healing capacity was performed using the obtained wound scores. The results of the wound score analysis are presented in Table 2.

In this study, the initial blood glucose measurement and diabetes induction in the experimental animals were conducted on day 0. Subsequently, blood glucose levels were rechecked on day 3, and it was found that the glucose levels in all experimental animals were >200 mg/dL. According to Panjaitan et al. [14] animals are considered diabetic if their blood glucose levels exceed 200 mg/dL. After confirming diabetes in the experimental animals, wound creation

Table 2. Results of the Mann-Whittney post-hoc test for the average diabetic burn wound scores in rats over 15 days, from day 8 to day 22

Days	Mean±SD					
	K1	K2	K3	K4	K5	K6
8	28±0^{a}	28±0^{a}	28±0^{a}	28±0^{a}	28±0^{a}	28±0^{a}
9	26±0^{a}	25.8±0.45^{a}	26±0^{a}	26±0^{a}	26±0^{a}	26±0^{a}
10	26±0^{a}	25.8±0.45^{a}	26±0^{a}	26±0^{a}	25.8±0.45^{a}	25.8±0.45^{a}
11	26±0^{a}	25.8±0.45^{a}	26±0^{a}	26±0^{a}	25.8±0.45^{a}	25.8±0.45^{a}
12	22.6±3.29^{a}	22.6±5.37^{a}	25.2±0.45^{a}	25.8±0.45^{a}	24.2±2.95^{a}	23.8±2.77^{a}
13	21.4±3.29^{a}	22.6±5.37^{a}	21.6±2.19^{a}	19.4±0.89^{a}	18.4±0.55^{a}	20±3.39^{a}
14	20.2±2.68^{b}	20.2±5.02^{b}	21.6±2.19^{b}	19±1.22^{ab}	18.4±0.55^{a}	16.2±3.42^{a}
15	20.2±2.68^{b}	18.8±3.9^{ab}	21.6±2.19^{b}	18.6±0.55^{ab}	17.8±1.3^{ab}	15.4±3.78^{a}
16	19±0^{bc}	17.8±2.68^{b}	21.6±2.19^{c}	15.2±3.03^{ab}	16±2.55^{ab}	14.4±3.21^{a}
17	19±0^{bc}	17.8±2.68^{b}	21.2±2.49^{c}	14.8±3.49^{ab}	14.8±1.92^{ab}	14.2±3.03^{a}
18	19±0^{bc}	17±4.47^{bc}	19±0^{c}	14.4±3.36^{ab}	12.6±4.16^{a}	11.8±1.1^{a}
19	17.8±2.68^{bc}	14±4.36^{ab}	19±0^{c}	13±5^{ab}	12.4±4.04^{ab}	11±1.41^{a}
20	17.8±2.68^{c}	11.4±4.39^{abc}	17.4±2.19^{bc}	12.6±5.08^{bc}	12±4.53^{b}	6.8±3.49^{a}
21	16.2±3.03^{bc}	11±4.53^{ab}	16.2±2.68^{c}	12.2±4.6^{abc}	10.6±4.39^{ab}	6.6±3.29^{a}
22	15.6±3.13^{d}	9.2±2.28^{bc}	13.6±1.14^{d}	9.6±1.52^{bc}	7.8±2.68^{ab}	4.6±3.58^{a}

Note: Alphabetical notations a, b, c, d on the same row indicate significant differences based on the Mann-Whitney test ($P<0.05$). Normal control (no treatment); K2: Positive control (Betadine ointment); K3: Negative control (ointment base); K4: 20% brotowali ointment; K5: 35% brotowali ointment; K6: 50% brotowali ointment.

were created on day 4, with the initial wound area showing redness, slight dampness, and some burned surface areas. From day 5 to day 7, the wounds were left untreated both orally and topically. On day 8, before treatment was administered, the wound condition was first observed. The macroscopic appearance and healing of diabetic burn wound on days 8 were evaluated in the following groups: normal control (no treatment), K2 (positive control, Betadine oinment), K3 (negative control; ointment base), K4 (20% brotowali ointment), K5 (35% brotowali ointment), and K6 (50% brotowali ointment). The wounds in all individual experimental animals showed similar changes ($P>0.05$), with the skin appearing white but without any scabs (score 28). Initially, the wounds displayed redness and burned surface areas. The differences in wound appearance on day eight compared with previous days indicate the occurrence of natural healing processes. The macroscopic appearance and healing of diabetic burn wound on days 9 showed changes in wound condition: normal control (no treatment), K2 (positive control, Betadine oinment), K3 (negative control, ointment base), K4 (20% brotowali ointment), K5 (35% brotowali ointment), K6 (50% brotowali ointment), characterized by reddish-white scabs on the wound surface (score 26), except in the K2 group where the scabs were reddish-brown (score 25.8).

Statistically, the K2 group had a lower average score; however, there was no significant difference compared with the other groups ($P>0.05$). By the end of the observation period (day 22), the macroscopic appearance and healing of diabetic burn wounds differed among the treatment groups: normal control (no treatment), K2 (positive control, Betadine oinment), K3 (negative control, ointment base), K4 (20% brotowali ointment), K5 (35% brotowali ointment), and K6 (50% brotowali ointment). The K5 and K6 groups displayed wound conditions with a whitish-brown crust, with K6 showing half of the crust detached (score 4.6) and K5 showing no crust detached (score 7.8). The K2 (score 9.2) and K4 (score 9.6) groups showed dark brown crusts with complete crust detachment. The K3 group exhibited a dark brown crust with no detachment (score 13.6), while the K1 group had a brown crust with nearly complete crust detachment (score 15.6). The average wound scores presented in Table II indicate that the K6 group demonstrated the best healing capacity, with the lowest average wound score. Although there was no statistically significant difference between the K6 and K5 groups ($P>0.05$), significant differences were observed compared with the K1, K2, K3, and K4 groups ($P<0.05$). Based on the average wound scores, the order of groups from the best to worst was K6, K5, K2, K4, K3, and K1, with scores evaluated based on crust color and amount of crust detachment.

4. Discussion

Wound healing is a complex and dynamic process [19, 20]. This process consists of three stages: inflammation, proliferation, and maturation [18-20]. The inflammatory phase typically occurs from the onset of the wound until the third day post-injury [14, 18, 20]. The proliferative phase usually begins around the third or fourth day [14, 20] and continues through the second week after injury [20]. The maturation phase, the longest phase of wound healing, typically begins during the second week after the injury [18] and can last for months or even years [14, 18, 20].

Each stage of wound healing exhibits distinct macroscopic characteristics [18, 19, 20]. The first stage, the inflammatory phase, involves the inflammatory response in which immune cells eliminate bacteria and remove dead tissue [20]. Additionally, this phase generates growth factors that stimulate the new tissue formation [20], promotes coagulation, recruits repair cells, releases cytokines [21], and fills the empty lumen with blood due to changes in capillary conditions [19]. Macroscopically, the inflammatory phase is characterized by a reddish wound surface [18-20] and a wet appearance [19].

The second stage is proliferative phase, which is characterized by the formation of epithelial cells, blood vessels, and new tissue [20, 21]. This phase is marked by scab formation [13, 14, 18, 19]. The proliferation phase ends with scab shedding, during which epithelial cells migrate from the wound edges towards its center [14, 18]. Scab detachment indicates that the underlying tissue has dried, signaling the transition to the maturation phase [20, 21]. The third final stage is maturation phase. During this phase, epithelialization occurs [18, 21] along with gradual tissue recovery through increased collagen deposition [14]. Epithelialization is observed when the scab falls off, ideally without leaving a scar [18], and the epithelial tissue appears pale white [21]. If a scar remains, it indicates that the scab has not fully matured, leading to a delay in the maturation phase as the wound may dry out and form a new scab [19].

In this study, the inflammatory phase of wound healing lasted five days, beginning immediately after wound creation (day four) and ending on day eight. This phase was characterized by wounds showing redness and a moist appearance on the day of wounding (day four). This observation is consistent with Priamsari and Yuniawati [18]; Rahman and Kamri [19], who reported that the inflammatory phase is marked by redness and moisture at the wound site. By the end of the inflammatory phase

(day eight), the wounds appeared whitish, but no visible scab formation was observed. According to Panjaitan et al. [14]; Rahman and Kamri [19], scab formation indicates that the wound healing has transitioned to the proliferative phase. Thus, the absence of a scab formation suggests that the healing process remains within the inflammatory phase.

On day nine, the wound healing process in all treatment groups entered the proliferative phase, as indicated by scab formation. By the end of the observation period (day twenty-two), wounds in all treatment groups remained in the proliferative phase and had not yet transitioned to the maturation phase, as evidenced by the presence of residual scabs and the lack of a pale appearance. This suggests that the scabs had not fully matured, resulting in a delay in advancing to the maturation phase due to the scabs re-forming as the wounds dried. This finding is consistent with Priamsari et al. [18] who note that the maturation phase begins when scabs detach completely without leaving residual marks.

Based on the wound observations, the 50% brotowali ointment demonstrated a superior healing effect compared with the 35% and 20% concentrations. This finding suggests that higher extract concentrations result in a greater amount of secondary metabolites, which are hypothesized to enhance wound healing capacity. Additionally, Betadine ointment also effectively promotes wound healing due to its povidone-iodine content, which controls bacterial infections and stimulates the formation of new blood vessels at the wound site [22].

In this study, the oral and topical preparations of brotowali stem extract acted synergistically to heal diabetic burn wound. Previous have shown that plants capable of healing diabetic wound through oral administration include bidara upas (*M. mammosa* (Lour.)) [12], black pepper (*P. nigrum*), peanuts (*Arachis hypogaea*) [13], and insulin (*Smallanthus sonchifolius*) [12]; however, their topical use has not been reported. Plants reported to heal diabetic wound topically in ointments formulations include binahong leaves (*Anredera scandes* (L.) Moq.) [23], lime peel (*Citrus aurantifolia*) [24], and *Mahkota dewa* (*Phaleria macrocarpa*) [25]. Binahong leaves ointment promotes diabetic gangrene healing through its flavonoids, saponins, triterpenoids, and tannins contents by accelerating the proliferative phase [23]. Lime peel ointment can heal diabetic gangrene with an ability comparable to that of the positive control, supported by its flavonoids, alkaloids, and saponins contents [24]. Furthermore, *M. dewa* ointment improves diabetic gangrene healing by reducing oxidative stress through its flavonoids, saponins, tannins, and phenolics [25].

Further qualitative phytochemical analysis revealed that the ethanol extract of brotowali stems contains secondary metabolites, including flavonoids, saponins, tannins, and steroids. These secondary metabolites contribute to the wound-healing capacity of brotowali ointment. Panjaitan et al. [14] state that the most common mechanisms by which secondary metabolites aid wound healing include antioxidants, anti-inflammatory agents, and antibacterial agents. Specifically, in the context of diabetic wound, flavonoids act as antioxidants by binding to excess free radicals produced at the wound site and as antibacterial agents by inhibiting bacterial invasion during the early phases of wound healing [12]. Moreover, flavonoids can enhance fibroblast proliferation, accelerate re-epithelialization, and improve wound contraction rates [3, 15]. Additionally, Nurwahita et al. [3] suggest that saponins can shorten the epithelialization period and prevent cellular damage. Saponins also stimulate vascular endothelial growth factor (VEGF) and increase the number of fibroblasts and macrophages migrating to the wound area [13].

Panjaitan et al. [14] explain that tannins aid in wound healing by acting as astringents, which help stop bleeding and promote faster wound drying, and enhance the formation of capillaries and fibroblasts. Additionally, in the context of diabetic wound healing, tannins provide anti-inflammatory effects [13]. Furthermore, steroids contribute to wound healing by preventing excessive growth of granulation tissue [22].

The limitation of this research is that the wound model created was limited to burn wounds only. Furthermore, this research involved not only topical preparations but also involves oral administration of the extract. In addition, this study was limited to experimental animals and has not yet been conducted in humans. According to Cachet et al. [25], patients who consume brotowali stems in the form of liquid extract or tablets regularly and continuously may develop hepatitis. However, Cachet et al. [25] also reported that patients with hepatitis patients caused by consuming brotowali stems can recover after several weeks without special medical treatment. In this regard, controlled clinical trials are needed to determine the safety of the proper use of brotowali.

Conclusion

We conclude that 20% brotowali extract ointment has a healing capacity for diabetic burn wound comparable to that of Betadine ointment. Furthermore, the 35% and 50% brotowali ointments exhibit superior healing capabilities.

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Compliance with ethical guidelines

The use of experimental animals and all study the procedures were approved by the Faculty of Health Sciences, Universitas Respati, Yogyakarta, Indonesia (Code: 044.3/FIKES/PL/V/2023).

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Authors' contributions

Conceptualization, study design, statistical analysis, and writing the original draft: Ruqiah Ganda Putri Panjaitan and Fitriyani Fitriyani; Data acquisition, experiments, data interpretation, Project administration, technical, and material support, review and editing: Ruqiah Ganda Putri Panjaitan, Fitriyani Fitriyani, Titin Titin, Muhammad Waseem Alam, and Aji Suhartoyo.

Conflict of interest

The authors declared no conflict of interest.

Data availability

The data that support the findings of this study are available from the corresponding author and Fitriyani Fitriyani upon reasonable request.

References

[1] Ashari A, Nurinda E, Fatmawati A. Increased insulin levels in male Wistar rats (Rattus norvegicus) induced by streptozotocin due to administration of ethanol extract of Tinospora crispa stems (Tinospora crispa L.). Indones Pharm Nat Med J. 2021; 5(1):1-8. [DOI:10.21927/inpharnmed.v5i1.1653]

[2] Dewi NLKA, Prameswari PND, Cahyaningsih E, Megawati, Agustini NPD, Juliadi D. Review: The use of plants as phytotherapy for diabetes mellitus. J Tradit Med Integr. 2022; 2(1):31-42. [DOI:10.36733/usadha.v1i3.5562]

[3] Nurwahita, Masriadi, Mahmud NU, Amelia R. The effect of ginger, lemongrass, clove extract ointment diabetes mellitus wound healing. Indones J Pharm Sci Technol. 2024; 11(1):62-71. [DOI:10.24198/ijpst.v11i1.47645]

[4] Santoso P, Rahayu D, Irawan H. Analysis of the application of gangrenous wound care in diabetic ulcer patients: literature review. J Keperawatan. 2022; 14(1):171-8. [Link]

[5] Kuhlmann M, Alberti WW, Mackensen YV, Ebbinghaus M, Williams R, Kyora FK, et al. Wound healing characteristics of a novel wouns healing ointment in an abrasive wound model: A randomised, intra-individual clinical investigation. Wound Med. 2019; 24(1):24-32. [DOI:10.1016/j.wndm.2019.02.002]

[6] Martani NS, Fatmaria F. Utilization of the antidote (Tinospora crispa L.) as a traditional medicinal plant by the Ngaju Dayak Tribe. J Pharmascience. 2020; 7(2):156-65. [DOI:10.20527/jps.v7i2.8799]

[7] Irawan C, Putri ID, Sukiman M, Utami A, Ismail, Putri RK, et al. Antioxidant activity of DPPH, CUPRAC, and FRAP methods, as well as activity of alpha-glucosidase inhibiting enzymes from Tinospora crispa (L.) stem ultrasonic extract. Pharmacogn J. 2022; 14(5):511-20. [DOI:10.5530/pj.2022.14.128]

[8] Sanjaya DA, Yuda PESK, Cahyaningsih E, Rahayu LRP, Putra MADM. Ethnomedicinal study in traditional medicine for diabetic foot ulcers (DFU) in Bali. J Farm Udayana. 2021; 10(02):172-6. [DOI:10.24843/JFU.2021.v10.i02.p11]

[9] Tarukbua YSF, Queljoe ED, Bodhi W. Phytochemical screening and toxicity test of ethanol extract of Tinospora crispa L. leaves using the BSLT test method. Pharmacon J Ilm Farm. 2018; 7(3):330-7. [DOI:10.35799/pha.7.2018.20600]

[10] Bare Y, Sari DRT, Meak LEC, Mogi MC. In silico study of columbin from Tinospora crispa L. as dihydrofolate reductase-thymidylate synthase (DHFR-TS) inhibitor. Bioscience. 2020; 6(1):12-17. [DOI:10.24036/0202261116090-0-00]

[11] Adnan AZ, Armin F, Sudji IR, Novida MD, Roesma DI, Ali HA, et al. In vitro anti-inflamation activity test of tinocrisposide and freeze-dried aqueous extract of Tinospora crispa stems on human red blood cell by increasing membrane stability experiment. Asian J Pharm Clin Res. 2019; 12(5):125-9. [DOI:10.22159/ajpcr.2019.v12i5.30690]

[12] Marchianti ACN, Ulfa EU, Sakinah EN. The dose dependence analysis of the water fraction of Merremia mammosa (lour.) extract on diabetic wound healing enhancement. Hiroshima J Med Sci. 2018; 67(Special Issue):29-34. [Link]

[13] Dwita LP, Hasanah F, Srirustami R, Repi, Purnomo R, Harsodjo S. Wound healing properties of Epiphyllum oxypetalum (DC.) Haw. leaf extract in streptozotocin-induced diabetic mice by topical application. Wound Med. 2019; 26(1):100160. [DOI:10.1016/j.wndm.2019.100160]

[14] Panjaitan RGP, Maulidya A, Raharjeng ARP, Chakma S. Healing capacity of leaves stalks extract of pasak bumi (Eurycoma longifolia Jack.) on incised wounds of mice skin. Pharmacogn J. 2020; 12(6):1351-6. [DOI:10.5530/pj.2020.12.186]

[15] Maru AD, Lahoti SR. Formulation and evaluation of ointment containing sunflower wax. Asian J Pharm Clin Res. 2019; 12(18):115-20. [DOI:10.22159/ajpcr.2019.v12i18.33199]

[16] Panjaitan RGP, Novitasari. Anti-diabetic activity of the red dragon fruit peel (Hylocereus polyrhizus) in ethanol extract against diabetic rats experimental animals. Pharmacogn J. 2021; 13(5):1079-85. [DOI:10.5530/pj.2021.13.140]

[17] Tumigolung DPU, Runtuwene MRJ, Wewengkang DS. The effectiveness of healing burns with ethanol extract ointment from soyogik leaves (Sauraia bracteosa DC) on male white rats of the Wistar strain (Rattus Norvegicus). Pharmacon. 2019; 8(2):371-9. [DOI:10.35799/pha.8.2019.29303]

[18] Priamsari MR, Yuniawati NA. Phytochemical screening and burn healing activity of ethanolic extract of Morinda citrifolia L. J Pharm. 2019; 8(1):22-8. [DOI: 10.373013/7sk16r03]

[19] Rahman S, Kamri AM. Test of the epithelialization effect of dragon tail leaf extract (Rhaphidophora pinnata (L.f) Schoot.) on mice. As-Syifaa J Farm. 2019; 11(1):75-81. [DOI:10.56711/jifa.v11i1.516]

[20] Lestari T, Syukur S, Revilla G, Rita RS, Rustini R. The burn wound healing procces: A review. Int J Progress Sci Technol. 2023; 40(1):77-88. [DOI:10.52155/ijpsat.v40.1.5565]

[21] Primadina N, Basori A, Perdanakusuma DS. The wound healing process is reviewed from the cellular and molecular mechanism aspects. Qanun Med. 2019; 3(1):31-43. [DOI:10.30651/jqm.v3i1.2198]

[22] Ginting GA, Asfianti V, Tarigan MH. Wound healing test of ethanol extract of torch ginger fruit (Etlingera eliator Jack.) on white mice. Forte J. 2022; 2(1):42-51. [DOI:10.51771/fj.v2i1.201]

[23] Ardinata IPR, Satriyasa BK, Sumardika IW. Fibrogenesis enhancement activity test of binahong leaf extract ointment (Anredera scandes (L.) Moq.) 10% in healing diabetic wounds in Sprague Dawley rats. Lumbung Farm J Pharm Sci. 2023; 4(1):101-7. [Link]

[24] Ulfa AM, Primadiamanti A, Alim FN. Testing the effectiveness of lime peel (Citrus aurantifolia) extract ointment formulation as a wound healing agent for type I diabetes in male rats. J Farm Malahayati. 2021; 4(2):126-37. [DOI:10.33024/jfm.v4i2.5246]

[25] Cachet X, Langrand J, Riffault-Valois L, Bouzidi C, Colas C, Dugay A, et al. Clerodane furanoditerpenoids as the probable cause of toxic hepatitis induced by Tinospora crispa. Sci Rep. 2018; 8(1):13520. [DOI:10.1038/s41598-018-31815-6] [PMID]