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Research Article

Utilization of Kombucha SCOBY Waste Modified by Fe_3O_4/ZIF-8 for Antibacterial Wound Application

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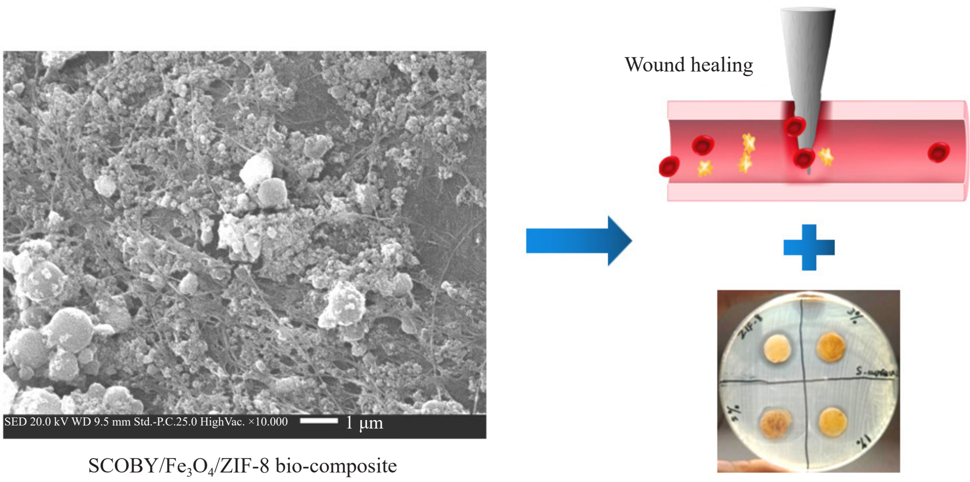
Graphical Abstract:



SCOBY/Fe_3O_4/ZIF-8 bio-composite

Antibacterial activity

Abstract: Wound dressing is an essential medical device used to cover wounds on the skin, but the lack of wound protection from bacterial infection has become a major issue nowadays. This research aims to develop a wound application from modified symbiotic culture of bacteria and yeast (SCOBY) with Fe_3O_4/zeolitic imidazolate framework-8 (ZIF-8) that can effectively protect the wound from bacterial infection. Cellulose-based SCOBY waste

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was obtained through the fermentation process of kombucha tea production at 25 °C for four days. This research studied the effect of varying concentrations of Fe_3O_4 in Fe_3O_4/ZIF-8 synthesis (1%, 3%, 5% wt), and different immersion times of SCOBY membrane in Fe_3O_4/ZIF-8 colloidal solution (60 and 120 minutes). For Fe_3O_4/ZIF-8 synthesis, magnetite (Fe_3O_4) with different concentrations was immobilized onto the zeolitic imidazolate framework-8 (ZIF-8) matrix; then, the SCOBY was immersed in the Fe_3O_4/ZIF-8 colloidal solution to form the SCOBY/Fe_3O_4/ZIF-8 biocomposite. The biocomposite characteristics were confirmed by scanning electron microscopy (SEM), fourier-transform infrared spectroscopy (FTIR), and X-ray diffraction (XRD). Moreover, the antibacterial activities were observed against *Escherichia coli* and *Staphylococcus aureus* bacteria using the disc diffusion method. Based on the results, the SCOBY/Fe_3O_4/ZIF-8 biocomposite with Fe_3O_4 concentration of 3% wt, and immersion time of 120 minutes showed the largest inhibition zone (21 ± 1 mm) against *E. coli*. It was found that the inhibition zone performed on *E. coli* bacteria was larger, indicating that *E. coli* is more resistant than *S. aureus*. This study demonstrated that the novel SCOBY/Fe_3O_4/ZIF-8 could be an excellent material for preparing antibacterial wound dressings and therefore warrants further investigation.

***Keywords*:** kombucha, zeolitic imidazolate framework-8, Fe_3O_4 nanoparticle, antibacterial activity, wound dressing

1. Introduction

Kombucha tea is a health drink from northeastern China produced through the fermentation process in a medium consisting of tea extract and symbiotic culture of bacteria and yeast (SCOBY) for 7-10 days.[1] In 2016, sales of kombucha tea reached $600 million and the growth is expected to continue up to 25% each year.[2] The bacteria contained in the kombucha tea belong to the genus *Acetobacter* and *Gluconobacter*, while the yeast belongs to genus *Saccharomyces*. The yeast ferments the sugar in kombucha tea into alcohol, which is then oxidized by bacteria to form acetic acid, giving the health drink its acidic taste.[3] However, the kombucha tea production process releases SCOBY membrane that is no longer used and becomes waste. Interestingly, SCOBY membrane has similar characteristics to bacterial cellulose (BC) membrane, which is known as a natural biopolymer with high mechanical strength, good water absorption, and outstanding biological compatibility.[4,5] Therefore, SCOBY membranes are becoming an alternative source of cellulose that can be used in many fields such as pharmaceutical and industrial sectors. Cellulose from SCOBY waste is becoming a sustainable multifunctional material and an environmentally responsible choice.[6] SCOBY membranes have some advantages, such as providing protection against ultraviolet rays and maintaining moisture,[7] having a strong and flexible fibers structure which provide mechanical support and rigidity to the membrane. These fiber properties give strength and stability to the membrane, which allows it to serve as a “home” for the bacterial and yeast colonies that contribute to the fermentation process. Although SCOBY membrane has different features due to the interaction of microorganisms involved in the Kombucha tea production, its characteristic similarity to cellulose membrane is crucial for its influence on the structure and consistency of the final product.[8] The membrane has been demonstrated as non-toxic, biocompatible, naturally degradable, and non-allergic material. Moreover, SCOBY membrane can be sterilized without affecting its characteristics.[9]

Metal organic frameworks (MOFs) are a group of coordination polymers composed of ion/metal nodes and functional organic ligands connected through coordination bonds. MOFs have received significant attention in biomedical research. Zeolitic imidazolate framework-8 (ZIF-8) is the most researched type of ZIF due to its ease of synthesis and production. Typical structure of a zeolitic imidazolate framework is formed through three-dimensional assembly of metal(imidazolate) tetrahedra. In general, ZIF-8 consists of Zn^{2+} ions and the ligand 2-methylimidazolate (2-meIm) with regular pore and channel shapes that allow the movement of certain molecules.[10,11] The material has been applied in various biomedical applications, including tissue engineering, drug delivery, and antimicrobial therapy.[12,13] MOFs have been widely applied for the immobilization of metal nanoparticles due to properties such as high porosity and high surface area.

Currently, metal oxide nanoparticles are developing very rapidly in biomedical applications as antibacterial wound healing agents.[14-17] Among the various types of nanomaterials, the ones that attract a lot of attention are nanometals because they have excellent optical and catalytic properties.[18-20] Magnetite nanoparticles (Fe_3O_4) are potential antibacterial wound healing agents that cause oxidative stress, which can disrupt the function of bacteria cells, making it difficult for bacteria to develop resistance to proteins within bacteria through the formation of free radicals, known as radical oxygen species (ROS).[14,21] ZIF-8 can be applied as promising antibacterial matrices for metal nanoparticle due to its large pore size and ability to release metal ions, thus enhancing antibacterial activity.[22,23]

This study focused on the development of antibacterial properties in SCOBY-based wound healing material by immobilizing Fe_3O_4 metal nanoparticle in ZIF-8 matrix to enhance the antibacterial release effect. Although wound dressings are essential for covering skin wounds, a significant current concern is their potential limitations in fully protecting against bacterial infection, as bacteria can still access and contaminate the wound site. So far, SCOBY/Fe_3O_4/ZIF-8 biocomposite has not been reported yet. The objective of this research was to investigate SCOBY/Fe_3O_4/ZIF-8 biocomposite composed of SCOBY cellulose material and Fe_3O_4/ZIF-8 as antibacterial potential agent that has significant uses in biomedical field. Moreover, antibacterial activities were tested against *Escherichia coli* and *Staphylococcus aureus* using the disc diffusion method.

2. Materials and methods

2.1 *Materials*

The materials used in this research included 2-methyl imidazole ($C_4H_6N_2$) obtained from Sigma-Aldrich, zinc nitrate hexahydrate ($Zn(NO_3)_2\cdot 6H_2O$) obtained from Sigma-Aldrich, iron (II) chloride tetrahydrate ($FeCl_2\cdot 4H_2O$) obtained from EMSURE, iron (III) chloride hexahydrate ($FeCl_3\cdot 6H_2O$) obtained from EMSURE, ammonium hydroxide (NH_4OH), sodium hydroxide (NaOH) obtained from EMSURE, SCOBY kombucha obtained from the "KombuchaForever" store, absolute ethanol (C_2H_5OH, 99.9%) obtained from EMSURE, methanol (CH_3OH, 99.9%) obtained from EMSURE, nutrient agar obtained from Sigma-Aldrich, black tea obtained from the "Heizl Official Store", sugar obtained from the "Papaya Official Store", Ampicillin, *Escherichia coli* ATCC 25922 and *Staphylococcus aureus* ATCC 25923, which were obtained from the bioprocess laboratory of Widya Mandala Surabaya Catholic University.

2.2 *Synthesis of ZIF-8*

$Zn(NO_3)_2\cdot 6H_2O$ (0.01 mol) and 2-methyl imidazole (0.08 mol) were each dissolved in 150 mL of methanol and stirred with a magnetic stirrer. Subsequently, the 2-methyl imidazole solution was added slowly to $Zn(NO_3)_2\cdot 6H_2O$ solution and stirred for 2 hours at room temperature. The white precipitate was collected by centrifugation, then washed twice with methanol and dried at 60 °C for ten hours.

2.3 *Synthesis of Fe_3O_4 nanoparticle*

$FeCl_3\cdot 6H_2O$ (0.004 mol) and $FeCl_2\cdot 4H_2O$ (0.002 mol) with a mole ratio of 2 : 1 were dissolved in 100 mL of distilled water separately, then the both colloidal solutions were mixed in a glass beaker and heated while stirring for ten minutes. After that, a 10% NH_4OH solution was added into the colloidal solution and stirred at 300, 400 and 500 rpm until pH 10. Then the solution was heated gradually for 30 minutes at temperatures of 40 °C, 60 °C and 80 °C, with the temperature increasing every ten minutes. Finally, the nanoparticle precipitate was separated by centrifugation, washed using distilled water and dried in an oven at 100 °C for 150 minutes.

2.4 *SCOBY membrane culture*

First, black tea (5 g) and sugar (100 g) were added to 500 mL of boiling water and stirred for 5 minutes. Subsequently, 500 mL of drinking water was added to the tea solution to speed up the cooling process, and the SCOBY starter was mixed into the tea solution. After thorough mixing, the culture solution was filtered and put 10 mL into a test tube for the fermentation process at room temperature for 4 days. Cellulose-based SCOBY membrane was formed with a diameter of 1.3 cm on the test tube surface. The SCOBY membrane was harvested, treated with 0.1 M NaOH solution for at 80 °C for 1 hour and washed with distilled water to pH 7.

2.5 *Synthesis of SCOBY/Fe_3O_4/ZIF-8*

Each ZIF-8 (0.5 g) and Fe_3O_4 NPs were homogeneously dispersed in 20 mL of ethanol using a sonicator. Then, the Fe_3O_4 NPs colloidal solution with varying weight percentages of 1, 3 and 5% each was dripped slowly onto the ZIF-8 solution. After 1 hour of stirring, the temperature increased to 75 °C and slowly added NaOH dissolved in 10 mL of ethanol until brown color appeared. Then the mixture was washed using ethanol until pH 7 and dried in an oven at 80 °C for 2 hours. After drying, Fe_3O_4/ZIF-8 was dissolved in distilled water with varying concentrations. Subsequently, the dried SCOBY biomembrane was immersed in the Fe_3O_4/ZIF-8 colloidal solution with varying immersion times of 60 and 120 minutes to form SCOBY/Fe_3O_4/ZIF-8 biocomposite.

2.6 *Characterizations*

The biocomposites were characterized using field emission scanning electron microscopy (FE-SEM) (JSM-6500F, JEOL, Japan) to determine the surface morphology of the sample. Fourier-transform infrared spectroscopy (FTIR) (FTS 3500, Bio-Rad Laboratories Sadtler Division, USA) was used to identify the special groups in the chemical compounds contained in a component. Furthermore, the crystal structure of biocomposite was analyzed by X-ray diffraction (XRD).

2.7 *Antibacterial test*

Antibacterial test was performed using the disc diffusion method against gram-negative bacteria *E. coli* and gram-positive bacteria *S. aureus*. The SCOBY membrane with a diameter of 1.3 cm was immersed in the Fe_3O_4/ZIF-8 colloidal solution for 60 and 120 minutes. The bacteria samples were spread on the surface of the agar using a sterile cotton swab. Biocomposite samples (SCOBY/Fe_3O_4/ZIF-8) and control (ZIF-8, Fe_3O_4, SCOBY) with a concentration of 25 mM were placed on the agar surface. Subsequently, all the samples were incubated for 24 hours at 37 °C. After that, the samples were observed by measuring the diameter of the inhibition zone. In addition, ampicillin antibiotic with a concentration of 25 mM was used as positive control.

3. Results and discussion

3.1 *Synthesis and characterization of SCOBY/Fe_3O_4/ZIF-8*

By reacting zinc nitrate hexahydrate (0.01 mol) and 2-methylimidazole (0.08 mol) using methanol as a solvent, a product was formed, called Zeolitic Imidazolate Frameworks-8 or commonly abbreviated as ZIF-8. In the synthesis of ZIF-8, methanol solvent was used because using methanol as a solvent could provide good product purity and uniform particle shape.[24] Figure 1a shows the morphology of ZIF-8 that has been synthesized in this research. The FE-SEM results of ZIF-8 synthesized in this research showed that the resulting product has a uniform shape and corresponds to the the rhombic dodecahedron shape reported in the literature.[25] The SCOBY/Fe_3O_4/ZIF-8 biocomposite was made by mixing Fe_3O_4 and ZIF-8 with NaOH and ethanol solvent. Then the SCOBY membrane was soaked in the Fe_3O_4/ZIF-8 colloidal solution and subsequently the particles of Fe_3O_4 attached to particles of ZIF-8 (Figure 1b) which occur due to the mixing of Fe_3O_4 and ZIF-8. From the synthesis results of the Fe_3O_4/ZIF-8 biocomposite, it was found that the yields for variations of Fe_3O_4 at 1%, 3%, and 5% wt were 72.8%, 77.8% and 84.4%, respectively. Figure 1c shows the SCOBY nanofibers which have been immersed in the Fe_3O_4/ZIF-8 to form SCOBY/Fe_3O_4/ZIF-8 biocomposite. The results of the SEM characterization showed that the Fe_3O_4/ZIF-8 biocomposite was successfully absorbed by the SCOBY matrix, which proves that the SCOBY has properties that enable it absorb liquids.[26]

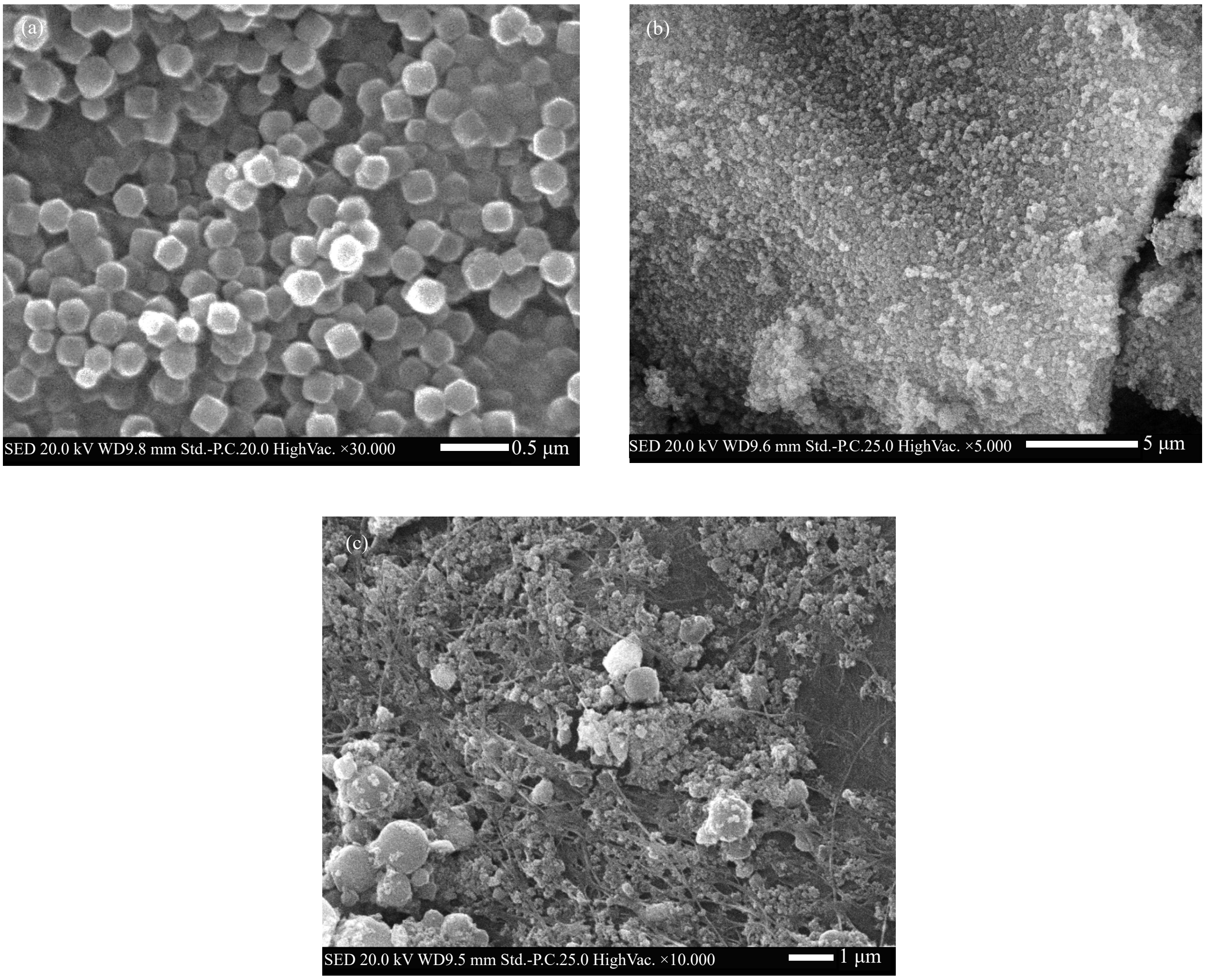

Figure 1. SEM analysis of (a) ZIF-8, (b) Fe_3O_4/ZIF-8, (c) SCOBY/Fe_3O_4/ZIF-8 biocomposite

The presence of functional groups in SCOBY/Fe_3O_4/ZIF-8 biocomposite was confirmed by FTIR analysis as shown in Figure 2. The OH group at wavenumber 3,345 cm^{-1} (3,433.98-3,362.84 cm^{-1}) and the C-O group at wavenumber 1,309 cm^{-1} (1,382-1,036 cm^{-1}) are characteristic of cellulose material from the SCOBY membrane. While the Fe-O group at wavenumber 598 cm^{-1} (544-673 cm^{-1}) indicates the presence of Fe_3O_4.[27] In addition, we have confirmed that Fe_3O_4 is a ferrimagnetic material that attracts magnets due to its superior magnetic properties[28] as shown in Figure S2 (in Appendix). The band at 408 cm^{-1} is associated with Zn-N stretching and the band at 759 cm^{-1} (675-995 cm^{-1}) represents the C-H group which shows ZIF-8 characteristics.[26,29] Therefore, these functional groups successfully confirmed the presence of cellulose, Fe_3O_4 and ZIF-8 in the SCOBY/Fe_3O_4/ZIF-8 biocomposite.

XRD analysis was used to determine the structure of the salts and their biocomposite membrane samples. The phase purity and crystallinity of ZIF-8, Fe_3O_4/ZIF-8 & SCOBY/Fe_3O_4/ZIF-8 are shown in Figure 3. The sharp peaks at 7.309° (011), 10.331° (002), 12.665° (112), 14.617° (022), 16.372° (013) and 17.957° (222) are characteristic of ZIF-8.[30] Then, the presence of ZIF-8 in Fe_3O_4/ZIF-8 biocomposite was confirmed by several peaks such as 29.759° (220), 35.644° (311), 57.479° (511) and 62.868° (440).[31] Furthermore, the structure of cellulose-based SCOBY was successfully proven by sharp peaks at 14.552° (101) and 22.8189° (002).[32]

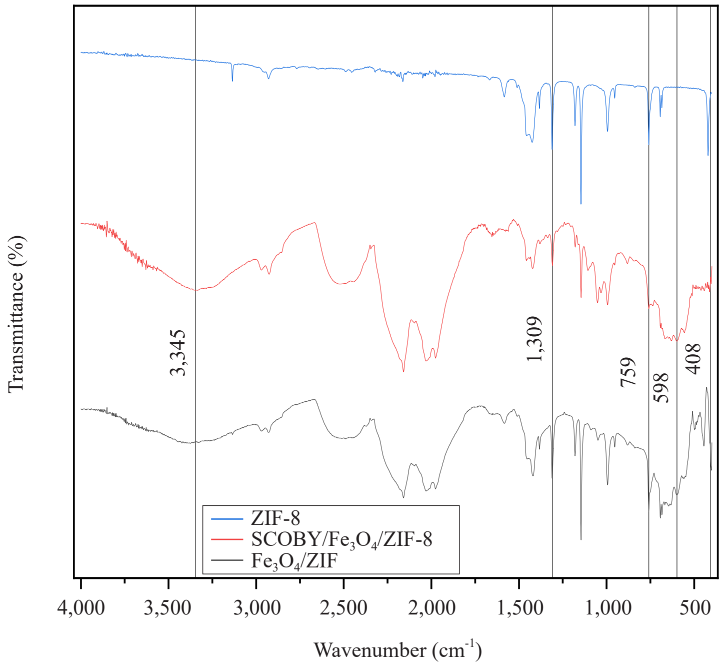

Figure 2. FTIR spectrum of ZIF-8, Fe_3O_4/ZIF-8 and SCOBY/Fe_3O_4/ZIF-8 biocomposite

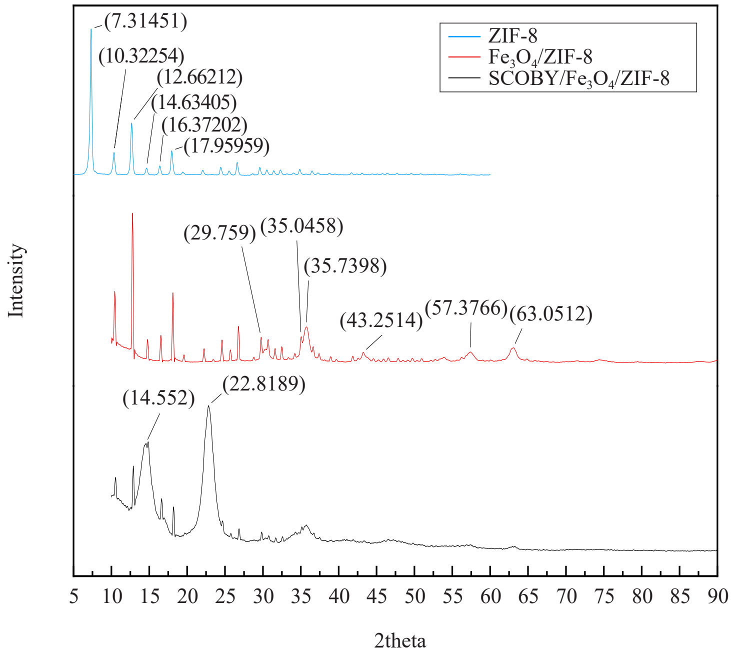

Figure 3. XRD results of ZIF-8, Fe_3O_4/ZIF-8 and SCOBY/Fe_3O_4/ZIF-8

3.2 *Antibacterial activity against Escherichia coli & Staphylococcus aureus*

Antibacterial activity test against gram-negative bacteria *E. coli* and gram-positive bacteria *S. aureus* was performed using the disc diffusion method. In the antibacterial activity test, the effect of varying concentrations of Fe_3O_4 nanoparticles in the Fe_3O_4/ZIF-8 synthesis and the SCOBY membrane immersion for 60 and 120 minutes in the

Fe_3O_4/ZIF-8 colloidal solution were studied. In addition, a SCOBY membrane immersed in a ZIF-8 solution was also performed as a control.

Figures 4 and 5 show the antibacterial test where a SCOBY immersion time of 60 minutes in Fe_3O_4/ZIF-8 against *E. coli* and *S. aureus* did not provide a significant effect (15-17 mm) due to the short immersion time. In Figures 6 and 7, the antibacterial effect increased when the SCOBY samples were soaked in Fe_3O_4/ZIF-8 for 120 minutes compared to 60 minutes due to the large absorption of Fe_3O_4/ZIF-8 in the SCOBY membrane.

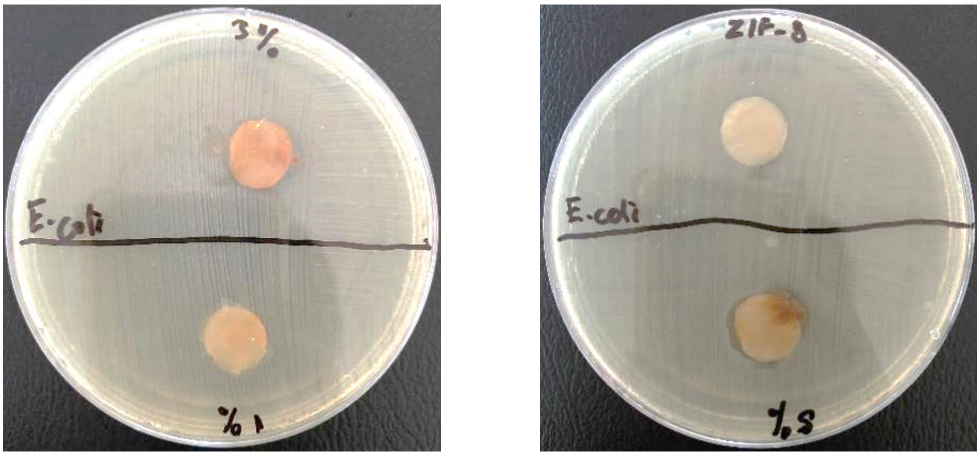

Figure 4. SCOBY immersion for 60 minutes in Fe_3O_4/ZIF-8 solution with Fe_3O_4 concentration of 1, 3, 5% wt and ZIF-8 as a control against *E. coli*

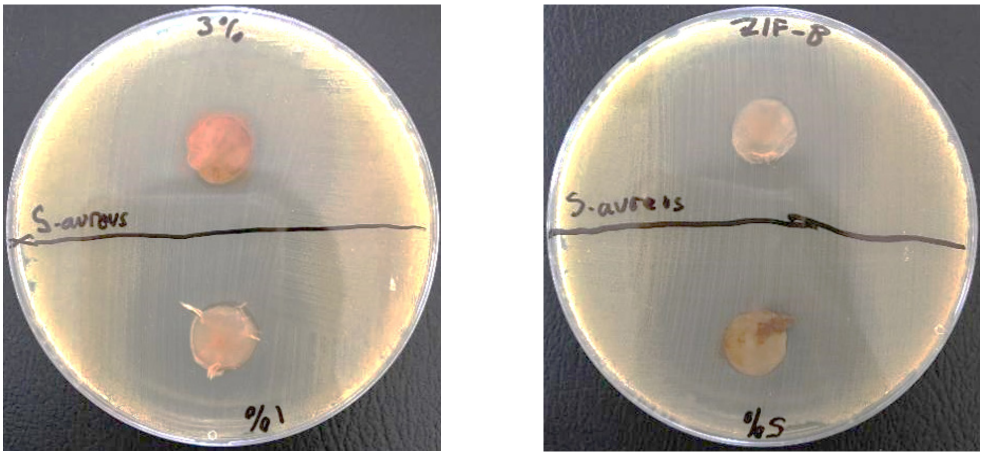

Figure 5. SCOBY immersion for 60 minutes in Fe_3O_4/ZIF-8 solution with Fe_3O_4 concentration of 1, 3, 5% wt and ZIF-8 as a control against *S. aureus*

From the results obtained (Figure 6 and Figure 7), the Fe_3O_4/ZIF-8 biocomposite with an Fe_3O_4 concentration of 3% wt has the highest inhibition zone compared to 1 and 5% wt. At a concentration of 3% wt, both components Fe_3O_4 and ZIF-8 could react well so that the antibacterial test results are higher than concentrations of 1% and 5% wt. When the concentration of Fe_3O_4 is high enough to exceed that of ZIF-8, the interaction will become saturated. Fe_3O_4 nanoparticles show a smaller antibacterial effect, while ZIF-8 shows a fairly good antibacterial effect, while the biocomposite which is a combination of ZIF-8 and Fe_3O_4 nanoparticles shows an antibacterial effect that tends to be more than the effect of ZIF-8 itself. The combination of ZIF-8 and Fe_3O_4 nanoparticles looks more effective when combined in biocomposite form. It was found that the zone of inhibition carried out on *E. coli* bacteria was larger, which shows that *S. aureus* is a bacterium that is more susceptible to antibacterials. The outer membrane of *E. coli* on its Gram-negative cell wall acts as a barrier, preventing many antibacterial materials from reaching their targets inside the cell.[33] Furthermore, we also measured the common antibiotic as positive control where ampicillin with a concentration of 3% wt exhibited 52 mm ± 1 mm against *E. coli* (Figure S1 in Appendix).

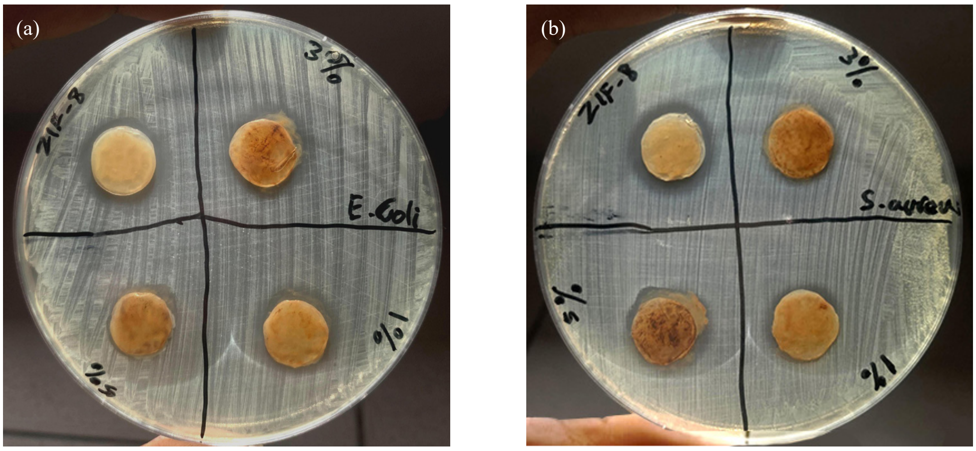

Figure 6. SCOBY immersion for 120 minutes in Fe_3O_4/ZIF-8 solution with Fe_3O_4 concentration of 1, 3, 5% wt and ZIF-8 against (a) *E. coli* and (b) *S. aureus*

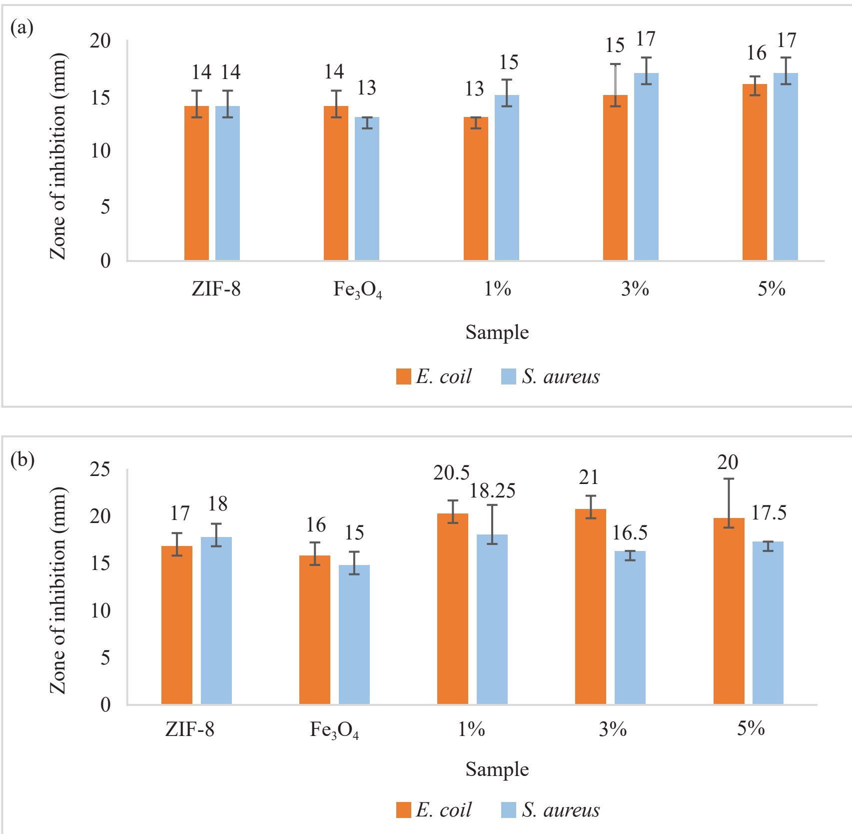

Figure 7. Inhibition zone against *E. coli* and *S. aureus*. (a) 60 minutes SCOBY immersion and (b) 120 minutes SCOBY immersion in Fe_3O_4/ZIF-8 solution, ZIF-8, and Fe_3O_4

Fe_3O_4 nanoparticles themselves can generate reactive oxygen species (ROS), which can also have negative effects on humans.[34] Excessive use of ROS effects can cause oxidative stress, damaging important cellular structures such as proteins, lipids and nucleic acids, and potentially leading to the onset of several diseases such as diabetes and metabolic disorders.[35] Therefore,combining Fe_3O_4 and ZIF-8 nanoparticles can overcome the shortcomings of Fe_3O_4.

Magnetic metal or oxide based nanoparticles in the biopolymer matrix biocomposite system are an active area of investigation.[36,37] With Fe_3O_4 nanoparticles, combining these two components can reduce the effect of excessive ROS on Fe_3O_4 nanoparticles. ZIF-8 is a porous nanomaterial with a high surface area that can absorb and immobilize Fe_3O_4 nanoparticles to form a core-shell structure. The shell of ZIF-8 acts as a barrier, preventing direct interaction of Fe_3O_4 nanoparticles with oxygen and other reactive species, thereby reducing excessive ROS formation.[38]

4. Conclusions

Symbiotic culture of bacteria and yeast (SCOBY) waste obtained from Kombucha tea fermentation has exhibited promising antibacterial and wound healing capabilities, which can be achieved by modifying SCOBY with Fe_3O_4/ZIF-8 as an antibacterial agent. From the results obtained, the biocomposite with a variation of 3% wt of Fe_3O_4 concentration and immersion time of SCOBY in Fe_3O_4/ZIF-8 for 120 minutes showed the highest inhibition zone (21 ± 1 mm) against *E. coli*. It was found that the zone of inhibition carried out on *E. coli* bacteria was larger, which shows that *S. aureus* is a bacterium that is more susceptible to antibacterials. In summary, SCOBY/Fe_3O_4/ZIF-8 is a promising, non-toxic biocomposite with potential for antibacterial wound healing applications. However, the biocomposite still needs further investigation regarding biocompatibility testing and antibacterial properties improvement. Furthermore, the most significant challenge in this research is the effectiveness of antibacterial wound materials in the healing process.

Conflict of interest

The authors declare no conflict of interest.

References

[1] Laavanya, D.; Shirkole, S.; Balasubramanian, P. Current challenges, applications and future perspectives of SCOBY cellulose of kombucha fermentation. *J. Clean Prod.* **2021,** *295*, 126454.

[2] St-Pierre, D. L. Microbial Diversity of the Symbiotic Colony of Bacteria and Yeast (SCOBY) and Its Impact on the Organoleptic Properties of Kombucha. Master's Thesis, University of Maine Repository, 2019.

[3] Leal, J. M.; Suárez, L. V.; Jayabalan, R.; Oros, J. H.; Escalante-Aburto, A. A review on health benefits of kombucha nutritional compounds and metabolites. *CyTA-J. Food* **2018,** *16*(1), 390-399.

[4] Zhang, Z. Y.; Sun, Y.; Zheng, Y. D.; He, W.; Yang, Y. Y.; Xie, Y. J.; Feng, Z. X.; Qiao, K. A biocompatible bacterial cellulose/tannic acid composite with antibacterial and anti-biofilm activities for biomedical applications. *Mater. Sci. Eng. C* **2020,** *106*, 110249.

[5] Li, Z.; Hu, W.; Dong, J.; Azi, F.; Xu, X.; Tu, C.; Tang, S.; Dong, M. The use of bacterial cellulose from kombucha to produce curcumin-loaded Pickering emulsion with improved stability and antioxidant properties. *Food Sci. Hum. Wellness* **2023,** *12*(2), 669-679.

[6] Besty Asthary, P.; Saepulloh; Sanningtyas, A.; Aditya Pertiwi, G.; Apriana Purwita, C.; Septiningrum, K. Optimasi produksi bacterial nanocellulose dengan metode kultur agitasi [Optimization of bacterial nanocellulose production in agitation culture methods]. *J. Selulosa* **2020,** *10*(2), 89-100.

[7] Rajwade, J. M.; Paknikar, K. M.; Kumbhar, J. V. Applications of bacterial cellulose and its composites in biomedicine. *Appl. Microbiol. Biotechnol*. **2015,** *99*(6), 2491-2511.

[8] Knöller, A.; Widenmeyer, M.; Bill, J.; Burghard, Z. Fast-growing bacterial cellulose with outstanding mechanical properties via cross-linking by multivalent ions. *Materials* **2020,** *13*(12), 1-8.

[9] Abdelraof, M.; Hasanin, M. S.; Farag, M. M.; Ahmed, H. Y. Green synthesis of bacterial cellulose/bioactive glass nanocomposites: Effect of glass nanoparticles on cellulose yield, biocompatibility and antimicrobial activity. *Int. J*

Biol. Macromol. **2019,** *138*, 975-985.
[10] Mphuthi, L. E.; Erasmus, E.; Langner, E. H. G. Metal exchange of ZIF-8 and ZIF-67 nanoparticles with Fe(II) for enhanced photocatalytic performance. *ACS Omega* **2021,** *6*(47), 31632-31645.
[11] Taheri, M.; Ashok, D.; Sen, T.; Enge, T. G.; Verma, N. K.; Tricoli, A.; Lowe, A.; Nisbet, D. R.; Tsuzuki, T. Stability of ZIF-8 nanopowders in bacterial culture media and its implication for antibacterial properties. *Chem. Eng. J.* **2021,** *413*, 127511.
[12] Wu, M. X.; Yang, Y. W. Metal-organic framework (MOF)-based drug/cargo delivery and cancer therapy. *Adv. Mater*. **2017,** *29*(23), 1606134.
[13] Zhang, X.; Chen, J.; Pei, X.; Wang, J.; Wan, Q.; Jiang, S.; Huang, C.; Pei, X. Enhanced osseointegration of porous titanium modified with zeolitic imidazolate framework-8. *ACS Appl. Mater. Interfaces* **2017,** *9*(30), S1-S9.
[14] Liu, C.; Zhao, X.; Wang, Z.; Zhao, Y.; Li, R.; Chen, X.; Chen, H.; Wan, M.; Wang, X. Metal-organic framework-modulated Fe_3O_4 composite Au nanoparticles for antibacterial wound healing via synergistic peroxidase-like nanozymatic catalysis. *J. Nanobiotechnology* **2023,** *21*, 427.
[15] Geleto, S. A.; Ariti, A. M.; Gutema, B. T.; Abda, E. M.; Abiye, A. A.; Abay, S. M.; Mekonnen, M. L.; Workie, Y. A. Nanocellulose/Fe_3O_4/Ag nanozyme with robust peroxidase activity for enhanced antibacterial and wound healing applications. *ACS Omega* **2023,** *8*(51), 48764-48774.
[16] Nagar, S.; Mukherjee, G. S.; Banerjee, M. Studies on the structural and magnetic properties of PMMA/Ni nanocomposite system prepared by embedding Ni nanoparticles in the PMMA films. *Mater. Chem. Phys*. **2024,** *314*, 128913.
[17] Nagar, S.; Mukherjee, G. S.; Banerjee, M. Structural and magnetic properties of Ni nanoparticles embedded in vinyl polymer nanocomposite films. *SPIN* **2023,** *13*(2), 2340011.
[18] Cai, N.; Li, C.; Han, C.; Luo, X.; Shen, L.; Xue, Y.; Yu, F. Tailoring mechanical and antibacterial properties of chitosan/gelatin nanofiber membranes with Fe_3O_4 nanoparticles for potential wound dressing application. *Appl. Surf. Sci*. **2016,** *369*, 492-500.
[19] Saraswaty, V.; Aji, E. S.; Ardiansyah, A.; Hanifah, A.; Puspitasari, N.; Santoso, S. P.; Hartono, S. B.; Risdian, C.; Chaldun, E. R.; Setiyanto, H. One-pot synthesis of green zinc oxide nanoparticles immobilized on activated carbon derived from pineapple peel for adsorption of Pb(II). *Karbala Int. J. Mod. Sci*. **2024,** *10*(2), 153-165.
[20] Puspitasari, N.; Retnoningtyas, E. S.; Gunarto, C.; Soetaredjo, F. E. The intra- and extracellular mechanisms of microbially synthesized nanomaterials and their purification. In *Green and Sustainable Approaches Using Wastes for the Production of Multifunctional Nanomaterials*; Elsevier, 2024; pp 273-288.
[21] Yao, H.; Liu, J.; Jiang, X.; Chen, F.; Lu, X.; Zhang, J. Analysis of the clinical effect of combined drug susceptibility to guide medication for carbapenem-resistant Klebsiella pneumoniae patients based on the Kirby-Bauer disk diffusion method. *Infect. Drug Resist*. **2021,** *14*, 79-87.
[22] Duan, C.; Meng, J.; Wang, X.; Meng, X.; Sun, X.; Xu, Y.; Zhao, W.; Ni, Y. Synthesis of novel cellulose-based antibacterial composites of Ag nanoparticles@ metal-organic frameworks@ carboxymethylated fibers. *Carbohydr. Polym*. **2018,** *193*, 82-88.
[23] Lewis, A.; Hazelton, P.; Butt, F. S.; Mazlan, N. A.; Wei, X.; Radacsi, N.; Chen, X.; Yang, Y.; Yang, S.; Huang, Y. Growth of nanostructured antibacterial zeolitic imidazolate framework coatings on porous surfaces. *ACS Appl. Nano. Mater*. **2022,** *5*(11), 16250-16263.
[24] Tsai, C. W.; Langner, E. H. G. The effect of synthesis temperature on the particle size of nano-ZIF-8. *Micropor. Mesopor. Mat*. **2016,** *221*, 8-13.
[25] Linder-Patton, O. M.; De Prinse, T. J.; Furukawa, S.; Bell, S. G.; Sumida, K.; Doonan, C. J.; Sumby, C. J. Influence of nanoscale structuralisation on the catalytic performance of ZIF-8: A cautionary surface catalysis study. *Cryst. Eng. Comm*. **2018,** *20*(34), 4926-4934.
[26] Puspitasari, N.; Arief, D.; Ismadji, S.; Saraswaty, V.; Santoso, S. P.; Retnoningtyas, E. S.; Putro, J. N.; Gunarto, C. Synthesis of novel bacterial cellulose-based silver-metal organic frameworks (BC@Ag-MOF) for antibacterial wound healing. *Fine Chem. Eng.* **2023,** *4*, 193-202.
[27] Winiarczyk, K.; Gac, W.; Góral-Kowalczyk, M.; Surowiec, Z. Magnetic properties of iron oxide nanoparticles with a DMSA-modified surface. *Hyperfine Interact*. **2021,** *242*, 48.
[28] Nguyen, M. D.; Tran, H.-V.; Xu, S.; Lee, T. R. Fe_3O_4 nanoparticles: Structures, synthesis, magnetic properties, surface functionalization, and emerging applications. *Appl. Sci.* **2021,** *11*(23), 11301.
[29] Prasad, A. G. D.; Zarei, M. FTIR spectroscopic studies on Cleome gynandra: Comparative analysis of functional groups before and after extraction. *Romanian J. Biophys*. **2012,** *22*, 137-143.
[30] Awadallah-F, A.; Hillman, F.; Al-Muhtaseb, S. A.; Jeong, H. K. On the nanogate-opening pressures of copper-

doped zeolitic imidazolate framework ZIF-8 for the adsorption of propane, propylene, isobutane, and *n*-butane. *J. Mater. Sci*. **2019,** *54*(7), 5513-5527.
[31] Kong, J.; Xu, S.; Dai, Y.; Wang, Y.; Zhao, Y.; Zhang, P. Study of the Fe_3O_4@ZIF-8@Sor composite modified by tannic acid for the treatment of sorafenib-resistant hepatocellular carcinoma. *ACS Omega* **2023,** *8*(42), 39174-39185.
[32] Wasim, M.; Khan, M. R.; Mushtaq, M.; Naeem, A.; Han, M.; Wei, Q. Surface modification of bacterial cellulose by copper and zinc oxide sputter coating for UV-resistance/antistatic/antibacterial characteristics. *Coatings* **2020,** *10*(4), 364.
[33] Haindongo, E. H.; Ndakolo, D.; Hedimbi, M.; Vainio, O.; Hakanen, A.; Vuopio, J. Antimicrobial resistance prevalence of *Escherichia coli* and *Staphylococcus aureus* amongst bacteremic patients in Africa: A systematic review. *J. Glob. Antimicrob. Resist*. **2023,** *32*, 35-43.
[34] Alili, L.; Chapiro, S.; Marten, G. U.; Schmidt, A. M.; Zanger, K.; Brenneisen, P. Effect of Fe_3O_4 nanoparticles on skin tumor cells and dermal fibroblasts. *Biomed. Res. Int*. **2015,** *2015*, 1-11.
[35] Pizzino, G.; Irrera, N.; Cucinotta, M.; Pallio, G.; Mannino, F.; Arcoraci, V.; Squadrito, F.; Altavilla, D.; Bitto, A. Oxidative stress: Harms and benefits for human health. *Oxid. Med. Cell. Longev*. **2017,** *2017*, 8416763.
[36] Nagar, S.; Mukherjee, G. S.; Banerjee, M. Structural and magnetic properties of Ni nanoparticles embedded in vinyl polymer nanocomposite films. *SPIN* **2023,** *13*(2), 2340011.
[37] Nagar, S.; Mukherjee, G. S.; Banerjee, M. Studies on the structural and magnetic properties of PMMA/Ni nanocomposite system prepared by embedding Ni nanoparticles in the PMMA films. *Mater. Chem. Phys*. **2024,** *314*, 128913.
[38] Abdelmigeed, M. O.; Sadek, A. H.; Ahmed, T. S. Novel easily separable core-shell Fe_3O_4/PVP/ZIF-8 nanostructure adsorbent: Optimization of phosphorus removal from fosfomycin pharmaceutical wastewater. *RSC Adv*. **2022,** *12*(20), 12823-12842.

Appendix

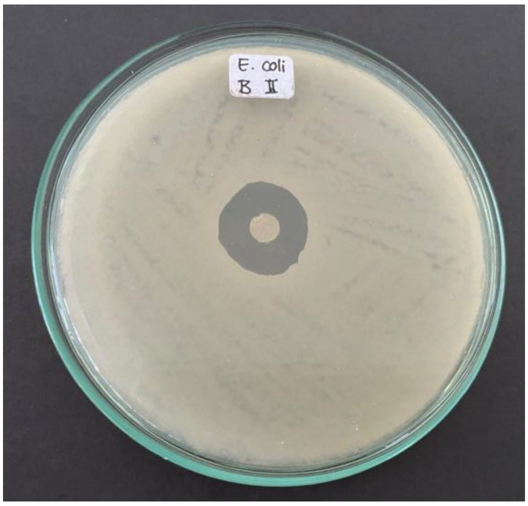

Figure S1. Antibacterial activity of Ampicillin (3 wt%) as positive control against *E. coli*

Figure S2. Fe_3O_4 samples (a) and ferrimagnetic properties of Fe_3O_4