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Review Article

Effectiveness of *Helichrysum italicum* Essential Oil on Wound Healing

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Abstract: *Helichrysum italicum* essential oils (EOs) has long been known as a medicinal agent, especially in wound healing. This is inseparable from the chemical compounds contained in it, such as abundant monoterpenes and sesquiterpenes. The purpose of this systematic review is to examine various studies on the effectiveness of *H. italicum* EOs on wound healing. Research data searches were conducted through Google Scholar, Proquest, Science Direct, SpringerLink, Wiley Online Library, Taylor & Francis Online and PubMed over the last 10 years by following inclusion and exclusion criteria. Obtained as many as 11 studies that match the criteria. Based on the review of these studies, it was found that *H. italicum* EOs has good wound healing abilities when viewed from various aspects such as its chemical components, biological activity and its effect on injured body tissues. These chemical components vary in type and amount because they are influenced by various aspects. Biological activities such as antioxidant, antimicrobial, and anti-inflammatory effects have also been shown to be contained in this oil because it affects various phases of wound healing, namely hemostasis, inflammation, proliferation and remodeling phases. This is also inseparable from the accuracy of the type of vehicle used.

Keywords: *Helichrysum italicum*; essential oils; wound healing; chemical components

1. INTRODUCTION

Essential oils (EOs) is naturally occurring aromatic molecules from plants. In general, this oil is known for its various health benefits. However, research on the application of this oil on skin health is still limited [1]. In addition, this oil is also known for its low toxicity, so it is relatively safe to use [2].

One source of EOs is the *Helichrysum* plant. The origin of the name *Helichrysum* comes from the Greek words, namely sun and gold (Ἥλιος/Helios and χρυσός/Chryos) which describe the bright yellow color of the flower heads [3], [4]. More than 500 species of *Helichrysum* are widely distributed between the Mediterranean region, Africa, Asia and Australia [4]. The species are very diverse related to the phenotype and profile of the metabolites contained therein [5]. *H. italicum* (Figure 1) is a typical Mediterranean plant scattered on the east coast and islands in the Adriatic Sea [2]. It usually grows in dry, rocky, or sandy soils around the Mediterranean region [4]. The first scientific study of the medicinal effects of *H. italicum* was carried out by Leonardo Santini. He researched the effect of this plant on the treatment of psoriasis fmmm 1940-1950. However, his findings were published in less important journals and were largely ignored [3].

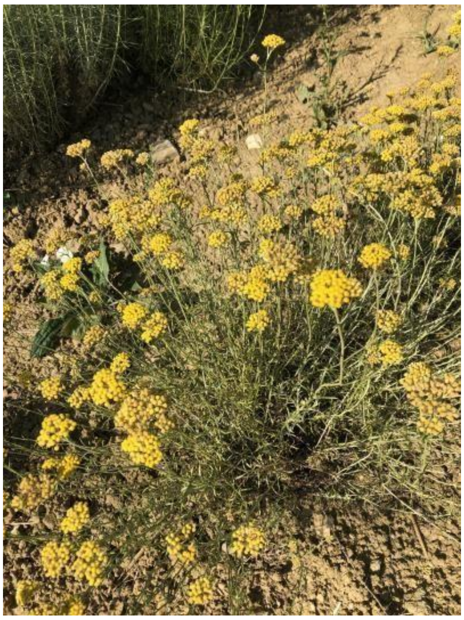

Figure 1. *Helichrysum italicum* Plants [9]

H. italicum EOs is often called "*Immortelle*" or immortal, because the flowers do not wither and the color remains alive even after being collected and dried. This species contains abundant monoterpenes and sesquiterpenes [6]. These compounds have been shown to have beneficial effects that have been studied, such as playing a role in anti-inflammatory, antimicrobial, antioxidant, anti-allergic, antimalarial and especially playing a role in wound healing. Recently, attention has been paid to the tissue regeneration and anti-inflammatory effects of *H. italicum* EOs, and it has been proven that this oil can be a potential agent for wound healing after reconstructive surgery [7], [8]. As a medicinal agent, this oil has been used for a long time and some products have also been sold in the market [9].

This review was systematically carried out using studies on the effectiveness of *Helichrysum italicum* EOs on wound healing over the past 10 years. The purpose of this systematic review is to examine various studies on the effectiveness of *Helichrysum italicum* EOs on wound healing.

2. WRITING METHODS

This systematic review was made in July 2022 by conducting searches through Google Scholar, Proquest, Science Direct, SpringerLink, Wiley Online Library, Taylor & Francis Online and PubMed over a span of 10 years. The search keywords were "Essential Oil" and ("Helichrysum italicum" or "Immortelle") and "Wound Healing". In this review, inclusion and exclusion criteria were made. The inclusion criteria were researched in English, published in the last 10 years, with in vitro and in vivo studies, and discussed the effects of *H. italicum* EOs on wound healing including its anti-inflammatory, anti-microbial and anti-oxidant effects. The exclusion criteria were non English studies, published over the last 10 years, and the other type of studies such as books, literature reviews, case reports, case series and others.

These studies met the restriction requirements when discussing the effectiveness of *H. italicum* EOs on wound healing. Evaluation of therapy using various parameters such as percentage of wound contraction, antimicrobial activity, antioxidant activity, histopathological analysis and others.

3. RESULT AND DISCUSSION

The analysed journals were selected from four different electronic databases such as Google Scholar, Proquest, Science Direct, SpringerLink, Wiley Online Library, Taylor & Francis Online and PubMed. Duplicates were removed automatically. Restrictions were carried out based on title and abstract screening. Then continued with full text screening for eligibility and finally 11 studies were obtained. The flow of the studies exclusion strategy is shown in Figure 2. Then the characteristics of each studies obtained are described in Table 1.

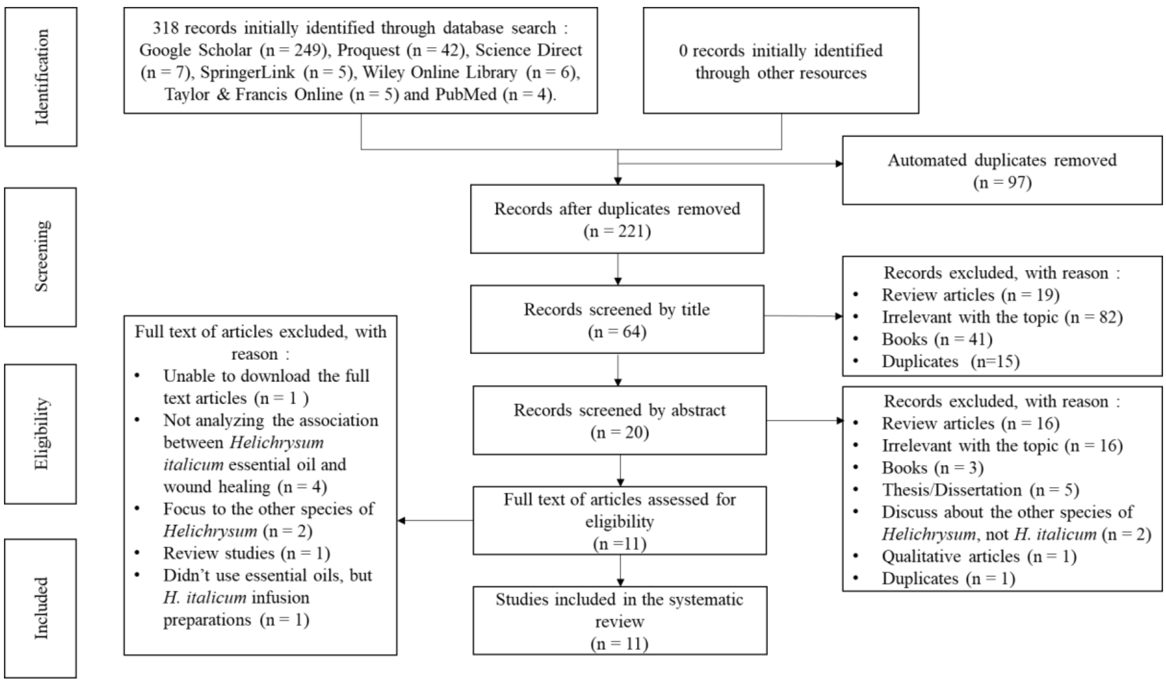

Figure 2. Article Exclusion Strategy Flow

Helichrysum italicum is Mediterranean plant originating from the *Asteraceae* family [10]. Various extracts can be made from this plant, including polar extract and EOs that are often used [11]. *H. italicum* EOs have various biological activities such as antimicrobial, anti-inflammatory and antioxidant [12]–[15]. These various effects cannot be separated from the chemical content in *H. italicum* EOs. There is study showing that this oil contains terpenes (monoterpene and sesquiterpene hydrocarbons) as the main components, such as α-curcumin (14.07%), neryl acetate (12.96%), α-pinene (12.38%), β-selinene (11.27%). and α-selinene (7.27%) [12]. This is supported by other study which also show terpenoids as the main component in the EOs of *H. italicum*, especially if obtained by the hydrodistillation method [16].

Table 1. Research Characteristics

Study (Year)	Model	Plant Part Used	Parameter	Result
Xuesheng H., Cody B. and Nicole S. (2017)[1]	In Vitro	Unspecified	Chemical components analysis by gas chromatography mass spectrometry (GC-MS) Protein critical for inflammation Immune response by enzyme-linked immunosorbent assay (ELISA) Tissue remodeling process	The major components of *H. italicum* are neryl acetate (35.5%), γ-curcumene (13.9%), α-pinene (8.9%) *H. italicum* EOs (0.01%) significantly inhibited the production of collagen I and III
Roberta A., et al. (2020)[17]	In Vitro	Leaves	Total phenols by Folin-Ciocalteau's method The antioxidant activity by ABTS assay The cytotoxicity of the compounds by MTT viability assay and the proliferative capabilities of fibroblasts by scratch assay	Total phenols from *H. italicum* from four concentrations (1 µl, 5 µl, 10 µl and 25 µl) were 49.55 ± 7.40, 199.36 ± 20.78, 378.05 ± 37.34 and 663.11 ± 42.71 in µg of gallic acid equivalent (GAE) No cytotoxic effects on cell proliferation, especially at low concentrations (1 µL/mL) and for prolonged culturing time. The effect on cell viability could be detected when cells were cultured for 96 hours in the presence of 1-10 µL/mL *H. italicum* extract ($p < 0.01$ and $p < 0.05$). Migration and proliferation of fibroblasts stimulated by *H. italicum* EOs extract were calculated by the percentage of wound closure at 24, 48 and 72 hours after the damage was caused, obtained at concentrations of 1 µl/ml (14.2 ± 3.4%, 19.3 ± 0.2% and 19.9 ± 0.0 %), 5 µl/ml (17.1 ± 3.0%, 22.8 ± 0.3% and 23.6 ± 0.0%) and 10 µl/ml (14.4 ± 1.9%, 16.0 ± 0.0% and 16.4 ± 0.0%).

Study (Year)	Model	Plant Part Used	Parameter	Result
Marijana A. et al. (2021)[12]	In Vitro and In Vivo	Flowers	Chemical components analysis by GC-MS Antioxidant activity by DPPH free radical scavenging assay, hydroxyl ion (.OH), nitric oxide (.NO), lipid peroxidation (LP) and ferric reduction antioxidant potential (FRAP) test Percentage of wound contraction Total hydroxyproline Redox status Histological observation	Forty-six of chemical components were identified in *H. italicum* EOs. Sesquiterpene hydrocarbons were found in almost 60% of the total, while monoterpene hydrocarbons accounted for 18.52% of the total. The main components detected were α-curcumin (14.07%), neryl acetate (12.96%), α-pinene (12.38%), β-selinene (11.27%), and α-selinene (7.27%). Antioxidant activity with DPPH, .OH and LP obtained IC50 respectively at 4.45 ± 0.44 μg/ml, 13.33 ± 1.11 μg/ml and 10.48 ± 1.22 μg/ml. While in FRAP, obtained 0.03 ± 0.00 mg AAE/ml *H. italicum* EOs. In .NO, no data were found. *H. italicum* gel and ointment on 7, 14 and 21 days after topical administration showed the percentage of wound closure was 42.3% and 39.87%, 84.21% and 82.39%, and 98.75% and 98.33%, respectively. *H. italicum* EOs gel and oinment showed significant improvement in comparison to negative control and vehicle groups, but did not show a significantly higher amount of hydroxyproline compared to the positive control group *H. italicum* gel and ointment significantly decreased the level of hydrogen peroxide (H_2O_2) release in relation to the negative control ointment and gel groups

Study (Year)	Model	Plant Part Used	Parameter	Result
				The most severe reparative fibrosis was observed in the negative control group . The first signs of the return of dermal adnexa, such as sebaceous and sweat glands and hair follicles, but still with a presence of fibrosis, were observed in the group's ointment and gel base.
Marija S. et al. (2021)[10]	In Vitro	Flowers	Chemical components analysis by GC-MS Antimicrobial activity Myeloperoxidase (MPO) activity NO production Arginase activity	Oil 1 contains α-pinene (16.8%), neryl acetate (21.2%) and γ-curcumin (16.8%) Oil 2 contains neryl acetate (35%), γ-curcumin (10.2%) and β-diketone (11.9%) Oil 3 contains α-pinene (19.9%) and neryl acetate (9.2%) Oil 4 contains α-pinene (4.9%) and neryl acetate (15.5%) The higher the levels of mono hydrocarbons and sesquiterpenes contained in the oil, the stronger the anti-inflammatory effect Oils 3 and 4 had higher antimicrobial activity than oils 1 and 2 because they showed a prominent antistaphylococcal effect, namely with MIC (Minimal Inhibitory Concentration) of 0.6 and 0.3 mg/ml, respectively Oils 1 and 4 showed significant potential in inhibiting MPO enzyme activity at the highest concentrations Oil 1 which is rich in mono and sesquiterpene hydrocarbons induces a decrease in NO production depending on the concentration

Study (Year)	Model	Plant Part Used	Parameter	Result
				The effect on arginase activity was only at the two lowest concentrations, namely 1 x 10^{-1} mg/ml and 1 x 10^{-2} mg/ml. Oil 4 showed prominent inhibition (38%, 37% and 42%, 32%, respectively).
R. Ahmed Khan and Sandy F. van Vuuren (2021)[18]	In Vitro	Unspecified	The inhibitory and bactericidal effect against two pathogens (*Clostridium perfringens* and *Clostridium septicum*) with The Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) assays	Strong activity is seen where the MIC value is 0.10 − 0.50 mg/ml and very strong activity is seen with an MIC value of < 0.10 mg/ml. EOs with a bactericidal efficacy ≤ 1.00 mg/ml was considered as noteworthy. *H. italicum* has MIC average and MBC average >8.00 mg/ml against *C. perfringens*, while MIC average is 0.32 ± 0.19 mg/ml and MBC average is >8.00 mg/ml against *C. septicum*
Vanja M.T. et al. (2021)[19]	In Vivo	Inflorescences	High-Performance Liquid Chromatography (HPLC) Analysis of SCO2 *H. italicum* EOs Trans-Epidermal Water Loss (TEWL) Skin pH Erythema index (EI)	From HPLC analysis it was found that *H. italicum* EOs contains chlorogenic acid (about 11.5%) and arzanol (4.59%) After four weeks of sample application on the skin, all of them led to a significant reduction of TEWL compared to the NC test-site In comparison with the NC test-site, pH changes were statistically significant only on the 30th day after the initial measurement (the 2nd day after discontinuation of treatment) for all tested samples During the long-term application of the samples for 28 days, there were no significant changes in EI

Study (Year)	Model	Plant Part Used	Parameter	Result
Asta J., Jurga B., Irena N. and Rasa G. (2022)[20]	In Vitro and In Vivo	Inflorescences	Chemical composition by GC-MS Antioxidant activity by DPPH assay The toxic activity by brine shrimp (*Artemia salina*) bioassay	Inflorescence EOs of *H. italicum* was characterized by sesquiterpene hydrocarbons: γ-curcumene (21.5 ± 2.50%), β-selinene (13.6 ± 1.65%), β-eudesmol (8.3 ± 0.35%), *α*-selinene (8.1 ± 0.55%) and *α*-pinene (6.5 ± 1.50) *H. italicum* inflorescences have higher antioxidant activity (0.35 ± 0.03 mmol/L, TROLOX equivalent) rather than *H. arenarium* inflorescences (0.27 ± 0.01 mmol/L, TROLOX equivalent) LC50 values varied from 15.99 to 23.42 μg/mL for *H. italicum* and *H. arenarium* EOs, respectively. According to Meyer's and Clarkson's toxicity criterion, these EOs are classified as highly toxic.
Marijana A. et al. (2022)[21]	In Vitro and In Vivo	Unspecified	Percentage of wound contraction Total hydroxyproline Systemic redox status Histopathological analysis	The Immortelle ointment showed significant wound contraction from day 7 to day 21 as compared to other groups. On the day 21, there was an average of 99.32% wound contraction in the Immortelle group, whereas the mean wound contraction in the negative control and ointment base group was 71.36% and 81.26% , respectively. The highest level of L-hydroxyproline was noticed in the group treated with Immortelle ointment. The results indicated that untreated control rats were associated with the highest release of all oxidative stress parameters. However, the application of immortelle EOs significantly decreased the levels of superoxide

Study (Year)	Model	Plant Part Used	Parameter	Result
				anion radical (O_2^-) and thiobarbituric acid-reactive substances (TBARS) as compared to NC group In the NC group, the scar was composed of hypercellular fibrous tissue with a predomination of fibroblasts and a small amount of extracellular fibrous fibres. On the other side, in a group OB and IO, we detected evident post excision scar maturation with increased collagen fibres density. Also, the expression of the matrix metalloproteinase 9 (MMP-9) was intensively reduced in these two groups
Géraldine L. et al. (2022)[22]	In Vitro	Aerial	Chemical components by GC-MS Transcriptomic analysis confirmed by quantitative reverse transcription PCR analysis Skin barrier protein immunofluorescence Lipid staining Analysis of ceramides using liquid chromatography mass spectrometry (LC-MS)	Oxygenated monoterpenes were the most abundant chemical class of compounds with 43.76% of the total content. Neryl acetate (NA) showed the largest relative abundance, accounting for 32.80% of the total, followed by neryl propionate (4.66%), linalool and italicene (4.29%) and nerol (2.01%) for this chemical class. Transcriptomic analysis revealed that 41.5% of *H. italicum* EOs-modulated genes were also regulated by neryl acetate and a selected panel of genes *H. italicum* EOs led to a more pronounced increase in expressions of involucrin protein (+111 ± 37%) than NA (+51 ± 18%) compared to the control (DMSO 0.5%). The increase in total ceramides was significantly higher in the presence of *H. italicum* EOs (0.1%) than NA

Study (Year)	Model	Plant Part Used	Parameter	Result
				(0.03%), ranging from +114.2% to +64.2% compared to DMSO 0.5%.
Katja B., Katja K. and Darja BM. (2022)[23]	In Vitro	Shoots and Inflorescences	Antioxidant activity by 1,1-diphenyl-2-picrylhydrazyl (DPPH) Cytotoxicity assay Antimicrobial and antibiofilm activity	The total antioxidant activity of the EOs was 684.66 ± 94.22 μg of ascorbic acid equivalents ml^{-1}, higher than antioxidant potential of the hydrosol (4.88 ± 0.59 μg of ascorbic acid ml^{-1}) The cytotoxicity of the EOs was higher when compared to hydrosol for all cell lines (180-times higher for immortalized cell lines Caco-2 and A375, approx. 900-times for primary epidermal melanocytes and 25.000-times higher for CCD112CoN primary colon fibroblasts). EOs have higher biofilm inhibition activity when compared to hydrosols, but their practical application may still be hampered by some limitations, such as strong smell and taste and difficulties in rinsing from surfaces.
Zenon W. et al. (2022)[24]	In Vitro	Herbs and Inflorescences	EOs content and composition Chemical analysis by GC-MS and gas chromatograph combined with a flame ionization detector (GC-FID) Total Phenolic Content Analysis of phenolic acids and flavonoids by HPLC Antioxidant activity by DPPH and ABTS scavenging capacity assays Antibacterial activity	The content of EOs was higher in the inflorescences (0.31 g x 100 g^{-1}) than in the herb (0.25 g x 100 gr^{-1}) Herb EOs have higher content of neryl acetate (20.27%) and α-pinene (10.42%) and lower content of nerol (4.49%) compared to inflorescence (16.38, 4.05, 15.73%, respectively) The herb contains flavonoids (rutoside) and phenolic acid (caffeic, rosmarinic, chlorogenic, neochlorogenic, isochlorogenic b and cichoric acid) of 3793.96 mg x 100 g^{-1}, higher than inflorescences (1862.04 mg x 100 g^{-1}, respectively)

Study (Year)	Model	Plant Part Used	Parameter	Result
				Methanol extracts from herbs and inflorescences have higher antioxidant activity than EOs. Meanwhile, when comparing herbs and inflorescences, herbs have higher antioxidant activity. Herb EOs inhibited the growth of *E. coli* and *P. aeruginosa* with MIC values equal to 32 mg x ml^{-1}. In turn, inflorescence EOs showed higher MIC values (64 mg x mL^{-1}) which is equivalent to less effective bacteriostatic power.

Apart from terpenes, several other studies have shown that phenolic compounds from *H. italicum* also play a role in generating beneficial biological activities. On a study, there were significant results of total phenolic compounds from *H. italicum* in four concentrations [17]. Phenolic compounds are divided into flavonoids and non-flavonoids. Examples of flavonoid compounds are 4,2′,4′,6′-tetrahydroxychalcone-2′-glucoside, kaempferol-3-glucoside, naringenin- glycoside, gnaphaliin, pinocembrin and tiliroside which have been shown to have antioxidant and anti-inflammatory activities. While non-flavonoid compounds such as pyrones, phloroglucinols and acetophenones [25]. One of the pyrones compounds, arzanol has been shown to have anti-inflammatory effects because it inhibits the release of pro-inflammatory cytokines and antioxidants [26], [27].

However, the composition of EOs depends on various influencing factors, namely population, altitude, climatic conditions, stage of development (before flowering, full bloom and after flowering), ecology, plant community, plant part, extraction method and others [12], [13], [28]. From the 11 studies reviewed, most of them used inflorescence and flower samples (6 studies) and 2 studies used leaves or aerial part of plants , but the other studies were unspecified (3 studies). This is in line with other studies that discuss the parts used for *H. italicum* EOs samples. From the study it was said that the flowers and leaves are the parts most often used for health problems [3].

In several studies using *H. italicum* EOs samples of different origin, it was found that the EOs content of each place had a different dominance of chemical components. Oil extracted from Italy, Bosnia and Herzegovina is dominated by α-pinene, neryl acetate and γ-curcumin. While oil from France is dominated by neryl acetate, γ-curcumin and β-diketone, oil from Serbia is dominated by α-pinene and neryl acetate [13], oil from the Adriatic coast is dominated by α-pinene and γ-curcumin [12], oil from Corsica island is dominated by neryl acetate [13], [22], oil from Tuscany is dominated by α-pinene and γ-curcumin [14], and oil from Sardinia are dominated by γ- curcumin. On the Greek island of Amorgos, this oil is dominated by geraniol. That is the result of various influencing factors, as already mentioned [12].

Other studies have shown that the chemicals neryl acetate and pinene have benefits in wound healing. Neryl acetate plays a role in skin barrier function by increasing the lipid and ceramide content in the stratum corneum by increasing the expression of enzymes related to the synthesis of ceramides required for the glucosylceramide pathway. These two compounds will increase keratinocyte differentiation, cell cohesion and the formation of a cornified envelope, thus facilitating wound healing in the epidermis. While pinene contributes to the deposition of collagen so as to produce scars with effective tensile strength [12], [22].

In addition to neryl acetate and pinene, there are studies that discuss other ingredients of *H. italicum* EOs. In this study, the chlorogenic acid content was 11.5% and arzanol 4.59%. Chlorogenic acid is a compound that can accelerate the excision wound healing process through its antioxidant potential and its significant ability to increase collagen synthesis, while arzanol inhibits the biosynthesis of pro-inflammatory mediators, so that these two compounds help in wound healing [19].

The content of *H. italicum* EOs also acts as an antimicrobial agent which also supports wound healing. Because each plant origin affects its content, the antimicrobial effect of each region is different. In a study discussing the antimicrobial effect, it was shown that EOs from Serbia had a higher antimicrobial effect than those from France, Italy, and Bosnia and Herzegovina because of their high antistaphylococcal effect [13]. In another study that discussed the antimicrobial effect, it was shown that EOs from Bosnia had the ability to inhibit only *Aspergillus brasiliensis*, while EOs from France had a low inhibitory ability against *Bacillus subtilis* and *Staphylococcus aureus*, but had strong inhibition against *Aspergillus brasiliensis* [29]. Meanwhile, in other studies, this oil is effective against gram-positive and negative bacteria because it can damage the bacterial cell membrane, causing the release of DNA [15]. This is in line with other studies showing that the content of α-pinene and β-pinene is able to destroy cellular integrity and inhibit the process of respiration and ion transport from microorganisms [30].

In another study discussing antimicrobial effects, we investigated the effect of *H. italicum* EOs on *C. perfringens* and *C. septicum*, the most common causes of gangrenous wounds. It has the strong activity against *C. septicum* (the average MIC was 0.32 ± 0.19 mg/ml) [18]. In addition to bacteria and fungi, this EOs also has an antiviral effect against HSV-1 (Herpes Simplex Virus Type 1) at a concentration of 100-400 g/ml [31].

In addition to having various benefits, it turns out that this oil also has a low level of cytotoxicity and genotoxicity, so it is safe to use [3]. This is supported by studies that reviewed the use of *H. italicum* EOs which found that its safety has been proven by many in vitro and in vivo studies in humans. However, research on its own benefits is still lacking, because most studies do not discuss *H. italicum* EOs separately, especially regarding wound healing [32], [33].

Wound healing is a dynamic process, in which there are several phases, namely hemostasis, inflammation, proliferation and remodeling phases [12]. Disruption in these phases will slow down the healing process, such as oxidative stress. This occurs due to an imbalance between the levels of antioxidants, ROS (Reactive Oxygen Species) and nitrogen species [17].

H. italicum EOs has been proven to have anti-inflammatory effects both through in vitro and in vivo tests [34]. NO (Nitric Oxide) is produced in macrophages and plays a role in the inflammatory process. NO is released into cells and tissues with the help of iNOS (inducible NO synthase) which oxidizes L-arginine to L-citrulline. In inflammation, proinflammatory cytokines stimulate iNOS resulting in excessive NO production, which can injure endothelial cells. In a study that showed *H. italicum* EOs can reduce the production of NO, it's depending on the amount of concentration [13].

In addition to NO, myeloperoxidase enzymes also play a role in the inflammatory phase. This enzyme is released from macrophages during inflammation and functions to catalyze the production of hypohalous acids (HOCl and HOBr) which are the strongest bactericidal oxidants, produced in vivo with H_2O_2 and halide ions. But if excessive, can be a threat of damage to the body's own cells. Research has shown that *H. italicum* EOs significantly inhibits the excessive increase in the activity of this enzyme, so it does not cause negative effects on the body [13].

While in the proliferative phase there is the enzyme arginase. This enzyme converts L-arginine into L-ornitine and urea to prevent excess ammonia and supplies L-ornitine to cells that function as cell proliferation, collagen formation and other physiological functions. However, in mammals, excess of this enzyme actually causes dysfunction of the nervous and cardiovascular systems. In a study comparing the content of *H. italicum* EOs, it was shown that the mono and sesquiterpene hydrocarbon content of *H. italicum* EOs had the greatest inhibitory effect on arginase activity compared to other ingredients.[13]

In addition, there are cells that play an important role in the wound healing phase, especially from the late inflammatory phase to remodeling, namely fibroblasts. These cells are known to secrete growth factors, cytokines, collagen and other extracellular components. These fibroblasts are also involved in the synthesis of extracellular matrix, along with the secretion of various growth factors such as transforming growth factor (TGF-β1) and basic fibroblast growth factor (b-FGF) which are capable of inducing hyaluronic acid secretion and matrix deposition [17]. In a study examining the role of fibroblasts in wound healing, one of which was *H. italicum* EOs, it was found that this oil can induce fibroblasts to induce type I and III collagen deposition during wound healing, thereby accelerating the wound repair process [12], [17], [21]. In the other study, *H. italicum* EOs (0.01%) significantly inhibited the production of collagen I and III [1].

As the main component that strengthens extracellular tissue, collagen plays an important role in wound healing. There are studies that examine collagen by looking at the hydroxyproline content. Hydroxyproline is an amino acid that plays a role in the synthesis, localization and maturation of collagen. In a study, there was a significant increase in the hydroxyproline content in the group treated with *H. italicum* EOs [12], [21].

As a treatment agent for wound healing, *H. italicum* EOs must use a vehicle to make treatment more effective. The most commonly used vehicle options are those based on emulsions and gels. The emulsion consists of 2 phases, namely as a controlled release system, where the substance is stored in the internal phase, then released in the external phase, and absorbed afterwards. While the gel has hydrophilic properties, so there is a risk of removing the active substance if it absorbs water. In terms of migration of the active substance to the skin, it is easier to use a gel, because of its hydrating effect. However, if the substance is hydrophobic and interacts with the hydrolipidic layer of the skin, then an emulsion is better than a gel. Thus an emulgel containing both is formed, which provides the advantages of good dispersibility, no fat and better stability of the active substance (especially hydrophobic) [19].

In a study comparing the effect of *H. italicum* ointment with other ointments, it showed a significant wound contraction from the 7th to the 21st day compared to other groups. On the 21st day there was an average wound contraction of 99.32% in the group given *H. italicum* ointment. Then also the histopathological results validate the wound healing effect of this ointment because it shows a clear increase in collagen fiber density in the post-excision scar [21].

With various benefits that have been reviewed from several selected journals, *H. italicum* EOs is proven to be safe and beneficial in wound healing. However, information regarding its use is still very limited, so further research is needed to maximize the benefits of this oil, so that it can become an alternative treatment agent in wound healing in the future.

4. CONCLUSION

Based on the studies that have been reviewed, *H. italicum* EOs shows good wound healing ability when viewed from various aspects such as its chemical components which are also influenced by various factors, its biological activity, and its effect on injured body tissues. This is also inseparable from the accuracy of the type of vehicle used for treatment, so that it can produce a good wound healing effect. In this study, of course, there are several limitations, such as the evaluation of heterogeneous measurements, varying treatment times, and varied research subjects.

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