# ANTIMICROBIAL ACTIVITY ASSESSMENT OF FATTY ACIDS EXTRACT FROM VIETNAM VIRGIN COCONUT OIL BY LIPASE FROM ASPERGILLUS NIGER IN ANTIBIOTIC-RESISTANCE BACTERIAL TREATMENT

**Doan Ngoc Tra My*, Phan Thi Cuc**
*The University of Danang - University of Science and Technology*

**ARTICLE INFO**

**Received:** 09/4/2025
**Revised:** 10/8/2025
**Published:** 11/8/2025

**KEYWORDS**

Virgin coconut oil
Lipase
*Aspergillus Niger*
Antibiotic resistance
*Staphylococcus Aureus*

**ABSTRACT**

This study aims to optimize the incubation time and enzyme concentration of lipase from *Aspergillus niger* for the extraction of fatty acids from virgin coconut oil. It also evaluates the antibacterial activity of the extracted samples against *Staphylococcus Aureus* and *Methicillin-Resistant Staphylococcus Aureus* using the agar diffusion method. The results show fatty acids extracted using lipase at an enzyme concentration of 1.5% and an incubation time of 48 hours achieved a hydrolysis degree of 20.88%. These sample demonstrated strong antibacterial activity against *S. aureus* and *Methicillin-Resistant Staphylococcus Aureus* when compared to chemical methods and the commercial antibiotic Vancomycin. Importantly, the fatty acids extract showed a lower minimum inhibitory concentration against *Methicillin-Resistant Staphylococcus Aureus* than *S. aureus* (0.313% vs 1.25%). These findings suggest that enzymatically extracted fatty acids offer a sustainaible and efective antibacterial alternative, particularly valuable in addressing antibiotic-resistant infection.

# ĐÁNH GIÁ HOẠT TÍNH KHÁNG KHUẨN CỦA CHIẾT XUẤT AXIT BÉO TỪ DẦU DỪA VIỆT NAM BẰNG ENZYM LIPASE TỪ NẤM *ASPERGILLUS NIGER* TRONG ĐIỀU TRỊ VI KHUẨN KHÁNG KHÁNG SINH

**Đoàn Ngọc Trà My*, Phan Thị Cúc**
*Trường Đại học Bách Khoa - Đại học Đà Nẵng*

**THÔNG TIN BÀI BÁO**

**Ngày nhận bài:** 09/4/2025
**Ngày hoàn thiện:** 10/8/2025
**Ngày đăng:** 11/8/2025

**TỪ KHÓA**

Dầu dừa nguyên chất
Lipase
*Aspergillus Niger*
Kháng kháng sinh
*Staphylococcus Aureus*

**TÓM TẮT**

Nghiên cứu này nhằm tối ưu hóa thời gian ủ và nồng độ enzyme Lipase từ *Aspergillus niger* để chiết xuất axit béo từ dầu dừa nguyên chất. Các mẫu chiết xuất được đánh giá hoạt tính kháng khuẩn đối với vi khuẩn *Staphylococcus aureus* và *Staphylococcus aureus kháng Methicillin* bằng phương pháp khuếch tán trên thạch. Kết quả cho thấy axit béo được chiết xuất bằng lipase ở nồng độ enzyme 1,5% và thời gian ủ 48 giờ đạt mức độ thủy phân 20,88%, thể hiện hoạt tính kháng khuẩn mạnh đối với *S. aureus* và *Staphylococcus aureus kháng Methicillin* khi so sánh với phương pháp hóa học và kháng sinh thương mại Vancomycin. Đáng chú ý, chiết xuất axit béo cho thấy nồng độ ức chế tối thiểu thấp hơn đối với *Staphylococcus aureus kháng Methicillin* so với *S. aureus* (0,313% so với 1,25%). Những phát hiện này cho thấy các acid béo được chiết xuất bằng phương pháp enzym là một giải pháp kháng khuẩn hiệu quả và bền vững, đặc biệt có giá trị trong việc điều trị các nhiễm trùng do vi khuẩn kháng kháng sinh gây ra.

**DOI: https://doi.org/10.34238/tnu-jst.12519**

* Corresponding author. *Email: dntmy@dut.udn.vn*

## 1. Introduction

Acne and eczema are two of the most prevalent dermatological conditions in Vietnam, affecting a significant portion of the population [1]. A primary contributing factor to these skin disorders is an imbalance in the skin microbiome, which facilitates the overgrowth of *Staphylococcus aureus* (S.aureus) in affected individuals, compared to those with healthy skin [2]. Infected acne is commonly treated with topical antibiotics, such as clindamycin combined with benzoyl peroxide (Ancaya, Duac) or erythromycin with benzoyl peroxide (Benzamycin). For severe cases, systemic therapies involving oral antibiotics and anti-inflammatory agents are required. However, when multiple treatment regimens fail to yield improvement, persistent acne may be attributed to the emergence of antibiotic-resistant bacteria-often referred to as superbugs. A prominent example is *Methicillin-Resistant Staphylococcus aureus* (MRSA), which has evolved mechanisms to resist a broad range of commonly prescribed antibiotics. One such mechanism includes the production of β-lactamase, an enzyme that hydrolyzes the β-lactam ring, a core structural component of antibiotics like penicillins and cephalosporins [3]. This enzymatic action renders these antibiotics ineffective, enabling MRSA to survive treatments with penicillin, tetracycline, and clindamycin. As a result, treating MRSA infections now necessitates the use of more potent and expensive antibiotics, such as Vancomycin and Daptomycin, which also require careful monitoring to avoid toxicity and prevent further resistance development. MRSA can spread from infected skin lesions to surrounding healthy skin, intensifying acne outbreaks. It can also be transmitted to other individuals through direct contact or shared personal items, highlighting its significant public health risk. Given the increasing prevalence of MRSA-related infections, there is a growing demand for alternative antimicrobial strategies. Natural antimicrobial agents are of particular interest due to their potential efficacy, affordability, and lower risk of inducing resistance.

Vietnam is also among the world's largest coconut producers, with the coconut industry playing a crucial role in the national economy. Virgin coconut oil (VCO) has been widely used in food, cosmetics, and traditional medicine. One of its key bioactive components is medium-chain fatty acids (MCFAs), including lauric acid, capric acid, and caprylic acid, which exhibit strong antimicrobial properties, making VCO a promising source of natural antibacterial agents [4], [5]. Various studies have explored extraction methods to enhance the antibacterial potential of coconut oil-derived fatty acids. The two primary methods for obtaining fatty acids from VCO are chemical and biological processes. Chemical methods (saponification) offer high extraction efficiency but may compromise bioactivity due to the use of harsh chemicals [6]. In contrast, biological methods (enzymatic hydrolysis) are considered environmentally friendly and may yield bioactive compounds with enhanced efficacy [7], especially helping to preserve the integrity of bioactive components compared to other methods [8]. However, comparative studies on the antibacterial effectiveness of fatty acids extracted via these two approaches remain limited.

Lipases are the key enzymes responsible for lipid hydrolysis, and they can be classified into three main groups based on their origin: microbial, plant and animal sources. While plant- and animal-derived lipases exhibit high hydrolytic efficiency, their industrial applications remain limited due to high production costs and complex extraction processes. In contrast, microbial lipases, particularly those from bacteria and yeast, are easier to produce, more cost-effective, and offer greater stability than their plant and animal counterparts [9]. Bacterial lipases primarily hydrolyze long-chain fatty acids [10], whereas yeast lipases favor short- and medium-chain fatty acids [11]. Several yeast-derived lipases, such as *Candida rugosa*, *Candida antarctica*, *Candida cylindracea*, and *Yarrowia lipolytica*, have been widely applied in industrial processes. Additionally, filamentous fungi such as *Aspergillus niger (A.niger)* hold significant potential for lipase production. This species is known for secreting various extracellular enzymes, including amylases, cellulases, and particularly lipases [12]. Although *A. niger* lipase has demonstrated

potential applications in the food, biotechnology, and lipid-processing industries, its use in coconut oil hydrolysis remains largely unexplored.

This study aims to investigate the hydrolysis conditions (time and concentration) for VCO using lipase enzymes derived from *A. niger*. Additionally, the antibacterial activity of the resulting fatty acids (FA) will be investigated against *S. aureus* and MRSA strains, compared with those obtained through conventional chemical methods. The goal of this research not only contributes to enhancing the economic value of coconut oil but also assess the potential of enzymatic-derived FAs as effective natural alternatives for treating antibiotic-resistant skin infections, thereby contributing to the development of cost-effective and sustainable therapeutic solutions.

## 2. Materials and methods

### *2.1. Chemical agents*

Virgin coconut oil (VCO) was supplied by Luong Quoi Coconut Co., Ltd. (Ben Tre Province, Vietnam). Lipase derived from *Aspergillus niger* (CAS 9001-62-1; 100g) was purchased from Oxford, India. The main chemicals used in this study were potassium hydroxide (KOH), hydrochloric acid (HCl), sodium dihydrogen phosphate dihydrate ($NaH_2PO_4 \cdot 2H_2O$), disodium hydrogen phosphate dodecahydrate ($Na_2HPO_4 \cdot 12H_2O$), absolute ethanol ($C_2H_5OH$), anhydrous sodium sulfate ($Na_2SO_4$), phenolphthalein ($C_{20}H_{14}O_4$), *n*-hexane, and other reagents purchased from GHTECH (China).

### *2.2. Fatty acid extraction by chemical hydrolysis*

The procedure for obtaining the fatty acid mixture of coconut oil involves several steps. Firstly, VCO is combined with ethanol in a three-necked flask in a volumetric ratio of 2:1, followed by gentle heating and stirring. To initiate *chemical hydrolysis,* an equal volume of 33% (w/v) sodium hydroxide (NaOH) was added (1:1), and stirring continues at a speed of 550 rpm until complete dissolution of the solution. The solution is poured into a beaker containing hot saturated NaCl solution and stirred until separation into layers with the appearance of a solid layer on the surface. The solid layer was carefully separated and washed with distilled water, thendissolved again in hot distilled water. To form the fatty acid mixture, 10% HCl is added to the solution until the pH 1. After hydrolysis, the reaction mixture was cooled to room temperature, and the hydrolyzed fatty acids were extracted using *n*-hexane. The organic layer containing free fatty acids was separated from the aqueous phase using a separatory funnel [13].

### *2.3. Fatty acid extraction by biological hydrolysis*

The VCO and gum arabic mixture (oil-to-water ratio = 1/6 (w/v); 12% gum arabic) was emulsified at 10.000 rpm for 5 minutes. A phosphate buffer solution was then added at a 1:4 ratio. The hydrolysis efficiency of lipase was evaluated at different concentrations. Next, the mixture was stirred to fully dissolve the lipase powder. The hydrolysis reaction was incubated in a shaking incubator for 24–48 hours at 37 °C. The hydrolyzed FAs was neutralized by adding an excess amount of KOH and separated with *n*-hexane. The FAs in the lower phase were converted back to their free form by adding an excess amount of HCl 4N solution (pH < 2). FAs were then continuously extracted using *n*-hexane and purified with a rotary evaporator to remove *n*-hexane from the mixture [14]. The hydrolysis degree (HD) was calculated using the following formula:

$$\mathrm{HD} = \frac{VKOH * MKOH * MFAS}{10 * m} \quad (\%) \qquad (1)$$

Where $V_{\mathrm{KOH}}$ : volume of the potassium hydroxide (KOH) solution used for titration (mL); n$M_{\mathrm{KOH}}$: molar concentration of the KOH solution (mol/L); $M_{\mathrm{FAs}}$ : average molecular weight of the fatty acids; $m$ : mass of the sample (g).

### *2.4. Antibacterial activity evaluation*

The antibacterial activity was evaluated using the agar well diffusion method. The study was conducted on *S. aureus* and MRSA, a Gram-positive bacterial strain obtained from the Department of Biotechnology, Faculty of Chemistry Engineering, Da Nang University of Science and Technology. Bacterial suspension was prepared by dissolving bacterial colonies in Mueller-Hinton Broth (MHB) to reach a final concentration of $10^6$ CFU/mL. Different test samples were added to corresponding wells on the agar plates, including: a negative control (DMSO 10% or *n*-hexane), a positive control (Vancomycin 30 µg/mL), virgin coconut oil (VCO), and fatty acid extracts at various dilutions in DMSO (100%, 50%, 25%, 10%) from both enzymatic and chemical extraction methods. The samples were allowed to diffuse into the medium for 2 hours at room temperature, and results were recorded after 24 hours of incubation at 37 °C [13]. The antibacterial activity of the FAs samples using the agar diffusion method was determined using the following formula:

$$DK(mm) = D - d \quad (2)$$

Where D: diameter of the sterile zone, d: diameter of the agar hole.

### *2.5. Determination of the minimum inhibitory concentration (MIC)*

The bacterial suspension was diluted in MHB to a final concentration of $10^6$ CFU/mL. Each well of a 96-well plate was filled with 50 µL of the bacterial suspension and 50 µL of the 10% acid sample, along with acid at various diluted concentrations. A separate control was prepared for color change comparison by replacing the bacterial suspension with the medium. The negative control containe DMSO 10% and the bacterial suspension, while the positive control included Vancomycin 30µg/mL and the bacterial suspension**.** The plate was incubated at 37°C for 24 hours**.** After incubation, 30 µL of 0.015% resazurin was added to each well, followed by an additional 2 hours of incubation at 37°C. The color change in the wells was then observed, and the MIC value was recorded [15].

### *2.6. Experiment analysis*

The variability degree of results was expressed in form of means ± standard deviation (mean ± SD) based on triplicates determinations (n = 3 for replicate plates). The data were statistically analyzed by one-way ANOVA analysis and compared using the least significant difference (LSD) test at $p < 0.0001$(****). It was done to compare between control and treatments, no statistically significant difference (ns).

## 3. Results and discussions

### *3.1. Evalulation of incubation time and concentration of lipase from Aspergillus Niger on the hydrolysis process*

Figure 1 illustrates the effect of lipase concentration and incubation time on the hydrolysis degree percent (HD%) of coconut oil using lipase from *A.niger*. The experiment was conducted with enzyme concentrations ranging from 0% to 2.5% (w/w), and the HD was measured at 24 and 48 hours of incubation. The results indicate that HD% increased with enzyme concentration, showing a positive correlation between lipase concentration and the extent of hydrolysis. At 0% enzyme concentration, minimal hydrolysis was observed, confirming that enzymatic activity is essential for FAs liberation. As the enzyme concentration increased, HD% progressively improved, reaching its peak at 2.5% enzyme concentration. Furthermore, the incubation time significantly influenced the hydrolysis process. At all enzyme concentrations, HD% was consistently higher at 48 hours compared to 24 hours, indicating that prolonged incubation allows for more extensive hydrolysis. However, the rate of increase in HD% between 24 and 48 hours appeared to diminish at higher enzyme concentrations, suggesting that the reaction may be approaching saturation beyond a certain threshold.

These findings suggest that both enzyme concentration and incubation time play crucial roles in optimizing the hydrolysis of coconut oil, with 2.5% lipase concentration and 48 hour incubation yielding the highest HD%. In conclusion, lipase concentration of 1.5% and incubation time of 2 hours were selected and used for subsequent experiments.

The results show that the hydrolysis efficiency is only moderate when compared to previously published studies that employed lipases from other microbial sources [16]. The optimal conditions for the hydrolysis process (buffer ratio, buffer pH, hydrolysis incubation time, etc.) also need to be further studied in order to enhance the extraction of FAs from VCO using *A. niger* lipase.

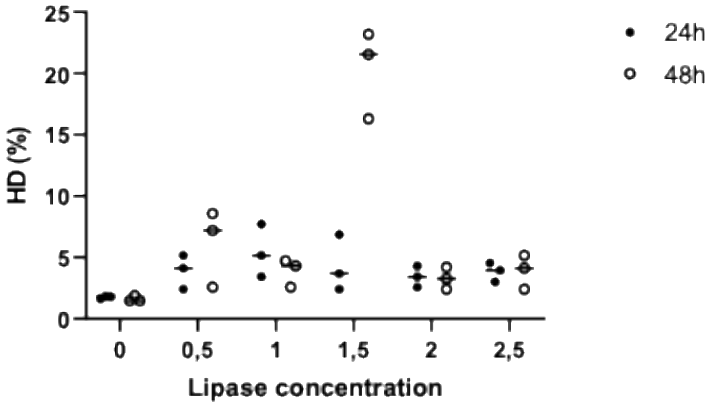


**Figure 1.** *Hydrolysis degree (HD%) of VCO at differnd Lipase concentration with incubation time 24 and 48 hour*

### *3.2. Fatty acid extracts from VCO by Lipase from Aspergillus niger exhibits a high Antibacterial Activiy on Staphyloccocus Aureus*

*Staphylococcus aureus* (*S. aureus*) is a harmful microorganism responsible for skin diseases such as eczema and is also a contributing factor in exacerbating dermatological conditions like psoriasis and acne [17]. Therefore, controlling *S. aureus* on the skin may help reduce the severity of these conditions. This study focuses on evaluating the antibacterial activity of FAs samples extracted from coconut oil using enzymatic and chemical methods against *S. aureus*, as well as virgin coconut oil (VCO), using the agar well diffusion method. Since the FAs were extracted using *n*-hexane, this solvent was used as a negative control. In all antibacterial assays, Vancomycin (30 µg) was used as a positive control.

Figure 2.a and 2.d illustrate that Vancomycin, a commercially available antibiotic, exhibited strong antibacterial activity with an inhibition zone diameter of 19 mm and he negative control (*n*-hexane) showed no inhibition zone confirming that the antibacterial activity observed in the test samples was due to their bioactive properties. While VCO did not produce a clear inhibition zone; however, FAs extracted via *chemical hydrolysis* (CH-100) exhibited antibacterial activity with an inhibition zone of 6 mm. Notably, FAs extracted via Enzymatic hydrolysis (EH-100) using lipase from *A. niger* demonstrated the highest antibacterial activity, equivalent with Vancomycin, with an inhibition zone diameter of 20 mm.

To optimize cost-effectiveness for industrial applications, it is crucial to select a lower concentration that retains antibacterial activity. Therefore, in this experiment, FAs samples extracted via the enzymatic (Figure 2.b) and chemical method (Figure 2.c) were diluted to 100%, 50%, 25%, and 10% using DMSO 10% as a solvent. Consequently, DMSO 10% was used as the negative control in this study. The experience was repeated 3 times and the summary results showed in Figure 2.d. This indicates that FAs extracts at all tested concentrations exhibited antibacterial effects against *S. aureus*. While the concentration of FAs extracted using the chemical method was reduced by half (from 100% to 50%), the diameter of the inhibition zone remained

unchanged (6 mm). However, for an inhibition zone of 20 mm at 100% concentration, the FAs extracted using the enzymatic method exhibited a proportional decrease in inhibition zone size with decreasing concentration. At a concentration of 10%, the antibacterial activity of FAs extracted using the enzymatic method was approximately equivalent to that of FAs extracted using the chemical method at 100%. This results indicates that the FAs extracted by the enzymatic extraction exhibits superior antibacterial efficacy compared to the chemical method, providing valuable insights for cost-effective formulation strategies in pharmaceutical and cosmetic applications.

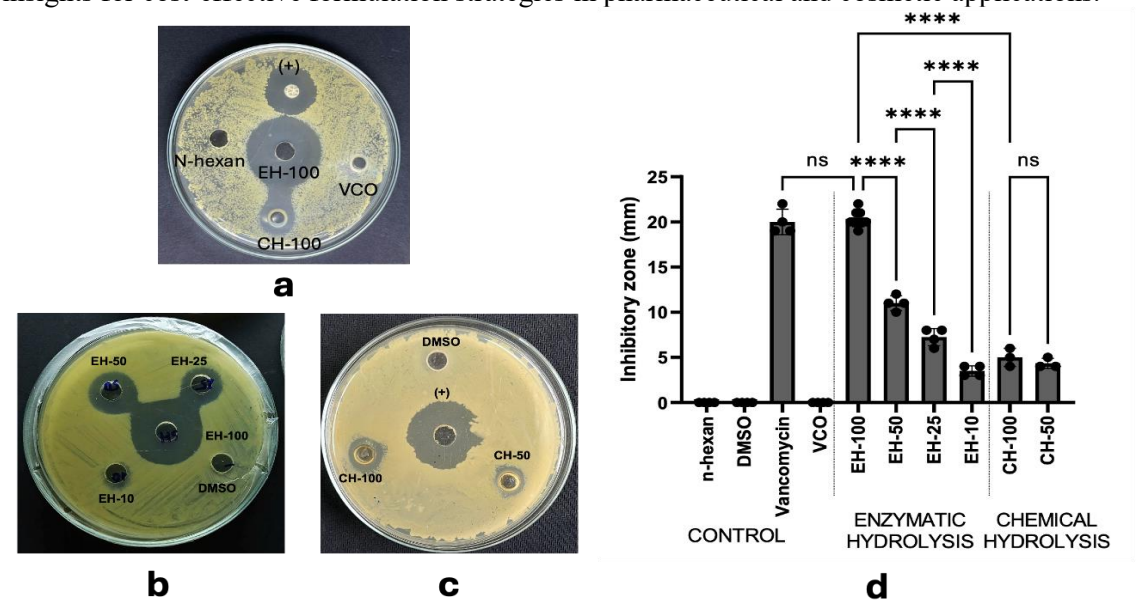


**Figure 2.** *Antimicrobial activity against S.Aureus of Virgin Coconut Oil (VCO), FAs extract by enzymatic hydrolysis (EH) and chemical hydrolysis (CH) at 100% (a) and EH (b), CH (c) at different concentration, Control (+): Vancomycin, Control (-): n-*hexane *or DMSO; Summary graph (d)*

### *3.3. Fatty acid extracts from VCO by Lipase from Aspergillus niger exhibits a high antibacterial activiy on Methicilin-Resistance Staphyloccocus Aureus*

The next aim of study was to assess their potential for FAs extracts in the treatment of antibiotic-resistant infections, *Methicillin-Resistant Staphylococcus aureus* (MRSA) - a highly resistant pathogen causing serious skin and soft tissue infections, offering a promising natural alternative to conventional antibiotics.

The antibacterial activity against MRSA of FAs extracted via chemical hydrolysis (CH) and enzymatic hydrolysis (EH) at, as well as unprocessed VCO, was compared. Figures 3.a and 3.d showed that Vancomycin, a standard antibiotic, was used as a positive control exhibited the large inhibition zone (20 mm), confirming its strong antibacterial activity. Figure 3.d indicates the summary results of Figures 3.b and 3.c. This indicates the FAs extract obtained via enzymatic hydrolysis at 100% concentration (EH-100) exhibited a significant inhibition zone (~21 mm), comparable to vancomycin, indicating potent antibacterial activity. As the concentration of the enzymatic hydrolysate decreased (EH-50, EH-25, EH-10), the inhibition zone progressively reduced, demonstrating a dose-dependent effect. EH-50 still exhibited notable activity (~10 mm), whereas EH-10 showed a much smaller inhibition zone (~5 mm), indicating a significant reduction in antibacterial potency at lower concentrations. Meanwhile, the FAs using chemical hydrolysis at 100 and 50% (CH-100 and CH-50) exhibited significantly lower antibacterial activity than EH-100, with inhibition zones of approximately 5–6 mm. Statistical analysis showed a significant difference in antibacterial activity between enzymatic and *chemical hydrolysis* at equivalent concentrations, suggesting that Enzymatic hydrolysis enhances the bioactive properties of the FAs

(Figure 3.d). No statistically significant difference (ns) was observed between CH-100 and CH-50, indicating that further dilution had minimal impact on antibacterial activity (Figure 3.d).

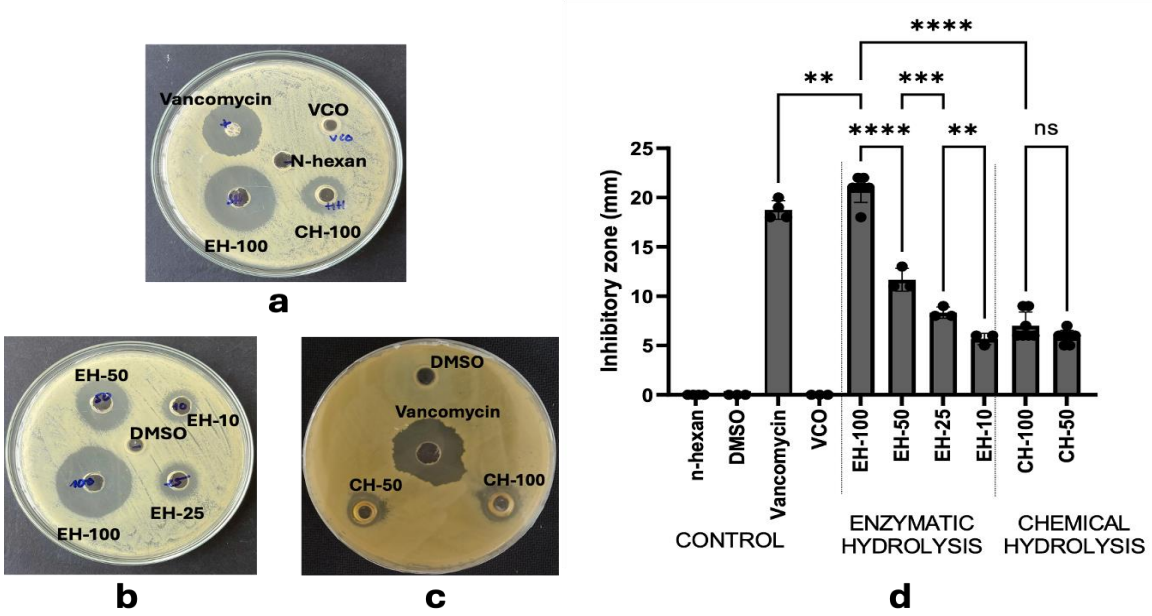


**Figure 3.** *Antimicrobial activity against MRSA of Virgin Coconut Oil (VCO), FAs extract by enzymatic hydrolysis (EH) and chemical hydrolysis at 100% (a) and EH (b), CH (c) at different concentration, Control (+): Vancomycin, Control (-): n-*hexane *or DMSO; Summary graph (d).*

### *3.4. Comparison of MIC of FAs extract by enzymatic and chemical hydrolysis*

Additionally, to confirm the antibacterial concentration, the Minimal Inhibitory Concentration (MIC) test was also conducted. This assay determines the lowest concentration of the FAs extract that effectively inhibits visible bacterial growth, providing a more precise evaluation of its antimicrobial potency.

For *S. aureus*: FAs extracted via *A. niger* lipase hydrolysis exhibited a MIC of 1.25% (Figure 4.a), which is considerably lower than that obtained by *chemical hydrolysis*, 2.5% (Figure 4.c). This indicates that the enzyme-mediated FAs extract exhibits stronger antibacterial activity at a lower concentration, suggesting the potential of this method in controlling the growth of *S. aureus*. For MRSA: The FAs extract obtained through *A. niger* lipase hydrolysis demonstrated an MIC of 0.313%, showing superior antibacterial activity against methicillin-resistant bacteria (Figure 4.b) In comparison, the *chemical hydrolysis* method showed an MIC of 2.5%, highlighting the significantly higher potency of lipase *A. niger* in combating MRSA at much lower FAs concentrations (Figure 4.d).

This highlighted result suggests that certain FAs present in the extract may have a more pronounced disruptive effect on resistant bacterial strains, potentially due to differences in membrane composition or biofilm-forming ability. While previous studies have proposed that medium-chain fatty acids (MCFAs), such as lauric acid, can integrate into the phospholipid bilayer and disrupt membrane integrity, we did not perform mechanistic assays (e.g., membrane permeability, reactive oxygen species generation, or protein denaturation) to validate this in our samples. Additionally, we have not yet identified the specific composition of the extracted FAs that dominate in the extract obtained through enzymatic versus chemical methods, as well as understanding the antibacterial mechanism against MRSA, is crucial for supporting future clinical pharmaceutical applications. This is a significant limitation that may obscure the identification of the most bioactive constituents.

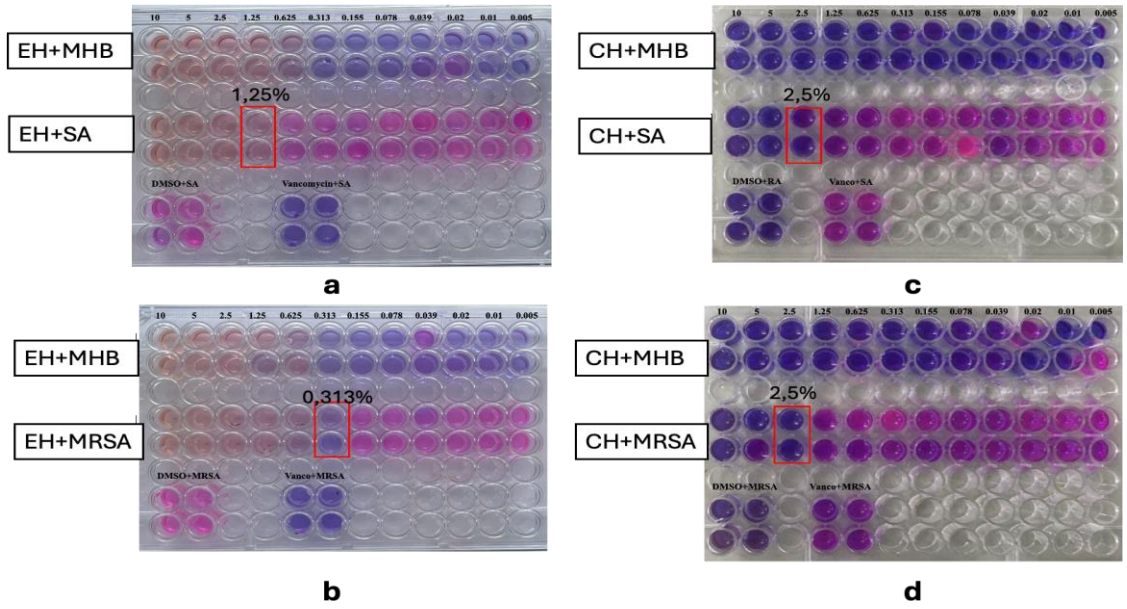


**Figure 4.** *Resazurin-based 96-well plate microdilution method to determine MIC value of FAs extract by enzymatic hydrolysis-EH (a, b) and chemical hydrolysis-CH (c, d) against S. aureus(a, c) or MRSA (b, d)*

However, these results underscore the advantages of using *A. niger* lipase in the hydrolysis of virgin coconut oil to extract FAs with strong antibacterial properties, particularly against antibiotic-resistant bacteria such as MRSA. This approach presents a promising development for the creation of natural antibacterial products, offering a safe and effective alternative in pharmaceutical and cosmetic applications.

## 4. Conclusions

One of the most innovative aspects of this research is the use of lipase from *A. niger* or enzymatic hydrolysis of VCO to obtain FAs. While enzymatic hydrolysis has been studied extensively, lipase from *A. niger* has not been widely used for this purpose despite being a cost-effective, readily available enzyme. The use of *A. niger* lipase offers significant economic advantages over other lipases, especially those derived from more expensive sources. The low cost of the enzyme, combined with its high catalytic efficiency, makes this approach highly attractive for industrial applications, particularly in natural skincare formulations. Our findings indicate that increasing the enzyme concentration and incubation time significantly improves the hydrolysis degree and enhances the release of bioactive FAs. Specifically, at an enzyme concentration of 1.5% and an incubation time of 48 hours, the hydrolysis degree reached 20.88%. These conditions resulted in an inhibition zone of 20 mm against *S. aureus*, a major cause of skin infections such as acnes, eczema, and other dermatological conditions, which surpassed the efficacy of chemical methods (5 mm), meanwhile, unprocessed VCO has not. Furthermore, FAs extracted via enzymatic hydrolysis also exhibited potent antibacterial activity against *Methicillin-Resistant Staphylococcus aureus* , with an inhibition zone of 21 mm, particularly MIC against MRSA is lower than *S. aureus* (0.313% vs 1.25%).

To conclude, this study highlights the superior antibacterial efficacy of enzymatically hydrolyzed FAs, positioning them as a promising natural alternative for addressing antibiotic-resistant infections. The findings underscore the potential of utilizing coconut oil-derived FAs as a sustainable and effective solution in the fight against resistant bacterial strains. This approach opens promising research direction for the development of natural antibacterial products, offering a safe, eco-friendly, and potent alternative for pharmaceutical and cosmetic applications. However, despite these positive results, certain limitations remain, warranting further research. Specifically,

HPLC analysis is needed to identify and quantify the specific FAs within enzymatic hydrolysis extract verus chemical methods. Moreover, assessing the antibiofilm activity against MRSA by using FAs extract is essential for understanding the antibacterial mechanism and resistance in antibiotic-resistant bacterial strains.

**Acknowledgments**

This work was supported by The University of Danang – University of Science and Technology, code number of Project: T2024-02-18.

REFERENCES

[1] T. T. Ha, T. T. Le, and T. T. T. An, "Curent situation and some factors related to hand eczema in medical staff at 108 Military Central Hospital," *Journal of 108 - Clinical Medicine and Pharmacy*, *Special Issue: Scientific Conference on the 70th Anniversary of 108 Military Central Hospital*, 2021, pp. 126–130.

[2] F. Khorvash, F. Abdi, H. H. Kashani, F. F. Naeini, and T. Narimani, "Staphylococcus aureus in acne pathogenesis: A case-control study," *Am. J. Med. Sci.*, vol. 4, no. 11, pp. 573–576, 2012, doi: 10.4103/1947-2714.103317.

[3] P. Nandhini *et al.,* "Recent Developments in Methicillin-Resistant Staphylococcus aureus (MRSA) Treatment: A Review," *Antibiotics.,* vol. 11, no. 4, 2022, Art. no. 606, doi: 10.3390/antibiotics11050606..

[4] G. Parfene, V. Horincar, A. K. Tyagi, A. Malik, and G. Bahrim, "Production of medium chain saturated fatty acids with enhanced antimicrobial activity from crude coconut fat by solid state cultivation of Yarrowia lipolytica," *Food Chem.*, vol. 136, no. 3–4, pp. 1345–1349, 2013, doi: 10.1016/j.foodchem.2012.09.057.

[5] M. Shilling *et al.*, "Antimicrobial effects of virgin coconut oil and its medium-chain fatty acids on clostridium difficile," *J. Med. Food*, vol. 16, no. 12, pp. 1079–1085, 2013, doi: 10.1089/jmf.2012.0303.

[6] Z. Fangfang, B. Xinpeng, G. Wei, G. Wang, Z. Shi, and C. Jun, "Effects of virgin coconut oil on the physicochemical, morphological and antibacterial properties of potato starch-based biodegradable films," *Int. J. Food Sci. Technol.*, vol. 55, no. 1, pp. 192–200, 2020, doi: 10.1111/ijfs.14262.

[7] J. Salimon, B. M. Abdullah, and N. Salih, "Hydrolysis optimization and characterization study of preparing fatty acids from Jatropha curcas seed oil," *Chem. Cent. J.*, vol. 5, no. 1, pp. 1–9, 2011, doi: 10.1186/1752-153X-5-67.

[8] F. D. Gunstone, *The Chemistry of Oils and Fats: Sources, Composition, Properties and Uses*. Oxford, UK: Wiley-Blackwell, 2004.

[9] C. T. Hou and Y. Shimada, "Lipases," in *Encyclopedia of Microbiology,* M. Schaechter, Ed., 3rd ed. Oxford, UK: Academic Press, 2009, pp. 229–242, doi: 10.1016/B978-012373944-5.00153-X.

[10] K. E. Jaeger, S. Ransac, B. W. Dijkstra, C. Colson, M. van Heuvel, and O. Misset, "Bacterial lipases," *FEMS Microbiol. Rev.*, vol. 15, no. 1, pp. 29–63, 1994, doi: 10.1016/0168-6445(94)90025-6.

[11] L. Nini, L. Sarda, L. C. Comeau, E. Boitard, J. P. Dubès, and H. Chahinian, "Lipase-catalysed hydrolysis of short-chain substrates in solution and in emulsion: A kinetic study," *Biochim. Biophys. Acta - Mol. Cell Biol. Lipids*, vol. 1534, no. 1, pp. 34–44, 2001, doi: 10.1016/S1388-1981(01)00172-X.

[12] M. Papagianni, "Advances in citric acid fermentation by Aspergillus niger: Biochemical aspects, membrane transport and modeling," *Biotechnol. Adv.*, vol. 25, no. 3, pp. 244–263, 2007, doi: 10.1016/j.biotechadv.2007.01.002.

[13] F. O. Nitbani, Jumina, D. Siswanta, and E. N. Solikhah, "Isolation and Antibacterial Activity Test of Lauric Acid from Crude Coconut Oil (Cocos nucifera L.)," *Procedia Chem.*, vol. 18, pp. 132–140, 2016, doi: 10.1016/j.proche.2016.01.021.

[14] M. C. P. Zenevicz, A. Jacques, A. F. Furigo, J. V. Oliveira, and D. de Oliveira, "Enzymatic hydrolysis of soybean and waste cooking oils under ultrasound system," *Ind. Crops Prod.*, vol. 80, pp. 235–241, 2016, doi: 10.1016/j.indcrop.2015.11.031.

[15] M. Elshikh *et al.*, "Resazurin-based 96-well plate microdilution method for the determination of minimum inhibitory concentration of biosurfactants," *Biotechnol. Lett.*, vol. 38, no. 6, pp. 1015–1019, 2016, doi: 10.1007/s10529-016-2079-2.

[16] T. A. V. Nguyen, T. D. Le, H. N. Phan, and L. B. Tran, "Hydrolysis Activity of Virgin Coconut Oil Using Lipase from Different Sources," *Scientifica* (Cairo), vol. 2018, Jan. 14, 2018, Art. no. 9120942, doi: 10.1155/2018/9120942

[17] H. Alexander *et al*., "The role of bacterial skin infections in atopic dermatitis: expert statement and review from the International Eczema Council Skin Infection Group," *Br. J. Dermato*l., vol. 182, no. 6, pp. 1331-1342, 2020, doi: 10.1111/bjd.18643.