Exploring the Anti-Inflammatory Potential of Ethyl Acetate Extract of *Lobophytum* sp. from Southeast Sulawesi Sea

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Abstract

Compounds derived from marine organisms have gained significant interest for their potential therapeutic effects on inflammatory diseases. Lobophytum sp., a soft coral species, has demonstrated anti-inflammatory properties by inhibiting the production of TNF-α and IL-1β, two crucial cytokines involved in the inflammatory process. This study assessed the anti-inflammatory potential of the ethyl acetate extract of Lobophytum sp. (LEA) using in vivo models. The extract was obtained through maceration using ethyl acetate. Phytochemical screening identified various bioactive compounds, including flavonoids, alkaloids, and terpenoids, known for their ability to modulate various biological pathways. To evaluate the anti-inflammatory effects, LEA was tested in a mouse model of xylene-induced ear edema, a common assay for inflammation. Diclofenac sodium, a well-known anti-inflammatory drug, was used as a positive control. LEA was administered at a dose of 0.05, 0.1, and 0.2 mg.mL^{-1}. The results revealed that LEA significantly reduced the swelling of the ear and suppressed the release of key pro-inflammatory cytokines, such as TNF-α and IL-1β. The expression levels of IL-1β and TNF-α were measured using the ELISA reader method. These findings suggest that the anti-inflammatory effects of LEA are likely mediated by the modulation of inflammatory signalling pathways, potentially involving the inhibition of cytokine production. The most effective dose in this study was 0.1 mg.mL^{-1}. Overall, the results highlight the promising therapeutic potential of LEA as a natural anti-inflammatory agent. Further investigation is needed to explore the underlying mechanisms of action and evaluate its clinical applications for treating inflammatory diseases.

Keywords: *Lobophytum sp. (LEA), Inflammation, Cytokine, TNF-α, IL-1β.*

Introduction

Soft corals play a crucial role in marine ecosystems and have attracted considerable attention due to their diverse chemical compositions and biological activities (Putra *et al.*, 2016; Nguyen *et al.*, 2022). Among them, the Lobophytum genus has been recognized as a valuable source of bioactive compounds with significant pharmacological potential. Primarily distributed in tropical and subtropical marine habitats, secondary metabolites derived from *Lobophytum* sp. have exhibited various biological activities, including antimicrobial, anticancer, and anti-inflammatory effects (Putra *et al.*, 2016; Arunasalam *et al.*, 2021). Notably, *Lobophytum* sp. species found in the Southeast Sulawesi Sea remain primarily unexplored despite their potential as a reservoir of novel bioactive compounds for therapeutic applications (Putra *et al.*, 2016; Zhang *et al.*, 2019).

Recent research has shown that marine-derived metabolites can modulate inflammatory pathways by suppressing pro-inflammatory cytokine release (Jegani *et al.*, 2024). *Lobophytum* sp., known for its extensive chemical diversity, has demonstrated promising anti-inflammatory properties, although its specific mechanisms of action are still poorly understood. Previous studies, including our research on Lobophytum as an anti-rheumatoid agent (Fristiohady *et al.*, 2024), have provided initial evidence supporting the role of this soft coral in

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inflammation regulation, reinforcing the need for further exploration of its bioactive metabolites. In particular, soft corals from the Southeast Sulawesi Sea present an opportunity to investigate further the anti-inflammatory potential of Lobophytum, which may lead to the discovery of novel compounds for managing inflammatory diseases (Roy *et al*., 2019; Fristiohady *et al*., 2024; Ke *et al*., 2025).

Inflammation is a crucial biological response to harmful stimuli and plays a significant role in developing various diseases, including autoimmune disorders, cardiovascular conditions, and neurodegenerative diseases (Chen *et* al., 2017; Chavda *et al*., 2024). While acute inflammation is essential for tissue repair and immune defence, prolonged or chronic inflammation can damage tissue and contribute to disease progression (Chen *et al*., 2017; Chavda *et al*., 2024; Hannoodee and Nasuruddin, 2024). Cytokines, small signalling molecules produced by immune cells, are central regulators of the inflammatory response. Among them, tumor necrosis factor-alpha (TNF-α) and interleukin-1 beta (IL-1β) are two key pro-inflammatory cytokines closely linked to the onset and progression of inflammation (Chen *et al*., 2017; Bhol *et al*., 2024). Increased levels of TNF-α and IL-1β have been associated with various inflammatory diseases, making them important targets for anti-inflammatory treatments (Chen *et al*., 2017; Liao *et al*., 2020).

TNF-α and IL-1β are primarily secreted by activated immune cells, such as macrophages, monocytes, and dendritic cells, and they act on various target cells including endothelial cells, fibroblasts, and neutrophils. Through these interactions, both cytokines regulate immune cell recruitment, enhance vascular permeability, and promote the release of additional inflammatory mediators, thereby amplifying the inflammatory response (Chen *et al*., 2017; Kany *et al*., 2019; Chen *et al*., 2024).

Conventional approaches to inflammation management predominantly involve using nonsteroidal anti-inflammatory drugs (NSAIDs) and corticosteroids. However, these treatments are often associated with undesirable side effects, underscoring the need for safer and more effective alternatives (Bindu *et al*., 2020). Marine natural products have emerged as promising candidates for anti-inflammatory agents due to their potential efficacy and low toxicity (Elbandy, 2022). In particular, soft corals have been found to contain bioactive compounds that can regulate key inflammatory mediators, such as cytokines, making them a valuable resource for the discovery and development of new anti-inflammatory therapies (Putra *et al.*, 2016; Yang *et al.*, 2023).

This study investigated the anti-inflammatory activity of Lobophytum soft corals collected from the Southeast Sulawesi Sea by evaluating their potential to suppress inflammation in a mouse model of xylene-induced ear edema. The xylene-induced ear edema model is a well-established method for simulating acute inflammatory responses, making it helpful in evaluating the anti-inflammatory potential of various compounds (Sun *et al*., 2018). Additionally, this study investigated the impact of Lobophytum extracts on regulating pro-inflammatory cytokines, specifically TNF-α and IL-1β, using the enzyme-linked immunosorbent assay (ELISA) technique. This method allows for quantitative analysis of cytokine levels following treatment, providing valuable insights into the molecular mechanisms underlying the anti-inflammatory effects of Lobophytum extracts (Alhajj *et al*., 2023).

Reducing TNF-α and IL-1β levels is a key therapeutic approach for managing inflammatory responses. Several existing anti-inflammatory treatments, including biologic agents that specifically target TNF-α (such as infliximab and etanercept) and IL-1β (such as anakinra), have been developed to inhibit these cytokines and are commonly used in the treatment of autoimmune diseases and chronic inflammatory. However, these therapies have potential side effects, emphasizing the need for safer alternative disorders (Dinarello *et al*., 2012; Li *et al*., 2021). Natural products derived from marine organisms offer a promising solution, as they have the potential to regulate pro-inflammatory cytokine levels effectively while minimizing adverse effects.

This study aims to establish scientific evidence supporting the anti-inflammatory potential of Lobophytum soft corals and their role in modulating inflammatory cytokine responses. By uncovering the mechanisms through which these marine-derived compounds regulate inflammation, this research seeks to contribute to developing new therapeutic approaches for treating inflammatory diseases.

Materials and Methods

The marine soft coral *Lobophytum* sp. was collected around Bintang Samudra Marine Education Park, Soropia District, Konawe Regency, Southeast Sulawesi, Indonesia, by SCUBA (Self Contained Underwater Breathing Apparatus) diving. *Lobophytum* sp. was classified by Biology Laboratory, Faculty of Teacher Training and Education, Halu Oleo University, Indonesia. The soft coral was cleaned from dirt and animals attached to its surface, washed with clean water, and soaked with ice cubes for 24 hours. Subsequently, the soft coral was cut into small pieces for compound extraction.

Extraction and preliminary identification of chemical constituents

The soft coral *Lobophytum* sp. was macerated with ethyl acetate (Merck®, Germany) in a tightly sealed container for three days. The ethyl acetate extract was then filtered using Whatman filter paper (Cytiva, Sweden) and concentrated using a rotary evaporator (Buchi®, Switzerland) at 50°C until crude extract was obtained (Fristiohady *et al*., 2024). The chemical composition of the extract was analyzed using methods reported in the literature. For the alkaloid test, a 1 mL sample was dissolved in 10 mL of chloroform ($CHCl_3$), followed by the addition of 4 drops of ammonium hydroxide (NH_4OH). Then, 10 drops of 2M sulfuric acid (H_2SO_4) were added, and the mixture was shaken. The aqueous layer was separated into another test tube, and Dragendorff's reagent was added, resulting in a red-orange precipitate, which indicated the presence of alkaloids or peptides (Jiangseubchatveera *et al*., 2017; Kancherla *et al*., 2019). For the terpenoid test, a 1 mL sample of the extract was mixed with 5 drops of chloroform and 5 drops of acetic anhydride, followed by 10 drops of sulfuric acid (H_2SO_4). The presence of terpenoids was confirmed by the formation of a brown or violet ring (Kancherla *et al*., 2019). A 1 mL sample of the extract was mixed with magnesium powder and 10 drops of hydrochloric acid (HCl) for the flavonoid test. A positive result was indicated by foam formation and a color change to transparent orange, pink, or red (Jiangseubchatveera *et al*., 2017).

In-vivo antiinflammatory test

Mice were acclimatized for one week and were provided food and water *ad libitum*. Routine observations were conducted to monitor their general condition, and the mice were weighed regularly following acclimatization; 24 mice were randomly assigned to six groups: the sham group (normal control), the naïve group (receiving 0.5% Na-CMC), the positive control group (receiving 0.2 mg of diclofenac sodium), and three treatment groups receiving 0.05, 0.1, and 0.2 $mg.mL^{-1}$ of ethyl acetate soft coral extract of *Lobophytum* sp., respectively. On day 8, an inflammatory response was induced using topical xylene. A 30 µL dose of xylene was applied to the inner and outer surfaces of the right ear in five groups using a micropipette. Ear thickness was measured 15, 30, and 45 min post-induction using an ultrasonic thickness gauge meter. After 45 minutes, the mice were sacrificed, and blood samples were collected for further analysis (Lee *et al*., 2021; Sahidin *et al*., 2023). The percentage of inflammation was calculated using the formula:

$$\% \text{ Inflammation} = [(D_t - D_0) / D_0] \times 100$$

Dt represents the ear thickness at a given time (t), and D_0 is the initial thickness at time zero. The percentage of inflammatory inhibition was determined using:

$$\% \text{ Inhibition} = [(a - b) / a] \times 100$$

Where a is the percentage of inflammation in the sham group, and b is the percentage of inflammation in the treatment group.

Measurement of levels of pro-inflammatory cytokines IL-1β and TNF-α

Following xylene induction, which resulted in edema, the mice received treatments as follows: ethyl acetate soft coral extract of *Lobophytum* sp. at doses of 0.05, 0.1, and 0.2 $mg.mL^{-1}$; 0.5% Na-CMC as the negative control; and 0.2 mg of diclofenac sodium as the positive control. Blood samples were collected once via cardiac puncture 45 minutes after xylene induction to assess IL-1β and TNF-α levels. The collected blood was transferred into tubes containing EDTA anticoagulant and processed according to the ELISA kit protocol. IL-1β and TNF-α levels were determined by measuring sample absorbance at 450 nm using the ELISA kit (Bioassay Technology Laboratory®) (Kara *et al*., 2018; Wang *et al*., 2023).

Statistical analysis

Data collected was presented as mean±SD and analyzed statistically using one-way ANOVA (Analysis of Variances) with a significance of 0.05.

Results and Discussion

The soft coral *Lobophytum* sp. was collected from Soropia Waters, Konawe Regency, Southeast Sulawesi. Its identification was conducted at the Biology Education Department Laboratory, Faculty of Teacher Training and Education, Halu Oleo University, Kendari, under registration number 2515/UN29. 18.1/PG/2021. *Lobophytum* sp. is characterized by its colony color, which varies between yellow, greenish-yellow, and grey. It has a finger-shaped upper surface and a broad capitulum positioned perpendicular to the capitulum surface, as Manuputty (2016) described.

Phytochemical screening of Lobophytum sp.

Phytochemical screening of *Lobophytum* sp. showed positive results for alkaloids, terpenoids, and flavonoids, indicating the presence of these bioactive compounds in the extract. The results of the screening are presented in Table 1. The presence of alkaloids suggests that *Lobophytum* sp. contains nitrogen-containing compounds, which are known for

their diverse pharmacological activities, including antimicrobial, anti-inflammatory, and anticancer properties (Putra *et* al., 2016; Wibowo *et al*., 2021; Sahidin *et al*., 2022). Detecting terpenoids confirms the presence of isoprene-based compounds, which play an essential role in chemical defence mechanisms. Terpenoids are widely recognized for their anti-inflammatory, antioxidant, and anticancer properties, making them valuable candidates for therapeutic applications (Putra *et* al., 2016; Siddiqui *et* al., 2024; Câmara *et al.,* 2024). The positive flavonoid test indicates the presence of polyphenolic compounds, which exhibit potent antioxidant and anti-inflammatory activities (Putra *et al*., 2016; Ullah *et al*., 2020). As summarized in Table 1, these findings highlight *Lobophytum* sp. as a promising source of bioactive compounds with potential pharmaceutical applications, particularly in developing anti-inflammatory and antioxidant agents.

Anti-Inflammatory activity of Lobophytum sp. ethyl acetate extract in a mice ear edema model

Inflammation is a biological response to harmful stimuli, including chemical irritants such as xylene, which induces edema by releasing inflammatory mediators. Synthetic anti-inflammatory drugs like diclofenac sodium are widely used but may cause adverse effects with long-term use. Therefore, exploring natural compounds with potential anti-inflammatory properties is essential (Singsai *et al*., 2020; Daud *et al.,* 2025). *Lobophytum* sp., a soft coral known to contain various bioactive compounds, has been investigated for its pharmacological properties, including anti-inflammatory effects (Putra *et* al., 2016; Fristiohady *et al*., 2024). This study aimed to evaluate the efficacy of the ethyl acetate extract of *Lobophytum* sp. in reducing xylene-induced ear edema in mice.

A xylene-induced ear inflammation test was conducted in mice to assess the anti-inflammatory potential of the ethyl acetate extract of *Lobophytum* sp. After xylene induction, the mice were treated with *Lobophytum* sp. extract at 0.05 mg.mL^{-1}, 0.1 mg.mL^{-1}, and 0.2 mg.mL^{-1}. The negative control group received 0.5% Na-CMC, while the positive control group was administered diclofenac sodium. Ear thickness was measured post-induction to evaluate the extent of inflammation. The results showed that the negative control group (Na-CMC) exhibited no reduction in ear thickness, indicating persistent edema. In contrast, the positive control (diclofenac sodium) significantly reduced inflammation compared to the negative control. Treatment with *Lobophytum* sp. extract significantly reduced xylene-induced edema compared to the negative control (*p*-value < 0.05), with the most notable effect observed in the 0.1 mg.mL^{-1} group. The 0.2 mg.mL^{-1}and 0.05 mg.mL^{-1} groups also showed reduced inflammation (Figure 1, Table 2). However, no significant difference was found between the 0.1 mg.mL^{-1} and 0.2 mg.mL^{-1} groups (*P*> 0.05), indicating that both concentrations provided similar anti-inflammatory effects. All extract-treated groups exhibited significantly lower anti-inflammatory effects than the positive control (*P* <0.05), suggesting that *Lobophytum* sp. extract has anti-inflammatory properties. However, its efficacy was not as strong as that of diclofenac sodium.

Ear edema is primarily caused by the synergistic action of inflammatory mediators that increase vascular permeability and promote blood flow. The anti-inflammatory activity of *Lobophytum* sp. extract may be attributed to its bioactive compounds, including flavonoids, alkaloids, and terpenoids, which are known for their anti-inflammatory properties. The findings suggest that *Lobophytum* sp. extract exhibits significant anti

Table 1. Phytochemical test results phytochemical screening of *Lobophytum* sp.

Chemical test	Result
Alkaloid	+
Flavonoid	+
Terpenoid	+

(+) = present

Table 2. Mice's ear edema thickness. Data is presented as mean±SD

Sample	Ear Edema (mm)			
	Pre-induction	Post-treatment with xylene		
		15 minutes	30 minutes	45 minutes
Control (-)	0.95±0.01	2.84±0.02	2.86±0.02	2.85±0.03
Control (+)	0.94±0.01	2.84±0.01	1.56±0.02	0.97±0.02
LEA 0.05 mg.mL^{-1}	0.95±0.02	2.84±0.02	2.27±0.03	1.77±0.03
LEA 0.1 mg.mL^{-1}	0.94±0.02	2.82±0.02	1.75±0.04	1.28±0.05
LEA 0.2 mg.mL^{-1}	0.94±0.01	2.83±0.02	1.77±0.03	1.30±0.02

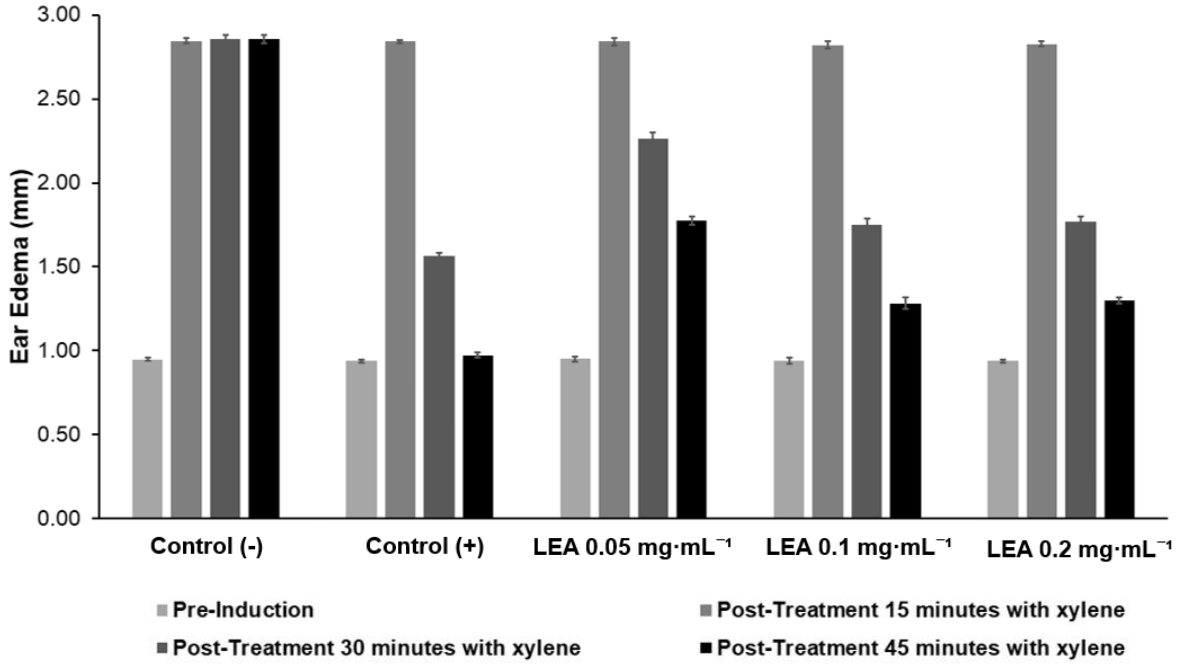

Figure 1. Mice ear thickness edema pre-xylene induction and post-treatment with the test samples. Data is presented as mean±SD (n=3)

inflammatory activity comparable to diclofenac sodium, particularly at concentrations of 0.1 mg.mL^{-1} and 0.2 mg.mL^{-1}. The bioactive compounds present in the extract may exert their effects by modulating inflammatory pathways, such as inhibiting cyclooxygenase (COX) enzymes, suppressing pro-inflammatory cytokines, or stabilizing cell membranes (Bakhsh *et al*., 2024; Fristiohady *et al*., 2024). Further studies are needed to elucidate the exact mechanism of action and to determine the potential of *Lobophytum* sp. extract as an alternative therapeutic agent for inflammatory conditions.

Based on Figure 2, the percentages of inflammation inhibition, from highest to lowest, are as follows: the positive control group (54.70%), the 0.1 mg.mL^{-1} extract group (46.25%), the 0.2 mg.mL^{-1} extract group (45.48%), the 0.05 mg.mL^{-1} extract group (29.79%), and the negative control group (0%). The 0.1 mg.mL^{-1} extract group demonstrated a notable anti-inflammatory effect, with a percentage of inhibition nearly equal to that of the positive control group (P< 0.05). A higher inhibition percentage reflects a greater capacity to mitigate xylene-induced inflammation, reinforcing the potential of *Lobophytum* sp. extract as an anti-inflammatory agent. Our findings align with previous reports on the anti-inflammatory properties of *Lobophytum* sp., as demonstrated in an in vivo study using a Freund's Complete Adjuvant-induced arthritis model, where the ethyl acetate extract administered at doses of 50, 100, and 200 mg·kg^{-1} BW produced significant anti-arthritic and anti-inflammatory effects, with the 200 mg·kg^{-1} BW dose showing the strongest activity (Fristiohady *et al*., 2024).

The anti-inflammatory activity of *Lobophytum* sp. extract may be attributed to its bioactive constituents, which are known to modulate inflammatory pathways by inhibiting pro-inflammatory cytokines and reducing oxidative stress. Flavonoids exert their anti-inflammatory effects through multiple mechanisms, including the inhibition of neutrophil degranulation and suppression of cyclooxygenase (COX) and lipoxygenase (LOX) enzyme activities, thereby preventing the formation of prostaglandins, which are key mediators of inflammation (Panche *et al*., 2016; Shamsudin *et al.*, 2022). Alkaloids contribute to anti-inflammatory activity by suppressing histamine release from mast cells (Javeed *et al*., 2024) and reducing monocyte TNF-α secretion (Yamazaki and Kawano, 2011). Meanwhile, terpenoids reduce inflammation by modulating cell signalling pathways, such as NF-κB activation (de las Heras and Hortelano, 2009).

Ethyl acetate extract of Lobophytum sp. reduces pro-inflammatory cytokine levels

The present study investigates the anti-inflammatory potential of the marine soft coral *Lobophytum* sp., collected from the Southeast Sulawesi Sea, focusing on its ability to suppress key pro-inflammatory cytokines, namely tumor necrosis factor-alpha (TNF-α) and interleukin-1 beta (IL-1β). Our findings demonstrate that *Lobophytum* sp.

exhibits significant anti-inflammatory activity, highlighting its potential as a natural source for developing therapeutic agents targeting inflammatory diseases (Fristiohady *et al*., 2024).

Inflammation is a fundamental biological response to injury or infection. However, when dysregulated, it can contribute to chronic diseases such as arthritis, cardiovascular diseases, and autoimmune disorders. Pro-inflammatory cytokines, including TNF-α and IL-1β, are critical in initiating and propagating the inflammatory response. Overproduction of these cytokines has been linked to the pathogenesis of numerous inflammatory disorders. Consequently, modulating the production of these cytokines presents a promising therapeutic strategy for treating such conditions (Chen *et al*., 2017; Chavda *et al*., 2024).

Further analysis was conducted using serum samples collected from treated mice to elucidate the potential mechanism underlying the anti-inflammatory properties of the ethyl acetate extract of *Lobophytum* sp. Serum levels of IL-1β and TNF-α were measured for each group (Figure 3, Table 3). Mice treated with Na-CMC (negative control) exhibited significantly elevated levels of TNF-α and IL-1β. In contrast, mice treated with diclofenac (positive control) displayed markedly reduced TNF-α and IL-1β levels, approaching normal physiological values. Similarly, treatment with the ethyl acetate extract of *Lobophytum* sp. led to a significant reduction in TNF-α and IL-1β levels, suggesting a possible mechanism involving the inhibition of TNF-α and IL-1β protein expression, thereby mitigating inflammation.

Furthermore, our in vitro experiments demonstrated a dose-dependent inhibition of TNF-α and IL-1β production following treatment with the *Lobophytum* sp. extract. These findings align with previous studies reporting the anti-inflammatory properties of marine soft corals, which produce bioactive compounds capable of modulating immune responses. Notably, bioactive metabolites isolated

Table 3. TNF-α and IL-1β levels in each group. Data are presented as mean±SD

Sample	TNF-α level (pg.mL^{-1})	IL-1β level (pg.mL^{-1})
Normal	63.17±1.80	10.89±0.60
Control (-)	334.91±2.07	97.76±1.03
Control (+)	68.56±0.76	14.90±0.66
LEA 0.05 mg.mL^{-1}	271.81±1.22	81.79±1.70
LEA 0.1 mg.mL^{-1}	163.26±2.17	46.99±1.77
LEA 0.2 mg.mL^{-1}	185.47±1.05	66.66±0.70

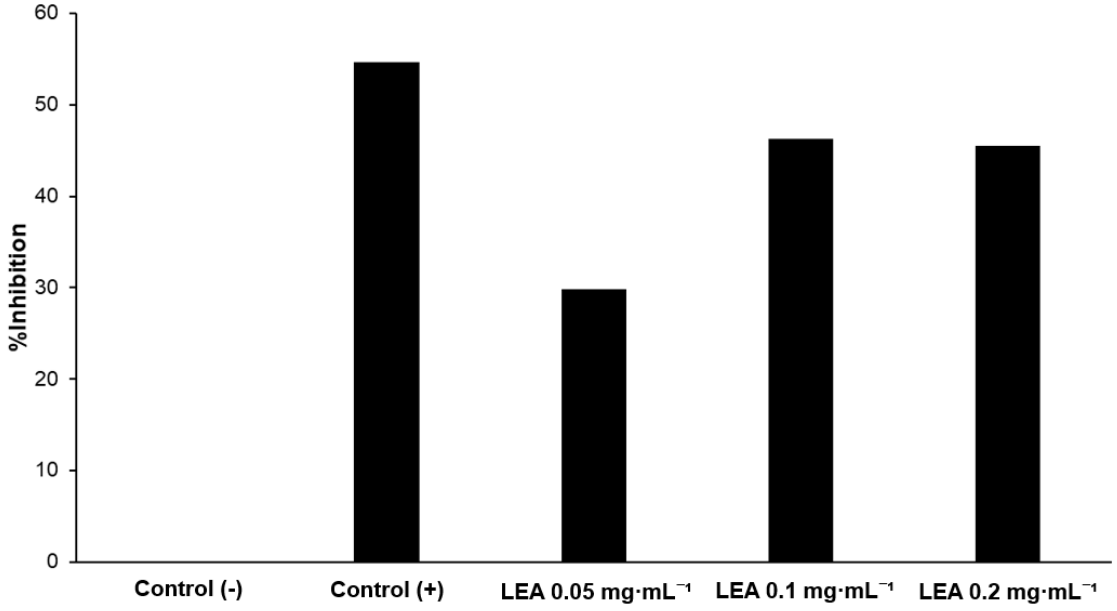

Figure 2. Percentage of Inhibition of Ear Inflammation in Mice.

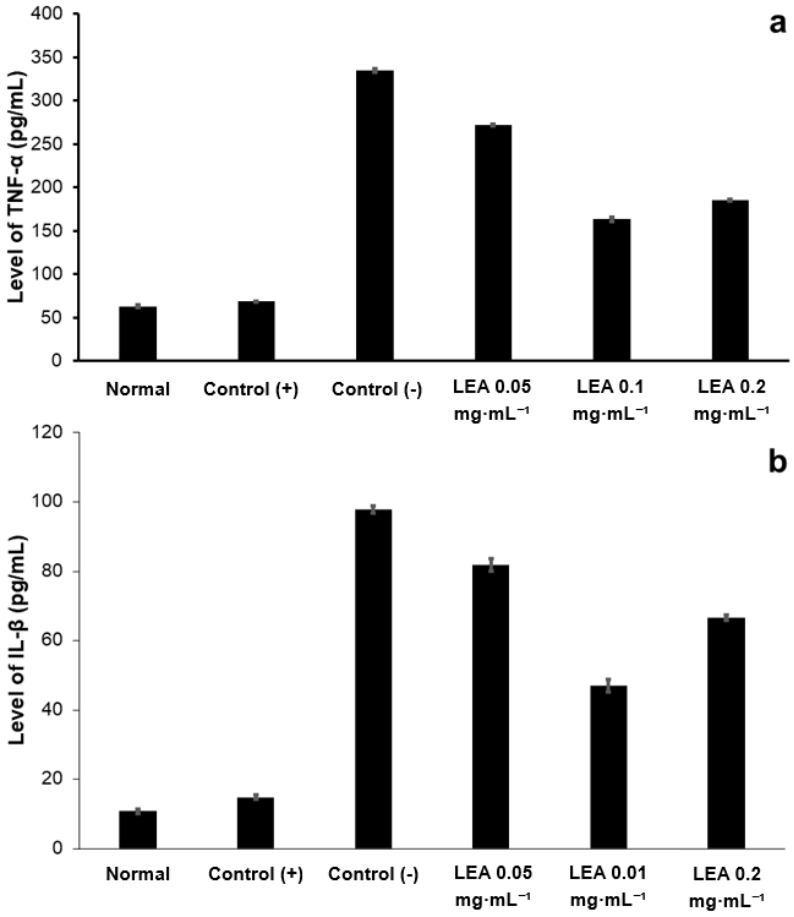

Figure 3. The (a) TNF-α; and (b) IL-β levels. Data are presented as mean±SD

from marine organisms have been shown to regulate the expression of pro-inflammatory cytokines by targeting signalling pathways such as nuclear factor kappa B (NF-κB) and mitogen-activated protein kinases (MAPK), both of which are crucial in the synthesis of TNF-α and IL-1β (Thao *et al*., 2014; Lee *et* al., 2024).

The suppression of TNF-α and IL-1β production observed in this study may be linked to secondary metabolites in *Lobophytum* sp., which could serve as a potential source of novel anti-inflammatory compounds. While the exact mechanism remains unclear, several pathways may be involved. One possibility is the inhibition of NF-κB, a key transcription factor that regulates pro-inflammatory cytokines. By blocking NF-κB activation, compounds from *Lobophytum sp.* may prevent TNF-α and IL-1β upregulation, thereby reducing inflammation. Another potential mechanism is the modulation of the MAPK signalling pathway, which involves kinases such as p38, JNK, and ERK. Marine-derived compounds have been shown to influence MAPK activation, which may contribute to their anti-inflammatory effects (Cuong *et al*., 2014; Bakhsh *et al*., 2024). Additionally, *Lobophytum* sp. may exert antioxidant effects, mitigating oxidative stress that often exacerbates inflammation. By reducing ROS levels, these compounds could help suppress inflammatory pathways and cytokine production (Thao *et al*., 2015; Mu'nisa, 2023).

Several marine soft coral species have been studied for their anti-inflammatory properties, and *Lobophytum* sp.'s bioactivity is consistent with other marine organisms' findings. For example, soft corals from the genera *Sinularia* and *Sarcophyton* exhibit potent anti-inflammatory effects by inhibiting cytokine production. Diverse bioactive compounds, such as alkaloids, terpenoids, and peptides, likely contribute to these effects (Tanod *et al*., 2019; Nguyen *et al*., 2022).

Although *Lobophytum* sp. shares similarities with other soft corals in its anti-inflammatory activity, each species produces distinct metabolites, which may lead to differences in their mechanisms of action. Given the vast chemical diversity of marine

soft coral metabolites, further investigation is needed to identify the specific compounds in *Lobophytum sp.* responsible for its bioactivity and evaluate their potential as drug candidates.

The suppressing of TNF-α and IL-1β by *Lobophytum* sp. highlights its potential as a therapeutic agent for inflammatory diseases. While TNF-α inhibitors like infliximab and adalimumab are effective, they are costly and associated with side effects. Natural compounds from marine organisms, including *Lobophytum* sp., could offer a more affordable and potentially safer alternative, with advantages such as better bioavailability and the ability to target multiple inflammatory pathways. Additionally, *Lobophytum* sp. may have therapeutic potential in conditions where IL-1β plays a key role, such as gout, osteoarthritis, and inflammatory bowel disease.

However, several aspects require further investigation. The molecular mechanisms underlying TNF-α and IL-1β suppression remain unclear, necessitating studies using gene expression analysis and protein assays. Since this study was conducted in vitro, in vivo validation in animal models is crucial to assess bioavailability, toxicity, and long-term effects. Furthermore, isolating and identifying the specific bioactive compounds in *Lobophytum* sp. using techniques like HPLC and MS will be essential to determine their therapeutic potential and support drug development.

Conclusion

In conclusion, the marine soft coral *Lobophytum* sp. from the Southeast Sulawesi Sea demonstrates significant anti-inflammatory activity by suppressing the production of pro-inflammatory cytokines TNF- α and IL-1β. In this study, the 0.1 $mg{\cdot}mL^{-1}$ dose of the ethyl acetate extract showed the strongest activity in reducing the expression levels of both cytokines. These findings highlight the potential of marine soft corals as a promising source of novel anti-inflammatory compounds. Given the rising prevalence of chronic inflammatory diseases, developing marine-derived therapeutic agents represents a valuable direction for future research. Further studies on the isolation of active metabolites and the elucidation of their mechanisms of action will be essential to advance *Lobophytum sp*. as a viable therapeutic candidate for inflammatory conditions.

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