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EFFECT OF ZINC ADDITION IN COPPER TO STRUCTURE, HARDNESS, CORROSION, AND ANTIBACTERIAL ACTIVITY

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ABSTRACT

Brass (CuZn) is widely used today due to better mechanical, thermal, physical, and chemical properties. The present research casts CuZn alloy by adding various Zn (6, 9, and 12 wt.%) to the Cu using gravity casting. Structure, hardness, corrosion, and antibacterial activity behavior were investigated using XRD, potentiostat, Vickers hardness equipment, and a digital camera. XRD confirmed that CuZn alloy has a cubic structure. The rise of the Zn content led to a decrease in the hardness and a shift to a more negative OCP potential at 1200 s measurement. The increase of the Zn content to 9 wt.% decreased the corrosion rate. Antibacterial activity observation found that all samples had no diffusion. Moreover, 24-hour post-contact observation found that the sample places removed remained clear of bacteria. The fluid contact test shows that the Cu6Zn sample has better antibacterial performance than others. The Cu6Zn sample successfully inhibited the growth of Escherichia coli in the 3rd hour, while Staphylococcus aureus was 100 % reduced in the 7th hour.

Keywords : XRD, Vickers, Electrochemical measurement, Staphylococcus aureus, Escherichia coli

1. Introduction

Brass (CuZn) is an alloy widely used in the defense, oil and gas industries, and health because it has better mechanical properties, thermal conductivity, and corrosion resistance (Basori et al., 2018) (Achiței et al., 2017). Widyastuti et al. state that CuZn alloy is commonly used for bullet casings (Widyastuti et al., 2023). Fadhil et al. used CuZn alloy for valves in the oil and gas industry (Fadhil et al., 2018). Tang et al. investigated CuZn alloys for cardiovascular applications (Tang et al., 2017).

CuZn alloys for medical equipment need to consider two parameters: corrosion resistance and antibacterial characteristics. Eccrine sweat and saline-infused for humans are near 0.9 % of NaCl. Minciuna et al. investigated CuZn casting alloy in a saline medium and found a corrosion current of around 25.325 $\mu\text{A}/\text{cm}^2$ (Minciuna et al., 2015). Gao et al. found that a reduction in thickness (50 to 60 %) of CuZn using cold rolling resulted in a significant decrease in corrosion current from 4.824 to 1.804 $\mu\text{A}/\text{cm}^2$ (investigating in 3.5 % NaCl) (Gao et al., 2021). Furthermore, some studies found that after cleaning the ambulance, 35.37 % of bacterial contaminants were still seen (Syamsuir et al., 2023). Vikke and Giebner found *Staphylococcus aureus* was detectable on ambulance equipment, though it was clean (Vikke & Giebner, 2016). *Staphylococcus aureus* is a type of bacteria that could cause skin disease and is hard to treat with traditional antibiotics. According to El-Mokhtar and Hetta, 46.1 % of *Staphylococcus aureus* is methicillin resistant (El-

Mokhtar & Hetta, 2018). Moreover, this bacteria also could contaminate medical implants and cause serious infections (Oliveira et al., 2018). Cu^{2+} and Zn^{2+} ions could act as antibacterial agents and inhibit *Staphylococcus aureus* growth (E. Zhang et al., 2021).

According to the literature review, Zn addition with composition 6, 9, and 12 wt.% into Cu has not been comprehensively investigated recently, especially for medical transportation. Therefore, the present research was casts CuZn alloy by adding various Zn to the Cu. Structure, hardness, corrosion, and antibacterial activity were investigated using XRD, Vickers hardness equipment, potentiostat, and digital camera.

2. Literature Review

Many techniques are used to make CuZn alloys, including gravity and investment casting (Hendrawan et al., 2021; Iecks et al., 2018). Gravity casting has the advantage of being easy, cheap, and quick to implement. Moreover, in the fabrication of CuZn alloys, one thing needs to be considered to produce specific properties, namely alloy composition. Researchers focused on adding various Zn compositions onto Cu for different purposes. Shahriyari et al. added Zn (5, 15, 20, and 30 wt.%), increasing the hardness due to the rise of the Zn content in the alloy (Shahriyari et al., 2022). Daniyan et al. found that hardness and tensile strength would increase with increasing Zn content (5, 10, 15, 20, and 30 %) in the alloy (Afolabi Daniyan et al., 2021). Situmorang et al. fabricated Cu with various Zn additions (10, 20, 38, and 45 wt.%) and found that the higher the Zn composition, the higher the antibacterial properties (Situmorang et al., 2019). Igelegbai et al. prepared Cu5Zn, Cu10Zn, Cu15Zn, Cu20Zn, and Cu30Zn using the sand casting method from scrap Cu and Zn resulting increase in hardness with an increased Zn content in the alloy (Igelegbai et al., 2017). Tang et al. melted Cu38Zn and Zn (Zn-xCu, x=1, 2, 3, and 4 wt.%) for cardiovascular implant applications. An increase in Cu concentration leads to an increase in tensile strength. The higher corrosion rate in the c-SBF solution is seen in the Zn-1Cu sample, around 33 $\mu\text{m}/\text{year}$; in contrast, lower corrosion rate are seen in pure Zn (around 22.1 $\mu\text{m}/\text{year}$) (Tang et al., 2017). Strzpek et al. investigated the mechanical properties of Cu and alpha brass (Cu2.5Zn and Cu6.5Zn) wire (\varnothing 3.8 mm). Increased Zn content causes increases in ultimate tensile strength, yield strength and hardness (Strzpek et al., 2019). Generally, a CuZn alloy with a Zn composition of less than 35 wt.% would produce a single alpha phase with an FCC crystal structure (Machuta et al., 2016).

3. Research Methods

3.1 Material Preparation

Sample preparation used the gravity casting method. Cu ingot (98.798 %) and Zn powder (99 %) were cast with the alloy composition Cu_xZn ($x=0, 6, 9$, and 12 wt.%, namely as Cu, Cu6Zn, Cu9Zn, and Cu12Zn, respectively). Melting was carried out in a crucible at 1100 °C under atmospheric pressure using a muffle furnace. The as-cast ingots, such as XRD, hardness, electrochemical measurement, and antibacterial activity observation, were cut for further characterization.

3.2 XRD Measurement

XRD was measured using the PANalytical (Cu $\text{K}\alpha_1$ $\lambda=1.5405980$) apparatus. XRD was scanned from 20 to 100°, using step size 0.0217°. The Highscore software was used to refine and collect the structure of the as-cast samples.

3.3 Hardness Measurement

Before testing, samples with 20×20×6 mm dimensions were polished using silicon carbide up to #3000 grit. The hardness test was conducted using the Vickers method. An FV-300e hardness test was performed on top of various samples using 1 kg of load. Ten repeatable measurements were conducted.

3.4. Electrochemical Measurement

Two electrochemical measurements were conducted in the present research, such as OCP and LSV, using Digi-Ivy (DY2311) potentiostat in 0.9 % NaCl at room temperature. OCP was scanned until 1200 s using a sampling scan rate of 0.02 s, while LSV was conducted using a scan rate of 1 mV/s. Cu/CuZn samples are used as the working electrode, platinum wire as the counter electrode, and Ag/AgCl as the reference electrode. LSV data was examined using the Tafel extrapolation method to see corrosion potential (E_{corr}) and current density (i_{corr}). The corrosion rate could be found by inserting i_{corr} in the following equation (Ahmad, 2006).

$$\text{Corrosion rate (mmpy)} = K \frac{M \times i_{corr}}{\rho \times n} \quad (1)$$

Where K is corrosion constant (3.27 mmpy), M is atomic weight (g/mol), ρ is material density (g/cm³), and n is the number of electrons involved.

3.5 Antibacterial Activity Observation

The sample dimension used for antibacterial activity is 20×20×6 mm. Before testing, samples with 20×20×6 mm dimensions were polished using silicon carbide up to #3000 grit. The experimental procedure for antibacterial activity is similar to that of the previous report (Syamsuir et al., 2023). Moreover, the recent study uses *Staphylococcus aureus* ATCC 25923 for Direct contact kill assay (24 hours) and Fluid contact assay (8 hours). In comparison, *Escherichia coli* 25922 was also used in the present study. Afterward, documentation was captured using a digital camera. In addition, 24-hour post-contact observation and Fluid contact test were also captured using a digital camera.

4. Results and Discussions

4.1 XRD

The diffraction pattern of Cu/CuZn with (111), (200), (311) and (222) planes can be seen in Figure 1. The four diffraction patterns match with the alpha phase, which is shown in alignment with another study (Heidarzadeh et al., 2022). The acquired XRD data was then analyzed using Highscore software, and the parameters are listed in Table 1. All samples had a cubic crystal structure, indicating that the Cu and Zn atoms dissolve into one other. Presenting Zn in the alloy reduces crystallite size from 227.20 to 109.95 nm and increasing Zn content from 6 to 12 wt.% leads to increasing a crystallite size from 109.95 to 316.26 nm. This behavior is similar to Özdemir and Karahan's study that showed Zn in the alloy leads to decreased crystallite size and increase in Zn content in the alloy leads to increased crystallite size (Özdemir & Karahan, 2014). Moreover, Micro strain depends on Zn content, which perfectly agrees with Karahan and Özdemir study (Karahan & Özdemir, 2014).

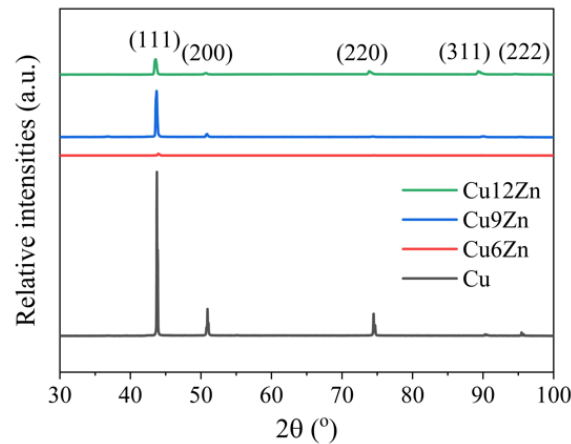


Fig. 1. XRD of various casting samples.

Table 1 - Crystallographic parameters of various casting samples.

Parameter	Sample			
	Cu	Cu6Zn	Cu9Zn	Cu12Zn
Crystal structure	cubic			
Lattice constant $a = b = c$ (Å)	2.964	8.929	2.981	2.964
Cell volume (Å ³)	26.03	90	26.49	26.05
d-spacing (Å)	1.69	1.18	1.69	2.07
Crystallite Size (nm)	227.20	109.95	204.95	316.26
Micro strain	0.37	0.53	0.16	0.32

4.2 Hardness

Figure 2 shows the average hardness of various casting samples. Nikhil et al. have found that pure Cu has a hardness of 140 HV when treated at 400 and 600 °C and then held for two hours, followed by quenching in tap water, resulting in a hardness of 100 and 60 HV (Nikhil et al., 2021). Therefore, the hardness of Cu may vary depending on heat treatment. In the present study, Cu re-casting has a hardness of 74.54 HV.

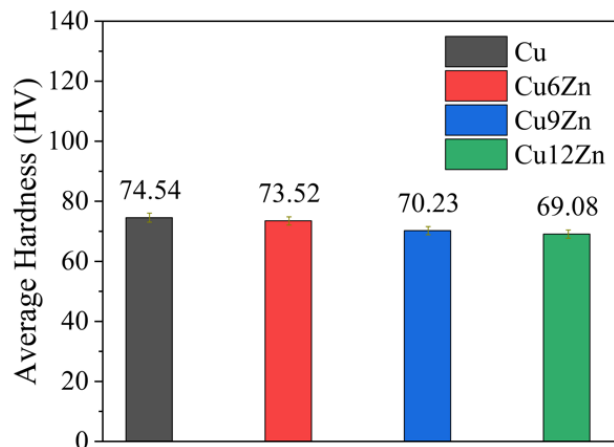


Fig. 2. Average hardness of various casting samples.

According to Figure 2, it can be seen that an increase in Zn content led to a decrease in the hardness. Another research study found that Cu (as-cast) has a hardness of around 100 HV, while Cu15 Zn (as-cast) has a hardness of 75 HV (Ezequiel et al., 2024). Nnakwo et al. also found that increased Zn content in the Cu alloy leads to decreased hardness (Nnakwo et al., 2021). According to a study by Qu et al. and García-Mintegui et al., pure Zn hardness is between 41-42 HV (García-Mintegui et al., 2021; Qu et al., 2020). Therefore, it could be concluded that Zn in the alloy leads to decreased hardness due to Zn hardness less than Cu.

4.3 OCP

Figure 3 shows the OCP measurement result of various casting samples in 0.9 % NaCl at room temperature. Generally, increased Zn content in the alloy promoted more negative potential, which perfectly agrees with the Cocco et al. study (Cocco et al., 2016). Dridi et al. have found that E_{OCP} CuZn30 and CuZn39 are -0.578 and -0.604 V/MSE at 3 % NaCl, which means an increase in the Zn promoted to more negative potential (Dridi et al., 2020). Cu, Cu6Zn, Cu9Zn, and Cu12Zn samples have E_{OCP} potential at 1200 s measurement -0.014, -0.023, -0.027, and -0.032 V vs Ag/AgCl, respectively.

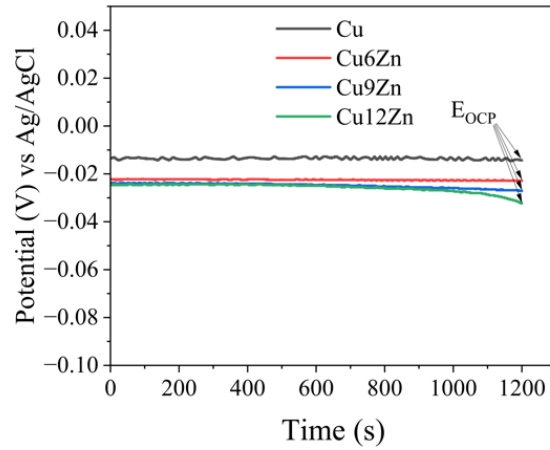


Fig. 3. OCP measurement of various casting samples.

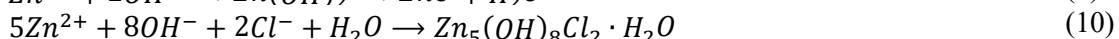
According to Figure 3, Cu, Cu6Zn, and Cu9Zn samples are steady at initial measurements until 1200 s, indicating that the protective layer formed has good protection. In contrast, the Cu12Zn sample is steady at initial measurements until 600 s, then moves in a more negative direction. These phenomena indicated that the formed protective layer had initially dissolved at 600 s; therefore, the measurement continuously moved forward in a negative direction until the measurement reached 1200 s.

4.4 LSV

Eccrine sweat and saline-infused for humans are nearly 0.9 % of NaCl (Bond & Lieu, 2014; Tayyab et al., 2021). Therefore, LSV measurement was conducted in 0.9 % NaCl at room temperature. Luo et al. found Cu₂O crystalline growth on the Cu surface when exposed to 0.9 % NaCl, while when exposed to pure water, Cu₂O crystalline was not seen (Luo et al., 2020). Commonly, Cu₂O crystallines are formed, which is preceded by the formation of CuCl when the specimen is tested in a chloride solution. Moreover, Zhang et al., in their study, found ZnO and Zn₅Cl₂(OH)₈·H₂O as corrosion products on top of Cu40Zn surfaces in a chloride environment (X. Zhang et al., 2016). The reaction of Cu in chloride solution is as follows (Milošev et al., 2024).



The reaction of Zn in chloride solution is as follows (Milošev et al., 2024).



LSV measurement results in 0.9 % NaCl can be seen in Figure 4.

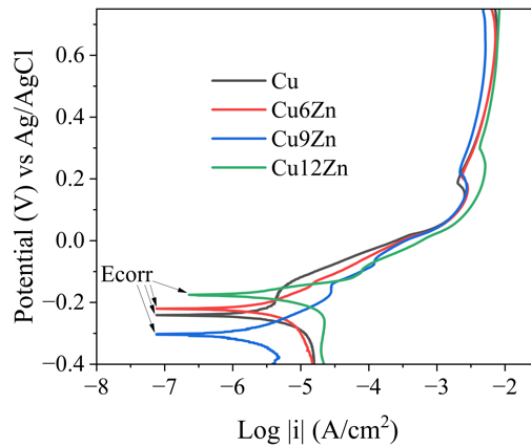


Fig. 4. LSV scans result of various casting samples.

According to Figure 4, corrosion E_{corr} and i_{corr} can be found using the Tafel extrapolation method. Moreover, the corrosion rate could be calculated by inserting i_{corr} into expression (1). Table 2 presented E_{corr} , i_{corr} , and the corrosion rate of various casting samples. It appears E_{corr} is independent of Zn addition; however, Zn is dependent on i_{corr} and corrosion rate (except for the Cu12Zn sample). This behavior is probably due to a protective layer that was formed. Compared to the OCP result, it can be seen that the Cu12Zn sample continuously moves forward in a negative direction from 600 until 1200 s of measurement. Therefore, that sample has a higher i_{corr} and corrosion rate.

Table 2 - Corrosion parameters of various casting samples.

Sample name	E_{corr} (V) vs Ag/AgCl	i_{corr} (A/cm ²)	Corrosion rate (mmpy)
Cu	-0.241	4.42×10^{-6}	5.13×10^{-2}
Cu6Zn	-0.220	3.33×10^{-6}	3.86×10^{-2}
Cu9Zn	-0.304	2.15×10^{-6}	2.49×10^{-2}
Cu12Zn	-0.175	6.19×10^{-6}	7.18×10^{-2}

Qu et al. have found that an increase in Zn content led to an increase in corrosion resistance, which perfectly agrees with the present study (except for Cu12Zn) (Qu et al., 2020). Milošev et al. investigated Cu, Cu10Zn, Cu40Zn, and Zn in 3 % NaCl and found i_{corr} after stabilized at 1 hour around 1.573, 1.456, and 2.114, and 5.21 $\mu\text{A}/\text{cm}^2$ respectively (Milošev et al., 2024). According to equation (1), i_{corr} strongly influences the corrosion rate. The more i_{corr} , the higher the corrosion rate. Moreover, a limitation in Zn content in the Cu alloy could influence the corrosion resistance. Presenting the Zn content ≤ 11 wt.% in the alloy could enhance the corrosion resistance; however, Zn of more than 10 wt.% could decrease corrosion resistance (Milošev et al., 2024).

4.5 Antibacterial Activity

Figure 5 shows the direct contact kill of *Staphylococcus aureus* and *Escherichia coli* after 24 hours of incubation. The present study focused on *Staphylococcus aureus*, but *Escherichia coli* was also used for comparison. There is no diffusion in the sample; therefore, the inhibition zone could not be seen.

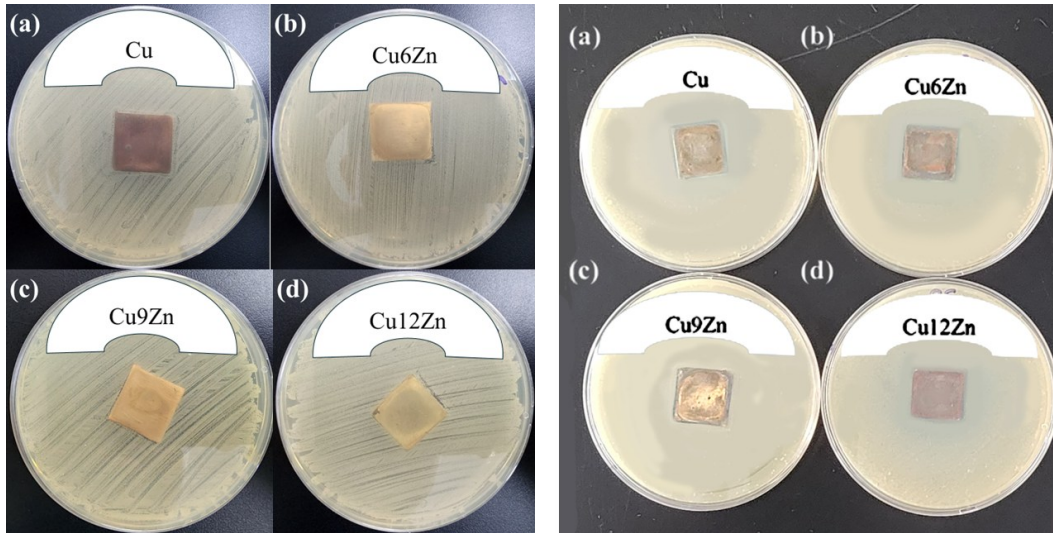


Fig. 5. Antibacterial activity test using *Staphylococcus aureus* (left) and *Escherichia coli* (right) (a) Cu, (b) Cu6Zn, (c) Cu9Zn, and (d) Cu12Zn.

Antibacterial activity after 24 hours of post-contact with various casting samples using *Staphylococcus aureus* and *Escherichia coli* can be seen in Figure 6. The removed sample places remain clear (with no regrowth) from bacterial activity. The antibacterial behavior was significantly influenced by Cu or Zn ions (Qu et al., 2020). Villapún et al. found that releasing Cu ions leads to the highest killing activity of *Staphylococcus aureus* (Villapún et al., 2016). Excess in the Cu ion could be bacteriostatic (Sabbouh et al., 2023). Moreover, Cu ions could be adsorbed on the cytoplasmic membrane surfaces, then penetrate the bacteria, react with sulfhydryl groups and cause the cell to die. Furthermore, Cu ions could form hydroxyl groups in the presence of oxygen in nature, which could destroy cell membranes (Chen et al., 2014; Dou et al., 2022). Hutchings et al. stated that Zn^{2+} successfully inhibits the growth of *S. epidermidis* (Hutchings et al., 2021). This behavior is associated with the generation of reactive oxygen or the formation of (Chen et al., 2014; E. Zhang et al., 2021). Cu killing is more effective in Gram negative bacteria because peptidoglycan affects the cell's susceptibility. The thicker the peptidoglycan layer, the harder it became for the Cu ions to reach the membrane (Xhafa et al., 2023).

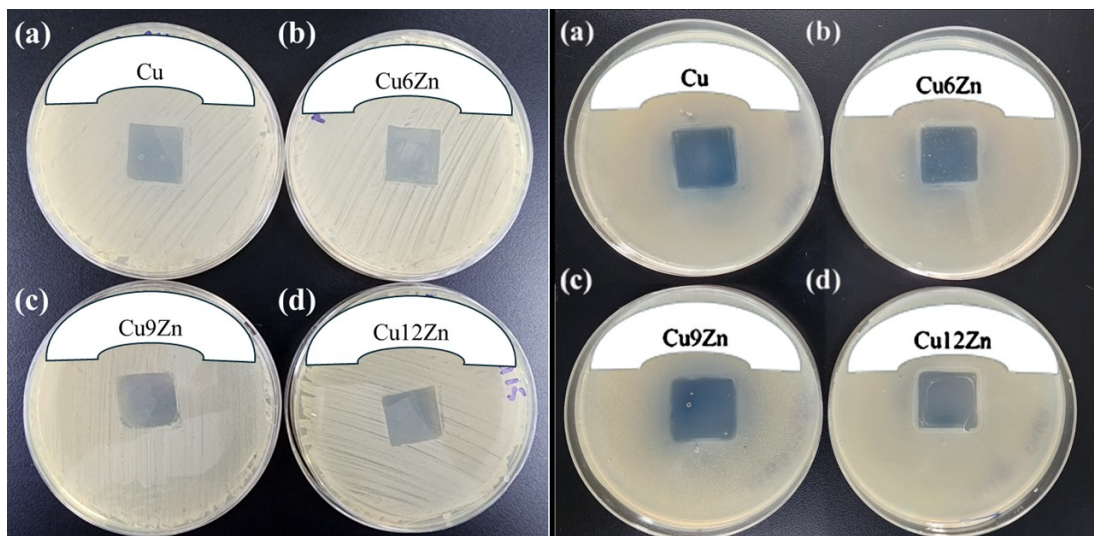


Fig. 6. Antibacterial activity after 24 hours of post-contact (regrowth assessment) towards *Staphylococcus aureus* (left) and *Escherichia coli* (right) (a) Cu, (b) Cu6Zn, (c) Cu9Zn, and (d) Cu12Zn

Figure 7 shows the fluid contact test of *Staphylococcus aureus* and *Escherichia coli*. The orientation of the test materials is mapped within the yellow box. For the Cu6Zn sample,

Escherichia coli was killed on the 3rd hour. However, there is no significant reduction within the fluid because there are no diffusible materials. Also, there is no visible growth after 3rd hour for the fluid in contact with the metal.

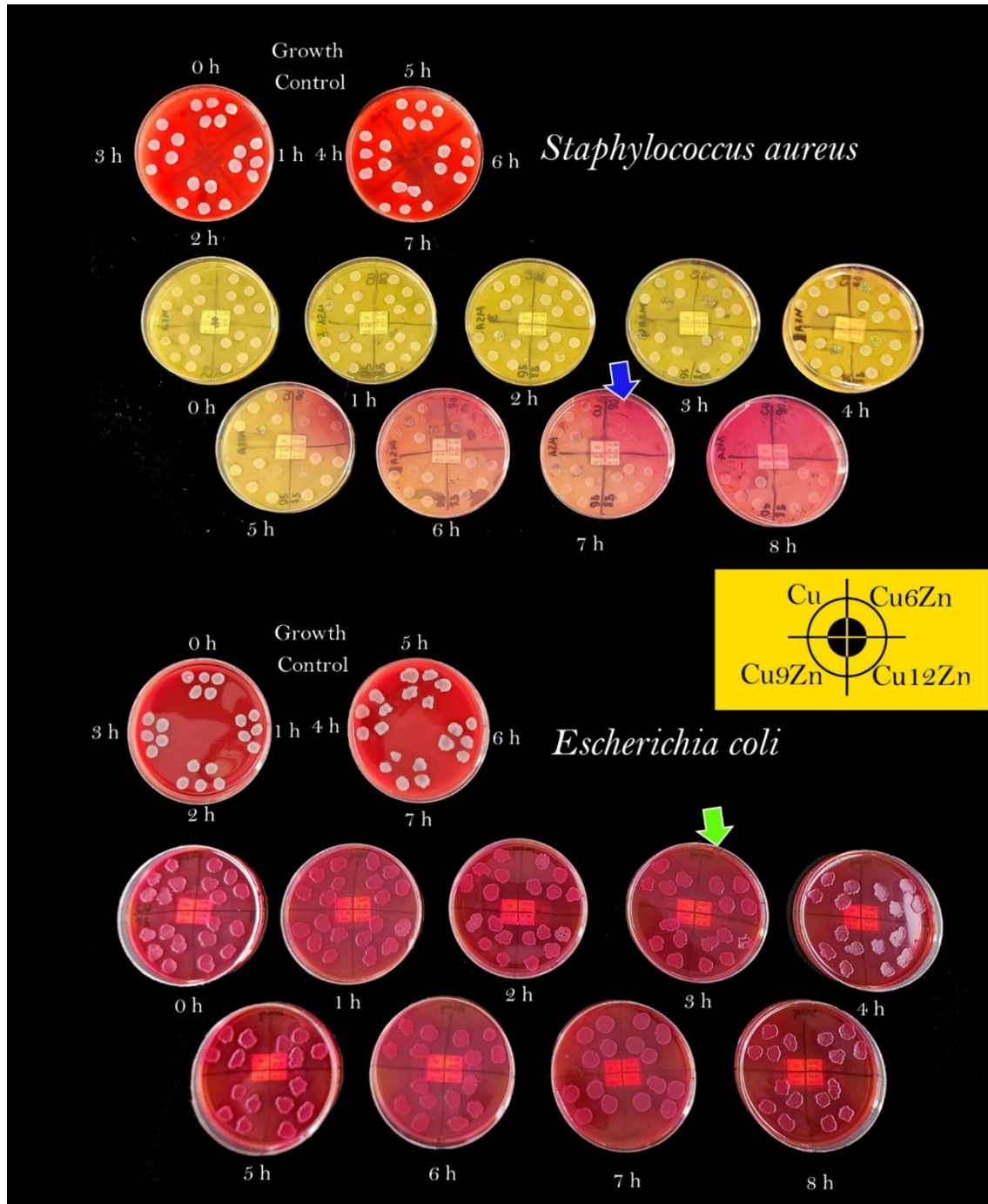


Fig. 7. Fluid contact test of *Staphylococcus aureus* and *Escherichia coli*

Moreover, it should be noted that the reduction of the colony is significant in the 7th hour for *Staphylococcus aureus* (blue arrow), but for *Escherichia coli*, the inhibition of *Escherichia coli* growth is shown within the 3rd hour of contact (green arrow), for Cu6Zn sample. The reduction of colonies is significant on the surface of the metal. While on the remaining fluid, the reduction is insignificant until 8 hours. This behavior is because *Staphylococcus aureus* and *Escherichia coli* have different membrane structures and thick cell walls, therefore could inhibit ion exchange and restrain the antibacterial effect of Cu and or Zn ions (Alzahrani et al., 2018; Di et al., 2022). Another reason Cu6Zn has better antibacterial performance than others are probably due to its

smaller crystallite size than others. Researchers found that smaller crystallite size promoting to enhance antibacterial effects (Sangeetha et al., 2015; Syamsuir et al., 2023). This behavior is attributed to an increased in surface area due to crystallite size reduced (Sangeetha et al., 2015).

5. Conclusion

CuZn has been successfully fabricated. XRD confirmed that CuZn alloy has a single alpha phase with an FCC crystal structure. The rise of the Zn content in the alloy led to a decrease in the hardness due to Zn hardness less than Cu and led to a shift to more negative OCP potential at 1200 s measurement. Moreover, the rise of the Zn content to 9 wt.% decreased the corrosion rate. It appears there is a limitation in Zn content in the copper alloy that influences the corrosion rate, as shown when Zn content around 12 wt. % is promoted to increase the corrosion rate. Antibacterial activity observation found that all samples had no diffusion. Moreover, 24-hour post-contact observation found that sample places removed from the sample remained clear of bacteria. The Cu₆Zn has better antibacterial performance than others, according to the fluid contact test, which shows that the reduction of the colony of *Staphylococcus aureus* is significant in the 7th hour. The inhibition of *Escherichia coli* growth is also shown within the 3rd hour of contact.

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2. BUKTI KONFIRMASI REVIEW

(6 November 2024)

Bukti dari OJS

6098 / Samura et al. / Effect of Zinc Addition in Copper to Structure, Hardness, Corrosion, and Antibacterial Activity

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Notifications

[JAETS] Editor Decision	2024-11-06 02:36 PM
[JAETS] Editor Decision	2024-11-13 08:01 AM
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[JAETS] Editor Decision	2024-12-13 02:34 PM

Bukti email

4 of 8

[JAETS] Editor Decision External Imported

Muhammad Luthfi Hamzah editor@jaets@gmail.com via journal.yrpipku.com
to me, Mustamina, Cahaya, Kartika, Suryo, Evi, Danie, Bambang, Muhammad, Ferry

Nov 13, 2024, 8:01 AM

Lise Samura, Mustamina Maulani, Cahaya Rosyidan, Kartika Fajarwati Hartono, Suryo Prakoso, Evi Ulina Margareta Situmorang, Daniel Edbert, Bambang Soegijono, Muhammad Yunan Hasbi, Ferry Budhi Susetyo

We have reached a decision regarding your submission to Journal of Applied Engineering and Technological Science (JAETS), "EFFECT OF ZINC ADDITION IN COPPER TO STRUCTURE, HARDNESS, CORROSION, AND ANTIBACTERIAL ACTIVITY".

Our decision is: Revisions Required



[JAETS] Editor Decision

2024-11-13 08:01 AM

Lisa Samura, Mustamina Maulani, Cahaya Rosyidan, Kartika Fajarwati Hartono, Suryo Prakoso, Evi Ulina
Margareta Situmorang, Daniel Edbert, Bambang Soegijono, Muhammad Yunan Hasbi, Ferry Budhi Susetyo:

We have reached a decision regarding your submission to Journal of Applied Engineering and Technological Science (JAETS), "EFFECT OF ZINC ADDITION IN COPPER TO STRUCTURE, HARDNESS, CORROSION, AND ANTIBACTERIAL ACTIVITY".

Our decision is: Revisions Required

Reviewer A:

Introduction :

While the text mentions that the addition of Zn in CuZn alloys with compositions of 6, 9, and 12 wt.% has not been comprehensively investigated recently, it would be useful to further elaborate on the specific gap in knowledge or application. For instance, it could mention whether there is a lack of research on the combined effects of Zn content on corrosion resistance, antibacterial properties, and mechanical properties, especially in the context of medical transportation (Research Gap Clarification).

The introduction references several studies related to the properties of CuZn alloys (e.g., corrosion resistance, antibacterial properties), but the connection to the specific research focus (medical transportation) could be more explicit. For example, it would be useful to explain how the research will extend current knowledge on CuZn alloys to address issues specific to medical transportation or medical equipment.

The introduction references several studies related to the properties of CuZn alloys (e.g., corrosion resistance, antibacterial properties), but the connection to the specific research focus (medical transportation) could be more explicit. For example, it would be useful to explain how the research will extend current knowledge on CuZn alloys to address issues specific to medical transportation or medical equipment. Suggestion: After mentioning existing research, transition smoothly into explaining why the application of CuZn alloys in medical transportation specifically needs further exploration, and how this research will address that need.

The introduction ends with a mention of the research objectives but does not provide a concluding summary or a clear statement of how the research contributes to advancing the field. A strong concluding sentence would help tie together the information and set the stage for the following sections of the paper.

Literature review :

While various studies are mentioned, the review does not explicitly highlight a research gap that this study aims to address. For example, it could mention that although there is ample research on the mechanical properties and antibacterial properties of CuZn alloys, there is a lack of comprehensive studies on their combined performance (e.g., how both properties interact or affect each other in specific applications such as medical transportation). Explicitly state the gap in current knowledge or research, especially in the context of this study's aim to investigate CuZn alloys for medical transportation, and how previous research has not sufficiently addressed this aspect.

The literature review mentions many studies but does not clearly tie them to the specific focus of the current research—CuZn alloys for medical transportation. While the properties of CuZn alloys (hardness, antibacterial, corrosion resistance) are discussed, the introduction of medical application as a central theme could be more strongly integrated throughout the review. Tie the studies more directly to the context of medical transportation, by explaining how CuZn alloys with varying Zn content may be suited for specific requirements in medical equipment (e.g., antimicrobial surfaces, durability in a medical environment, etc.).

The review could be more explicit in discussing the novelty of the current research, especially in light of recent studies. For instance, while the studies cited discuss various Zn content ranges and their effects, few studies appear to focus on the specific Zn compositions (6, 9, and 12 wt.%) or applications for medical transportation, which could be a key contribution of this research. Highlight the novelty of the study more clearly. For example, indicate that research on CuZn alloys with Zn compositions in the range of 6–12 wt.% for medical transportation purposes has not been thoroughly investigated, making the current study unique.

The review provides citations from a wide range of studies, but there is no mention of particularly recent or high-impact studies (e.g., from the last 3-5 years) that could demonstrate ongoing trends or breakthroughs in CuZn alloy research. Providing a more up-to-date review could enhance the relevance of the review and the study. Incorporate more recent studies to provide a more current perspective on CuZn alloys, especially research conducted in the last 3-5 years.

The review lacks a conclusive paragraph that ties together the findings from the various studies. A summary of key findings, as well as an explicit statement on how the research builds upon or addresses the gaps in existing knowledge, would help set up the research objectives more clearly. Conclude the literature review with a summary that synthesizes the key findings from the studies and explicitly connects them to the goals of the present research.

Research Methods :

While the alloy composition (Cu, Cu₆Zn, Cu₉Zn, and Cu₁₂Zn) is provided, the exact weight of Cu and Zn used in each alloy should be clarified, especially since Cu is in ingot form and Zn is in powder form. Additionally, it would be useful to mention any prior treatments or cleaning steps for the materials before casting. Specify the exact quantities (in grams or other units) of Cu ingot and Zn powder used for each composition. Also, provide any details on material preparation steps (e.g., cleaning, drying) before casting.

The method of gravity casting is mentioned, but more details on the casting setup could be helpful. For instance, were the Cu and Zn components melted separately before mixing, or were they directly combined in the crucible? What type of crucible was used (material, size, etc.), and what was the cooling rate of the cast samples? Include more detailed information on the casting procedure, including the preparation of the materials, mixing process, crucible specifications, and cooling rate, if available.

XRD Measurement : Provide additional information on the data analysis process using Highscore software, such as peak identification and phase determination. Mention if any reference materials were used for comparison.

Add details on sample cleaning or any surface treatments that were done prior to polishing. Clarify whether any surface defects were removed to ensure the accuracy of the hardness measurements.

Result and discussion :

Interpretation of XRD Data: While the XRD results and crystal structure are discussed well, it would be useful to elaborate on the significance of the observed phases in terms of material properties (e.g., mechanical, corrosion resistance). The statement that “all samples had a cubic crystal structure” should be linked to how this structure relates to the properties of the CuZn alloy, especially in terms of the alloy's performance in real-world applications (e.g., corrosion, strength). Suggestion: Add a brief explanation of why the cubic crystal structure is significant for the alloy's properties, and how it may affect the material's performance in specific applications such as corrosion resistance or mechanical strength.

Provide a clearer explanation of the relationship between crystallite size, microstrain, and material properties like hardness and corrosion resistance. Discuss whether the trends observed are desirable for the specific application of CuZn alloys.

Relate the hardness results to the potential applications of the CuZn alloy, such as its suitability for medical implants, corrosion-resistant parts, or mechanical components. Discuss whether the decrease in hardness with Zn addition is an advantage or limitation based on the alloy's intended use.

Analyze the data from Figure 2 in more detail, including commenting on any trends, anomalies, or unexpected results. Provide a more in-depth interpretation of the hardness values, especially in relation to Zn content.

References to Other Studies: The comparison with previous studies (e.g., Nikhil et al., Ezequiel et al., Nnakwo et al.) is appropriate, but it could be expanded. For instance, is the observed trend in your study consistent with the findings of other researchers, or are there notable differences? Also, what could explain the discrepancies, if any, in the results? Suggestion: Expand the comparison to other studies by addressing whether your findings align or diverge from theirs, and discuss possible reasons for any differences.

The discussion of antibacterial activity is good, but it would benefit from a deeper analysis of the results. For example, the statement about *Staphylococcus aureus* showing significant reduction after 7 hours and *Escherichia coli* after 3 hours is helpful, but there could be more explanation as to why this difference exists, beyond the basic membrane structure explanation. Suggestion: Provide a more detailed analysis of why *Staphylococcus aureus* and *Escherichia coli* show different inhibition times, discussing the different types of antibacterial activity (e.g., ion release, surface interaction) and how the alloy's properties may influence these activities.

Linking Results to Research Gap: The discussion could benefit from stronger linkage to the research gap outlined in the introduction and literature review. For example, the studies on Zn content in CuZn alloys are mentioned, but there could be more emphasis on how your study fills the gap or provides new insights. Suggestion: Tie the results back to the research gap and highlight how the findings contribute to advancing knowledge in the field, particularly regarding the antibacterial properties of CuZn alloys in medical or industrial

applications. Implications of Findings: While the results are discussed in relation to previous studies, the broader implications of these findings for real-world applications are not fully explored. For instance, how do the hardness and antibacterial properties affect the potential use of CuZn alloys in medical devices, valves, or other applications? Suggestion: Discuss the broader implications of your findings in terms of real-world applications, such as medical implants, industrial machinery, or other sectors where CuZn alloys are used.

Recommendation: Revisions Required

Reviewer B:

Abstract:

Please rewrite your abstract. Currently, it reads too narratively and focuses more on the procedural aspects of the research rather than providing a concise summary of the entire study. The abstract should include the following key points:

1. Purpose of the study: Briefly state the aim of your research and explain why the study was conducted.
2. Methodology: Outline the methods used to address the research problem.
3. Results: Summarize the key findings of your study.
4. Implications: Discuss the practical and theoretical implications of your results. How can the findings be applied in both theory and practice?
5. Contribution: State the value of your study. What is the unique contribution of your research to the field of study?

Introduction:

The introduction section needs to be expanded to provide more context and clarity. Please ensure that it includes the following:

1. Background: Clearly describe the context of the study and provide an overview of the current state-of-the-art in the field. This section should offer readers an understanding of the existing knowledge and research gaps.

2. Problem Statement: Identify the problem your study addresses. Explain why the problem is significant and why readers should be interested in the research.
3. Proposed Solution: Once the problem is identified, clearly outline the contribution of your work. Highlight the novel aspects of your research and provide a framework that connects it with the existing literature. You should cite at least 8 recent journal articles to support this section.
4. Literature Gaps: Summarize the existing gaps in the literature that your research intends to fill, and present the specific aim of your study.
5. Use of International Sources: Please ensure that you use international journal references, not local sources, to strengthen the relevance of your work.

Literature Review:

While the literature review covers relevant works, it lacks a critical analysis and synthesis of the literature. It currently reads as a list of studies, but a deeper, more critical evaluation is necessary. Here's how to improve it:

1. Critical Analysis: Provide a more comprehensive review of the literature, offering critical commentary and drawing connections between studies to highlight trends, contradictions, or gaps in knowledge. This should not just be a factual listing but a thoughtful analysis of the research.
2. Recent References: Include more recent journal articles (published from 2019-2024) to ensure the literature review reflects the current state of the field. This will also help support the argument for the novelty and relevance of your research.
3. Theoretical Framework: The review should present the theoretical framework underpinning your study. Explain how the selected literature connects with your research questions and objectives.
4. Selection of Literature: Describe how the literature was selected for inclusion. Explain the criteria used to ensure a comprehensive and representative review of the state of the art.

Methodology:

Please clarify the methodology section by addressing the following:

1. Justification of Methods: Avoid conceptual descriptions and instead explain why each method was chosen for your study. Discuss the rationale behind selecting specific approaches, tools, and techniques.
2. Participants and Characteristics: Provide detailed information about the participants (if applicable) and their characteristics. Who were the subjects, and why were they chosen for this research?

3. Data Collection and Instruments: Describe the data to be collected and the instruments used to gather that data. Include an explanation of how the instruments were applied to ensure reliability and validity in your results.
4. Explanation of Images: If any images are included in the methodology, add an explanation of what information can be gleaned from them.

Results and Discussion:

1. Discussion Structure: The results and discussion sections should be presented in continuous prose, without numbering or bullet points. Ensure that there is a smooth flow between sections, with connecting sentences to guide the reader.
2. Comparison with Previous Studies: Your discussion should compare your findings with previous research. Please include at least 15 relevant studies from international journals. This will help contextualize your results and demonstrate how your research fits within the broader body of knowledge.
3. Generalizations: Conclude the discussion with a broader perspective, offering generalizations or conclusions based on your findings.

Conclusion:

The conclusion should summarize the main outcomes of the study without using numbered points. It should reflect on the implications of your findings, both theoretical and practical. Highlight the significance of your study and why it contributes to the existing literature.

References:

1. Minimum Number of References: Include at least 40 references, with a minimum of 80% sourced from SCOPUS, Web of Science (WoS), or other reputable international journals.
2. Citations within the Text: Ensure that all references are properly cited within the text. Avoid listing references without citation. Use a reference management tool like Mendeley to ensure proper formatting and consistency.
3. Recent Sources: Prioritize recent journal articles (preferably from 2019 onward) to maintain the relevance and currency of your work.

Recommendation: Revisions Required

3. BUKTI MENJAWAB REVIEWER

(17 November 2024)

Title : Effect of Zinc Addition In Copper to Structure, Hardness, Corrosion, and Antibacterial Activity

Authors : Lisa Samura, Mustamina Maulani, Cahaya Rosyidan, Kartika Fajarwati Hartono, Suryo Prakoso, Evi Ulina Margareta Situmorang, Daniel Edbert, Bambang Soegijono, Muhammad Yunan Hasbi, Ferry Budhi Susetyo

Corresponding author : Lisa Samura

#Editorial Office of Journal of Applied Engineering and Technological Science (JAETS)

We have reached a decision regarding your submission to Journal of Applied Engineering and Technological Science (JAETS), "EFFECT OF ZINC ADDITION IN COPPER TO STRUCTURE, HARDNESS, CORROSION, AND ANTIBACTERIAL ACTIVITY".

Our decision is: Revisions Required

Response:

We are grateful for this decision. In the following Table, we provide a point-by-point response explaining how we have addressed each reviewer's comments. Thank you.

Reviewer #1:

No.	Comment from Reviewer	Author Response
a	Introduction :	
	1. While the text mentions that the addition of Zn in CuZn alloys with compositions of 6, 9, and 12 wt.% has not been comprehensively investigated recently, it would be useful to further elaborate on the specific gap in knowledge or application. For instance, it could mention whether there is a lack of research on the combined effects of Zn content on corrosion resistance, antibacterial properties, and mechanical properties, especially in the context of medical transportation (Research Gap Clarification).	CuZn corrosion has been compared to Al alloy when used in the ambulance as follows: "Gao et al. found that a reduction in thickness (50 to 60 %) of CuZn using cold rolling resulted in a significant decrease in corrosion current from 4.824 to 1.804 $\mu\text{A}/\text{cm}^2$ (investigating in 3.5 % NaCl) (Gao et al., 2021). Moreover, aluminum (Al) alloy widely used in transportation sector such as ambulance (Blanco et al., 2022; Vandersluis et al., 2020). Liu et al. have found Al alloy corrosion

		<p>current between 4.868-5.251 A/cm² in a 3.5% NaCl medium (Liu et al., 2020). Comparing the studies of Liu et al. and Gao et al., Al alloy has a higher corrosion current than CuZn (Gao et al., 2021; Liu et al., 2020). Corrosion current significantly influences the corrosion rate, and a rise in the corrosion current would enhance the corrosion rate.”</p> <p>The killing or inhibition mechanism of bacterial activity is explained as follows: “The killing mechanism of bacterial activity inseparable from the ions released by the alloy (Qu et al., 2020). Cu²⁺ ions could be adsorbed on the cytoplasmic membrane surfaces, then penetrate the bacteria, react with sulfhydryl groups, and cause the cell to die (Zeng et al., 2022). The released Zn²⁺ ions could penetrate the cell membrane and cause cell death (Du et al., 2021). Zhang et al. have stated that Cu²⁺ and Zn²⁺ ions could act as antibacterial agents and inhibit <i>Staphylococcus aureus</i> growth (E. Zhang et al., 2021).”</p> <p>Thank you.</p>
	2. The introduction references several studies related to the properties of CuZn alloys (e.g., corrosion resistance, antibacterial properties), but the connection to the specific research focus (medical transportation) could be more explicit. For example, it would be useful to explain how the research will extend current knowledge on CuZn alloys to address issues specific to medical transportation or medical equipment.	<p>Reviewer comment number a.2 is similar to a.3. Therefore, we answer in the number a.3.</p> <p>Thank you.</p>
	3. The introduction references several studies related to the properties of CuZn alloys (e.g., corrosion resistance, antibacterial properties), but the connection to the specific research focus (medical transportation) could be more explicit. For example, it would be useful to explain how	<p>The connection has been added as follows: “CuZn alloys for medical equipment in transportation such as ambulances need to consider two parameters: corrosion resistance and antibacterial characteristics. Commonly NaCl</p>

	<p>the research will extend current knowledge on CuZn alloys to address issues specific to medical transportation or medical equipment.</p> <p>Suggestion: After mentioning existing research, transition smoothly into explaining why the application of CuZn alloys in medical transportation specifically needs further exploration, and how this research will address that need.</p>	<p>media was used to investigate corrosion for ambulance equipment. This condition due to ambulance equipment commonly exposure from medical patients eccrine sweat and saline-infused (Baker & Wolfe, 2020; Tayyab et al., 2021). Furthermore, some studies found that after cleaning the ambulance, 35.37 % of bacterial contaminants were still seen (Syamsuir et al., 2023).</p> <p>Viegas et al. found <i>Staphylococcus aureus</i> was detectable on firefighter's ambulance equipment (Viegas et al., 2021). <i>Staphylococcus aureus</i> is a type of bacteria that could cause skin disease and is hard to treat with traditional antibiotics. <i>Staphylococcus aureus</i> bacteria tend to have methicillin resistance. According to Tajik et al., 38.4 % of <i>Staphylococcus aureus</i> methicillin resistant was found in Tehran community (Tajik et al., 2020). Moreover, this bacteria also could contaminate orthopedic implants and cause serious infections (Pietrocola et al., 2022).”</p> <p>Furthermore:</p> <p>“Azizian et al. investigated CuZn alloys microstructure, mechanical properties and cytotoxicity for cardiovascular applications (Azizian et al., 2024). Riaz et al. investigated the structural and biological properties of CuZn alloy for orthopedic applications (Riaz et al., 2024). Moreover, Sabbouh et al. did the sonification of CuZn in an alkali solution to enhance the antibacterial inhibition zone (Sabbouh et al., 2023). Moreover, Syamsuir et al. have investigated the antibacterial activity of <i>Staphylococcus aureus</i> by presenting a Cu layer for ambulance equipment (Syamsuir et al., 2023).”</p> <p>Thank you.</p>
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	<p>4. The introduction ends with a mention of the research objectives but does not provide a concluding summary or a clear statement of how the research contributes to advancing the field. A strong concluding sentence would help tie together the information and set the stage for the following sections of the paper.</p>	<p>The end of the introduction has been revised to:</p> <p>“According to the literature review, research on CuZn alloys with Zn compositions in the range of 6–12 wt.% for medical transportation purposes has not been thoroughly investigated. As mentioned above, the killing mechanism of bacterial activity depend on Cu and Zn ions. CuZn alloy can transform into Cu and Zn ions. Therefore, the present research casts CuZn alloy by adding various Zn to the Cu to investigate optimum composition, resulting in a higher killing mechanism of bacterial activity. Moreover, different alloy compositions would result in different electrochemical behavior and mechanical properties. The present study investigated structure, hardness, electrochemical behavior, and antibacterial activity using X-ray diffraction (XRD), Vickers hardness equipment, potentiostat, and digital camera.”</p> <p>Thank you</p>
b	Literature review :	
	<p>1. While various studies are mentioned, the review does not explicitly highlight a research gap that this study aims to address. For example, it could mention that although there is ample research on the mechanical properties and antibacterial properties of CuZn alloys, there is a lack of comprehensive studies on their combined performance (e.g., how both properties interact or affect each other in specific applications such as medical transportation). Explicitly state the gap in current knowledge or research, especially in the context of this study’s aim to investigate CuZn alloys for medical transportation, and how previous research has not sufficiently addressed this aspect.</p>	<p>In the literature review, we want to show the reader that CuZn alloy composition was investigated between 2019 and 2024. According to the literature review, the reader could know CuZn with compositions Cu6Zn, Cu9Zn, and Cu12Zn have not been examined, especially for corrosion and bacterial activity.</p> <p>Thank you.</p>
	<p>2. The literature review mentions many studies but does not clearly tie them to the specific focus of the current research—</p>	<p>As mentioned above (answered in b.1 point). In the literature review, we want to show the reader that CuZn</p>

	<p>CuZn alloys for medical transportation. While the properties of CuZn alloys (hardness, antibacterial, corrosion resistance) are discussed, the introduction of medical application as a central theme could be more strongly integrated throughout the review. Tie the studies more directly to the context of medical transportation, by explaining how CuZn alloys with varying Zn content may be suited for specific requirements in medical equipment (e.g., antimicrobial surfaces, durability in a medical environment, etc.).</p>	<p>alloy composition was investigated between 2019 and 2024. Of course, between those years, finding research directly related to medical equipment in ambulances was difficult. Therefore, we only describe other people's research that has been carried out. However, in the introduction section, we try to explain the problems that exist in ambulances.</p> <p>Thank you.</p>
	<p>3. The review could be more explicit in discussing the novelty of the current research, especially in light of recent studies. For instance, while the studies cited discuss various Zn content ranges and their effects, few studies appear to focus on the specific Zn compositions (6, 9, and 12 wt.%) or applications for medical transportation, which could be a key contribution of this research. Highlight the novelty of the study more clearly. For example, indicate that research on CuZn alloys with Zn compositions in the range of 6–12 wt.% for medical transportation purposes has not been thoroughly investigated, making the current study unique.</p>	<p>The novelty has been revised to: “According to the literature review, research on CuZn alloys with Zn compositions in the range of 6–12 wt.% for medical transportation purposes has not been thoroughly investigated.”</p> <p>Thank you.</p>
	<p>4. The review provides citations from a wide range of studies, but there is no mention of particularly recent or high-impact studies (e.g., from the last 3-5 years) that could demonstrate ongoing trends or breakthroughs in CuZn alloy research. Providing a more up-to-date review could enhance the relevance of the review and the study. Incorporate more recent studies to provide a more current perspective on CuZn alloys, especially research conducted in the last 3-5 years.</p>	<p>The introduction and literature review section has been revised to recent literature (2019-2024).</p> <p>Thank you.</p>
	<p>5. The review lacks a conclusive paragraph that ties together the findings from the various studies. A summary of key findings, as well as an explicit statement on how the research builds upon or addresses the gaps in existing knowledge, would help set up the research objectives more clearly. Conclude the literature</p>	<p>The introduction and literature review sections have been revised. Thank you.</p>

	review with a summary that synthesizes the key findings from the studies and explicitly connects them to the goals of the present research.	
c	Research Methods :	
	1. While the alloy composition (Cu, Cu ₆ Zn, Cu ₉ Zn, and Cu ₁₂ Zn) is provided, the exact weight of Cu and Zn used in each alloy should be clarified, especially since Cu is in ingot form and Zn is in powder form. Additionally, it would be useful to mention any prior treatments or cleaning steps for the materials before casting. Specify the exact quantities (in grams or other units) of Cu ingot and Zn powder used for each composition. Also, provide any details on material preparation steps (e.g., cleaning, drying) before casting.	<p>Unfortunately, we cannot present the exact weight in the methodology. We do not want to confuse the reader because the Zn dust used in the sample preparation many looses were poured into the crucible because they evaporated during direct contact with melted Cu. However, we present the composition of the formed alloy based on the XRF test results. Furthermore, cleaning was conducted for the Cu ingot and the crucible.</p> <p>Thank you.</p>
	2. The method of gravity casting is mentioned, but more details on the casting setup could be helpful. For instance, were the Cu and Zn components melted separately before mixing, or were they directly combined in the crucible? What type of crucible was used (material, size, etc.), and what was the cooling rate of the cast samples? Include more detailed information on the casting procedure, including the preparation of the materials, mixing process, crucible specifications, and cooling rate, if available.	<p>The method of gravity casting has been added as follows:</p> <p>“Before melting was conducted, the ingot and the apparatus, such as the crucible, were cleaned using water to avoid impurities and then dried. The Cu ingot was first filled into a silicon carbide crucible (3kg) and then inserted into a muffle furnace. Melting was carried out in a crucible at 1100 °C under atmospheric pressure. After the Cu has melted, remove it from the muffle furnace, mix it with Zn powder, stir it manually, and pour it into a permanent mold. For comparison, Cu ingot was melted and poured into a permanent mold without Zn addition. The as-cast ingots were cut for further characterization, such as XRD, hardness, electrochemical measurement, and antibacterial activity observation.”</p> <p>Thank you.</p>
	3. XRD Measurement : Provide additional information on the data analysis process using Highscore software, such as peak identification and phase determination. Mention if any reference materials were used for comparison.	<p>The XRD measurement has been revised.</p> <p>“XRD was measured using the PANalytical (Cu Kα1 λ=1.5405980) apparatus. XRD was scanned from 20 to 100°, using step size 0.0217°. The</p>

		<p>Highscore software was used to refine and collect peak, phase and crystallographic parameters of as-cast samples. By using that software, full width at half maximum (FWHM) are also found. FWHM data is used to calculate crystallite size.”</p> <p>Thank you.</p>
	<p>4. Add details on sample cleaning or any surface treatments that were done prior to polishing. Clarify whether any surface defects were removed to ensure the accuracy of the hardness measurements.</p>	<p>Sample cleaning for hardness measurement has been added as follows:</p> <p>“Afterward, the polished sample was cleaned using water, followed by alcohol, and then dried using drier equipment.”</p> <p>Thank you.</p>
d	Result and discussion :	
	<p>1. Interpretation of XRD Data: While the XRD results and crystal structure are discussed well, it would be useful to elaborate on the significance of the observed phases in terms of material properties (e.g., mechanical, corrosion resistance). The statement that “all samples had a cubic crystal structure” should be linked to how this structure relates to the properties of the CuZn alloy, especially in terms of the alloy’s performance in real-world applications (e.g., corrosion, strength). Suggestion: Add a brief explanation of why the cubic crystal structure is significant for the alloy's properties, and how it may affect the material’s performance in specific applications such as corrosion resistance or mechanical strength.</p>	<p>XRD result has been elaborated as follows.</p> <p>“According to Jinlong et al., the FCC quantity of surface energy is (111)>(001) >(110), and the FCC sample with the (111) plane has the lowest corrosion rate (Jinlong et al., 2016). Moreover, the FCC sample with the preferred orientation of the (111) plane has the highest surface atomic density (Soegijono et al., 2020).”</p> <p>Thank you.</p>
	<p>2. Provide a clearer explanation of the relationship between crystallite size, microstrain, and material properties like hardness and corrosion resistance. Discuss whether the trends observed are desirable for the specific application of CuZn alloys.</p>	<p>The correlation between crystallite size and hardness has been discussed in the hardness section. Moreover, strain, (111) plane, and FCC correlate to corrosion behavior.</p> <p>Thank you.</p>
	<p>3. Relate the hardness results to the potential applications of the CuZn alloy, such as its suitability for medical implants, corrosion-</p>	<p>“Several transportation sectors, such as ambulance equipment, are made from Al alloy (Blanco et al., 2022;</p>

	<p>resistant parts, or mechanical components. Discuss whether the decrease in hardness with Zn addition is an advantage or limitation based on the alloy's intended use.</p>	<p>Vandersluis et al., 2020). According to Hajizadeh et al., Al alloy hardness is between 32-52 HV (Hajizadeh et al., 2017). Therefore, all specimens have hardness still higher than Al alloy.”</p> <p>Thank you.</p>
	<p>4. Analyze the data from Figure 2 in more detail, including commenting on any trends, anomalies, or unexpected results. Provide a more in-depth interpretation of the hardness values, especially in relation to Zn content.</p>	<p>More analysis has been added by comparing crystallite to the measured hardness.</p> <p>Thank you.</p>
	<p>5. References to Other Studies: The comparison with previous studies (e.g., Nikhil et al., Ezequiel et al., Nnakwo et al.) is appropriate, but it could be expanded. For instance, is the observed trend in your study consistent with the findings of other researchers, or are there notable differences? Also, what could explain the discrepancies, if any, in the results? Suggestion: Expand the comparison to other studies by addressing whether your findings align or diverge from theirs, and discuss possible reasons for any differences.</p>	<p>A comparison with other study discussions has been expanded.</p> <p>“Nikhil et al. have found that pure Cu has a hardness of 140 HV when treated at 400 and 600 °C and then held for two hours, followed by quenching in tap water, resulting in a hardness of 100 and 60 HV (Nikhil et al., 2021). The higher heat treatment temperatures led to a decrease in the hardness of the pure Cu. Therefore, the hardness of Cu may vary depending on heat treatment.”</p> <p>“Nnakwo et al. also found that increased Zn content in the Cu alloy leads to decreased hardness due to increased in grain size and solid solution region (Nnakwo et al., 2021).”</p> <p>Thank you.</p>
	<p>6. The discussion of antibacterial activity is good, but it would benefit from a deeper analysis of the results. For example, the statement about Staphylococcus aureus showing significant reduction after 7 hours and Escherichia coli after 3 hours is helpful, but there could be more explanation as to why this difference exists, beyond the basic membrane structure explanation. Suggestion: Provide a more detailed analysis of why Staphylococcus aureus and Escherichia coli show different inhibition times, discussing the different types of</p>	<p>“Cu killing is more effective in Gram-negative bacteria (e.g., <i>Escherichia coli</i>) because peptidoglycan affects the cell’s susceptibility. The thicker the peptidoglycan layer, the harder it became for the Cu ions to reach the membrane (Soltani et al., 2020; Xhafa et al., 2023). Therefore, <i>Escherichia coli</i> was killed in the 3rd hour.”</p> <p>Thank you.</p>

	antibacterial activity (e.g., ion release, surface interaction) and how the alloy's properties may influence these activities.	
7.	Linking Results to Research Gap: The discussion could benefit from stronger linkage to the research gap outlined in the introduction and literature review. For example, the studies on Zn content in CuZn alloys are mentioned, but there could be more emphasis on how your study fills the gap or provides new insights. Suggestion: Tie the results back to the research gap and highlight how the findings contribute to advancing knowledge in the field, particularly regarding the antibacterial properties of CuZn alloys in medical or industrial applications.	<p>“The Cu₆Zn sample could be used as an alternative material for medical equipment in ambulances.”</p> <p>Thank you.</p>
8.	Implications of Findings: While the results are discussed in relation to previous studies, the broader implications of these findings for real-world applications are not fully explored. For instance, how do the hardness and antibacterial properties affect the potential use of CuZn alloys in medical devices, valves, or other applications? Suggestion: Discuss the broader implications of your findings in terms of real-world applications, such as medical implants, industrial machinery, or other sectors where CuZn alloys are used.	<p>“Several transportation sectors, such as ambulance equipment, are made from Al alloy (Blanco et al., 2022; Vandersluis et al., 2020). According to Hajizadeh et al., Al alloy hardness is between 32-52 HV (Hajizadeh et al., 2017). Therefore, all specimens have hardness still higher than Al alloy.”</p> <p>Thank you.</p>

Reviewer #2:

No.	Comment from Reviewer	Author Response
a	<p>Abstract:</p> <p>Please rewrite your abstract. Currently, it reads too narratively and focuses more on the procedural aspects of the research rather than providing a concise summary of the entire study. The abstract should include the following key points:</p>	<p>Thank you. We appreciate this constructive comment. Generally, we used the IMRaD method to create abstracts. Our responses are as follows:</p>
	<ol style="list-style-type: none"> 1. Purpose of the study: Briefly state the aim of your research and explain why the study was conducted. 2. Methodology: Outline the methods used to address the research problem. 	<ol style="list-style-type: none"> 1. The aim of the research has been added. 2. The methodology has been stated: “Structure, hardness, corrosion, and antibacterial activity behavior were investigated using XRD,

	<ol style="list-style-type: none"> 3. Results: Summarize the key findings of your study. 4. Implications: Discuss the practical and theoretical implications of your results. How can the findings be applied in both theory and practice? 5. Contribution: State the value of your study. What is the unique contribution of your research to the field of study? 	<p>potentiostat, Vickers hardness equipment, and a digital camera.”</p> <ol style="list-style-type: none"> 3. The result of several characterizations such as XRD, potentiostat, Vickers hardness equipment, and a digital camera has been stated. 4. “The Cu₆Zn sample could used as an alternative material in medical equipment for ambulances.” 5. Similar to point 4. <p>Thank you.</p>
b	<p>Introduction:</p> <p>The introduction section needs to be expanded to provide more context and clarity. Please ensure that it includes the following:</p>	<p>Thank you. We appreciate this constructive comment. Generally, an introduction has been revised. Our responses are as follows:</p>
	<ol style="list-style-type: none"> 1. Background: Clearly describe the context of the study and provide an overview of the current state-of-the-art in the field. This section should offer readers an understanding of the existing knowledge and research gaps. 2. Problem Statement: Identify the problem your study addresses. Explain why the problem is significant and why readers should be interested in the research. 3. Proposed Solution: Once the problem is identified, clearly outline the contribution of your work. Highlight the novel aspects of your research and provide a framework that connects it with the existing literature. You should cite at least 8 recent journal articles to support this section. 4. Literature Gaps: Summarize the existing gaps in the literature that your research intends to fill, and present the specific aim of your study. 5. Use of International Sources: Please ensure that you use international journal references, not local sources, to strengthen the relevance of your work. 	<ol style="list-style-type: none"> 1. The background has been enhanced to describe the problem statement clearly, step by step. 2. A problem in the present study is that Staphylococcus aureus was detectable on firefighters' ambulance equipment. This bacterium is methicillin-resistant. 3. The solution to the problem statement is presenting CuZn to kill Staphylococcus aureus. More than 8 journals have been cited. 4. The literature gap is mentioned in the literature review and stated at the end of the introduction. 5. All references used international journals (major from Elsevier). <p>Thank you.</p>
c	<p>Literature Review:</p> <p>While the literature review cofill andvant works, it lacks a critical analysis and synthesis of the literature. It currently reads as</p>	<p>Thank you. We appreciate this constructive comment. Generally, a literature review has been revised. Our responses are as follows:</p>

	a list of studies, but a deeper, more critical evaluation is necessary. Here's how to improve it:	
	<ol style="list-style-type: none"> 1. Critical Analysis: Provide a more comprehensive review of the literature, offering critical commentary and drawing connections between studies to highlight trends, contradictions, or gaps in knowledge. This should not just be a factual listing but a thoughtful analysis of the research. 2. Recent References: Include more recent journal articles (published from 2019-2024) to ensure the literature review reflects the current state of the field. This will also help support the argument for the novelty and relevance of your research. 3. Theoretical Framework: The review should present the theoretical framework underpinning your study. Explain how the selected literature connects with your research questions and objectives. 4. Selection of Literature: Describe how the literature was selected for inclusion. Explain the criteria used to ensure a comprehensive and representative review of the state of the art. 	<ol style="list-style-type: none"> 1. In the literature review, we want to show the reader that CuZn alloy composition was investigated between 2019 and 2024. According to the literature review, the reader could show CuZn with compositions Cu₆Zn, Cu₉Zn, and Cu₁₂Zn have not been investigated, especially for corrosion and bacterial activity. 2. All references cited in the literature review are up to date (2019-2024). 3. The majority of references in the literature review discuss about CuZn casting. 4. All literature reviews were selected for CuZn casting. 5. A literature review was presented to the reader, which found a research gap for our study. <p>Thank you.</p>
d	<p>Methodology:</p> <p>Please clarify the methodology section by addressing the following:</p>	<p>Thank you. We appreciate this constructive comment. Generally, a methodology has been revised and detailed to make the reader easy to understand. Our responses are as follows:</p>
	<ol style="list-style-type: none"> 1. Justification of Methods: Avoid conceptual descriptions and instead explain why each method was chosen for your study. Discuss the rationale behind selecting specific approaches, tools, and techniques. 2. Participants and Characteristics: Provide detailed information about the participants (if applicable) and their characteristics. Who were the subjects, and why were they chosen for this research? 3. Data Collection and Instruments: Describe the data to be collected and the instruments used to gather that data. 	<ol style="list-style-type: none"> 1. The alloy's composition was selected because no one has ever researched it. Other researchers commonly use all apparatus used in the casting process for the present study. 2. The present study conducts experiments; therefore, we don't add any participants outside the research team. 3. The data was collected according to actual measurements using calibrated apparatus and people who are experts in using the tool.

	<p>Include an explanation of how the instruments were applied to ensure reliability and validity in your results.</p> <p>4. Explanation of Images: If any images are included in the methodology, add an explanation of what information can be gleaned from them.</p>	<p>So, there is no need to doubt its truth.</p> <p>4. There is no image in this section.</p> <p>Thank you.</p>
e	Results and Discussion:	
	<p>1. Discussion Structure: The results and discussion sections should be presented in continuous prose, without numbering or bullet points. Ensure that there is a smooth flow between sections, with connecting sentences to guide the reader.</p> <p>2. Comparison with Previous Studies: Your discussion should compare your findings with previous research. Please include at least 15 relevant studies from international journals. This will help contextualize your results and demonstrate how your research fits within the broader body of knowledge.</p> <p>3. Generalizations: Conclude the discussion with a broader perspective, offering generalizations or conclusions based on your findings.</p>	<p>1. The result and discussion have been well arranged.</p> <p>2. The results and discussion compare findings to those of other researchers (more than 15 relevant studies).</p> <p>3. All characterization (XRD) was well connected to hardness, corrosion behavior, and antibacterial activity.</p> <p>Thank you.</p>
f	Conclusion:	
	<p>The conclusion should summarize the main outcomes of the study without using numbered points. It should reflect on the implications of your findings, both theoretical and practical. Highlight the significance of your study and why it contributes to the existing literature.</p>	<p>A conclusion has been revised to be more comprehensive.</p>
g	References:	
	<p>1. Minimum Number of References: Include at least 40 references, with a minimum of 80% sourced from SCOPUS, Web of Science (WoS), or other reputable international journals.</p> <p>2. Citations within the Text: Ensure that all references are properly cited within the text. Avoid listing references without citation. Use a reference management tool like Mendeley to ensure proper formatting and consistency.</p>	<p>1. References cited in this paper are Scopus indexed (majority from Elsevier publisher). We used more than 40 references</p> <p>2. We used Mendeley to make citations (APA ver. 7)</p> <p>3. All references were cited in the introduction, and the literature review is up to date (2019-2024). However, we cited references from the last ten years for the results and discussion section.</p>

	3. Recent Sources: Prioritize recent journal articles (preferably from 2019 onward) to maintain the relevance and currency of your work.	
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EFFECT OF ZINC ADDITION IN COPPER TO STRUCTURE, HARDNESS, CORROSION, AND ANTIBACTERIAL ACTIVITY

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ABSTRACT

Brass (CuZn) is widely used today due to better mechanical, thermal, and chemical properties. The present research fabricated CuZn alloy by adding various Zn (6, 9, and 12 wt.%) to the Cu using gravity casting. Casts CuZn alloy by adding various Zn to the Cu to investigate optimum composition were resulting highest inhibited of bacterial activity. In addition, the structure, hardness, and electrochemical behavior of the alloy were also investigated using XRD, Vickers hardness and potentiostat equipment. XRD confirmed that CuZn alloy has an alpha phase, and a FCC crystal structure. The rise of the Zn content in the alloy led to an increase in crystallite size, a decrease in the hardness and a shift to a more negative OCP potential at 1200 s measurement. Enhancing the Zn content to 9 wt.% in the alloy lead to decrease the corrosion rate. Moreover, 24-hour post-contact observation found that the sample places removed remained clear of bacteria. The Cu6Zn sample successfully inhibited the growth of *Escherichia coli* in the 3rd hour, while *Staphylococcus aureus* was 100 % reduced in the 7th hour. The Cu6Zn sample could be used as an alternative material for medical equipment in ambulances.

Keywords : XRD, Vickers, Electrochemical measurement, *Staphylococcus aureus*, *Escherichia coli*

1. Introduction

Brass (CuZn) is an alloy widely used in the national defense, oil and gas industries, and health because it has better mechanical properties, thermal conductivity, and corrosion resistance (Bhavsar & Bali, 2023; Wang et al., 2023; Widyastuti et al., 2023). CuZn alloys for medical equipment in transportation such as ambulances need to consider two parameters: corrosion resistance and antibacterial characteristics. Commonly NaCl media was used to investigate corrosion for ambulance equipment. This condition due to ambulance equipment commonly exposure from medical patients eccrine sweat and saline-infused (Baker & Wolfe, 2020; Tayyab et al., 2021). Furthermore, some studies found that after cleaning the ambulance, 35.37 % of bacterial contaminants were still seen (Syamsuir et al., 2023).

Viegas et al. found *Staphylococcus aureus* was detectable on firefighter's ambulance equipment (Viegas et al., 2021). *Staphylococcus aureus* is a type of bacteria that could cause skin disease and is hard to treat with traditional antibiotics. *Staphylococcus aureus* bacteria tend to have methicillin resistance. According to Tajik et al., 38.4 % of *Staphylococcus aureus* methicillin resistant was found in Tehran community (Tajik et al., 2020). Moreover, this bacteria also could contaminate orthopedic implants and cause serious infections (Pietrocola et al., 2022).

Several researchers were interested in investigating the corrosion behavior of CuZn alloy in NaCl medium (Abed & Dawood, 2022; Chen et al., 2024; Gao et al., 2021; Yin et al., 2021). Abed and Dawood investigated the corrosion behavior of Cu40Zn alloy in 3.5% NaCl and found a corrosion rate of around 0.037 mmpy (Abed & Dawood, 2022). Yin et al. investigated the corrosion behavior of Cu alloy in NaCl medium were immersed in different times. More time is immersed, resulting in more corrosion resistance of Cu (Yin et al., 2021). Chen et al. investigated Cu alloy in NaCl medium and found Cu potential around -0.305 V vs SCE and Zn potential around -1.165 V vs SCE (Chen et al., 2024). Gao et al. found that a reduction in thickness (50 to 60 %) of CuZn using cold rolling resulted in a significant decrease in corrosion current from 4.824 to 1.804 $\mu\text{A}/\text{cm}^2$ (investigating in 3.5 % NaCl) (Gao et al., 2021). Moreover, aluminum (Al) alloy widely used in transportation sector such as ambulance (Blanco et al., 2022; Vandersluis et al., 2020). Liu et al. have found Al alloy corrosion current between 4.868-5.251 A/cm^2 in a 3.5% NaCl medium (Liu et al., 2020). Comparing the studies of Liu et al. and Gao et al., Al alloy has a higher corrosion current than CuZn (Gao et al., 2021; Liu et al., 2020). Corrosion current significantly influences the corrosion rate, and a rise in the corrosion current would enhance the corrosion rate.

Recently, researchers have been interested in investigating CuZn alloy for medical applications (Azizian et al., 2024; Riaz et al., 2024; Sabbouh et al., 2023). Azizian et al. investigated CuZn alloys microstructure, mechanical properties and cytotoxicity for cardiovascular applications (Azizian et al., 2024). Riaz et al. investigated the structural and biological properties of CuZn alloy for orthopedic applications (Riaz et al., 2024). Moreover, Sabbouh et al. did the sonification of CuZn in an alkali solution to enhance the antibacterial inhibition zone (Sabbouh et al., 2023). Moreover, Syamsuir et al. have investigated the antibacterial activity of *Staphylococcus aureus* by presenting a Cu layer for ambulance equipment (Syamsuir et al., 2023).

The killing mechanism of bacterial activity inseparable from the ions released by the alloy (Qu et al., 2020). Cu^{2+} ions could be adsorbed on the cytoplasmic membrane surfaces, then penetrate the bacteria, react with sulfhydryl groups, and cause the cell to die (Zeng et al., 2022). The released Zn^{2+} ions could penetrate the cell membrane and cause cell death (Du et al., 2021). Zhang et al. have stated that Cu^{2+} and Zn^{2+} ions could act as antibacterial agents and inhibit *Staphylococcus aureus* growth (E. Zhang et al., 2021).

According to the literature review, research on CuZn alloys with Zn compositions in the range of 6–12 wt.% for medical transportation purposes has not been thoroughly investigated. As mentioned above, the killing mechanism of bacterial activity depend on Cu and Zn ions. CuZn alloy can transform into Cu and Zn ions. Therefore, the present research casts CuZn alloy by adding various Zn to the Cu to investigate optimum composition, resulting in a higher killing mechanism of bacterial activity. Moreover, different alloy compositions would result in different electrochemical behavior and mechanical properties. The present study investigated structure, hardness, electrochemical behavior, and antibacterial activity using X-ray diffraction (XRD), Vickers hardness equipment, potentiostat, and digital camera.

2. Literature Review

Many techniques are used to make CuZn alloys, including gravity and investment casting (Hendrawan et al., 2021; Ziat et al., 2020). Gravity casting is simple, inexpensive, and can rapidly fill complex geometry (Huang et al., 2024; Nuryadi et al., 2020). Moreover, in the fabrication of CuZn alloys, one thing needs to be considered to produce specific properties, namely alloy composition. Researchers focused on adding various Zn compositions onto Cu for different purposes. Strzpek et al. investigated the mechanical properties of Cu and alpha brass (Cu2.5Zn and Cu6.5Zn) wire (\varnothing 3.8 mm). Increased Zn content causes increases in ultimate tensile strength, yield strength and hardness (Strzpek et al., 2019). Situmorang et al. fabricated Cu with various Zn additions (10, 20, 38, and 45 wt.%) and found that the higher the Zn composition, the higher the antibacterial properties (Situmorang et al., 2019). Iqbal et al. melted Cu28.7Zn using a furnace and cast to investigate hardness and morphology (Iqbal et al., 2021). Shahriyari et al. added Zn (5, 15, 20, and 30 wt.%), increasing the hardness due to the alloy's rise in the Zn content

(Shahriyari et al., 2022). Akhyar et al. melted Cu_{28.7}Zn using a gas furnace and cast to investigate the tensile strength (Akhyar et al., 2023). Morath et al. have created Zn_{0.8}Cu and Zn_{1.5}Cu using casting methods to investigate the biological aspect of arterial implants (Morath et al., 2024). Azizian et al. have added Cu with compositions 1, 2, and 5 wt.% to CuZn alloy by melting in the induction furnace to investigate microstructure, mechanical properties, and cytotoxicity for cardiovascular application (Azizian et al., 2024). Generally, a CuZn alloy with a Zn composition of less than 37 wt.% would produce a single alpha phase with an FCC crystal structure (Clement & Auger, 2023; Mousavi et al., 2020).

3. Research Methods

3.1 Material Preparation

Cu ingot (98.798 %) and Zn powder (99 %) were melted and cast with the alloy composition Cu_xZn (x=0, 6, 9, and 12 wt.%, namely as Cu, Cu₆Zn, Cu₉Zn, and Cu₁₂Zn, respectively) and then confirmed the formed alloy using XRF (Table 1). Before melting was conducted, the ingot and the apparatus, such as the crucible, were cleaned using water to avoid impurities and then dried. The Cu ingot was first filled into a silicon carbide crucible (3kg) and then inserted into a muffle furnace. Melting was carried out in a crucible at 1100 °C under atmospheric pressure. After the Cu has melted, remove it from the muffle furnace, mix it with Zn powder, stir it manually, and pour it into a permanent mold. For comparison, Cu ingot was melted and poured into a permanent mold without Zn addition. The as-cast ingots were cut for further characterization, such as XRD, hardness, electrochemical measurement, and antibacterial activity observation.

Table 1 – Chemical composition of various casting samples.

Sample	Element (wt.%)					
	Cu	Zn	Al	Si	P	Fe
Cu	98.798	-	0.135	0.444	0.384	0.238
Cu ₆ Zn	92.308	6.384	0.135	0.444	0.384	0.345
Cu ₉ Zn	89.228	9.497	0.135	0.444	0.384	0.312
Cu ₁₂ Zn	87.075	11.557	0.135	0.444	0.384	0.405

3.2 XRD Measurement

XRD was measured using the PANalytical (Cu K α 1 λ =1.5405980) apparatus. XRD was scanned from 20 to 100°, using step size 0.0217°. The Highscore software was used to refine and collect peak, phase and crystallographic parameters of as-cast samples. By using that software, full width at half maximum (FWHM) are also found. FWHM data is used to calculate crystallite size.

3.3 Hardness Measurement

Before testing, samples with 20×20×6 mm dimensions were polished using silicon carbide up to #3000 grit. Afterward, the polished sample was cleaned using water, followed by alcohol, and then dried using drier equipment. The hardness test was conducted using the Vickers method. An FV-300e hardness test was performed on top of various samples using 1 kg of load. Ten repeatable measurements were conducted.

3.4. Electrochemical Measurement

Two electrochemical measurements were conducted in the present research, such as open circuit potential (OCP) and Linear sweep voltammetry (LSV), using Digi-Ivy (DY2311) potentiostat in 0.9 % NaCl at room temperature. OCP was scanned until 1200 s using a sampling scan rate of 0.02 s, while LSV was conducted using a scan rate of 1 mV/s. Cu/CuZn samples are used as the working electrode, platinum wire as the counter electrode, and Ag/AgCl as the reference electrode. LSV data was examined using the Tafel extrapolation method to see corrosion potential (E_{corr}) and current density (i_{corr}). The corrosion rate could be found by inserting i_{corr} in the following equation (Soegijono et al., 2020).

$$\text{Corrosion rate (mmpy)} = C \frac{M \times i_{\text{corr}}}{\rho \times n} \quad (1)$$

Where C is corrosion constant (3.27 mmpy), M is atomic weight (g/mol), ρ is material density (g/cm³), and n is the number of electrons involved.

3.5 Antibacterial Activity Observation

The sample dimension used for antibacterial activity is 20×20×6 mm. Before testing, samples were polished using silicon carbide up to #3000 grit. The experimental procedure for antibacterial activity is similar to that of the previous report (Syamsuir et al., 2023). Moreover, the recent study uses *Staphylococcus aureus* ATCC 25923 for Direct contact kill assay (24 hours) and Fluid contact assay (8 hours). In comparison, *Escherichia coli* 25922 was also used in the present study. Afterward, documentation was captured using a digital camera. In addition, 24-hour post-contact observation and Fluid contact test were also captured using a digital camera.

4. Results and Discussions

4.1 XRD

The diffraction pattern of Cu/CuZn with (111), (200), (311), and (222) planes can be seen in Figure 1. The four diffraction patterns match the alpha phase, which is shown to align with another study (Heidarzadeh et al., 2022). The acquired XRD data was then analyzed using Highscore software, and the parameters are listed in Table 2. All samples had a face-centered cubic (FCC) crystal structure, indicating that the Cu and Zn atoms dissolve into one another. According to Jinlong et al., the FCC quantity of surface energy is (111) > (001) > (110), and the FCC sample with the (111) plane has the lowest corrosion rate (Jinlong et al., 2016). Moreover, the FCC sample with the preferred orientation of the (111) plane has the highest surface atomic density (Soegijono et al., 2020).

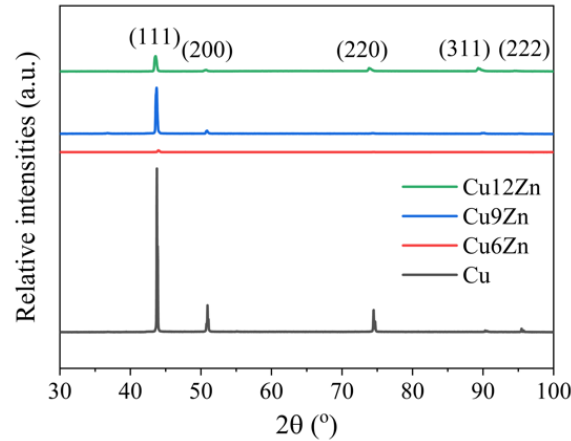


Fig. 1. XRD of various casting samples.

Table 2 - Crystallographic parameters of various casting samples.

Parameter	Sample			
	Cu	Cu6Zn	Cu9Zn	Cu12Zn
Crystal structure	FCC			
Lattice constant a = b = c (Å)	2.964	8.929	2.981	2.964
Cell volume (Å ³)	26.03	90	26.49	26.05
d-spacing (Å)	1.69	1.18	1.69	2.07
Crystallite Size (nm)	227.20	109.95	204.95	316.26
Micro strain	0.37	0.53	0.16	0.32

Presenting Zn in the alloy reduces crystallite size from 227.20 to 109.95 nm and increases Zn content from 6 to 12 wt.%, leading to an increase in crystallite size from 109.95 to 316.26 nm.

This behavior is similar to Özdemir and Karahan's study that showed Zn in the alloy leads to decreased crystallite size, and an increase in Zn content in the alloy leads to increased crystallite size (Özdemir & Karahan, 2014). Moreover, the microstrains of the as-cast sample are independent of Zn content, which perfectly agrees with Karahan and Özdemir's study ((Karahan & Özdemir, 2014). **The smallest microstrain is seen in the Cu9Zn sample.**

4.2 Hardness

Figure 2 shows the average hardness of various casting samples. Nikhil et al. have found that pure Cu has a hardness of 140 HV when treated at 400 and 600 °C and then held for two hours, followed by quenching in tap water, resulting in a hardness of 100 and 60 HV (Nikhil et al., 2021). The higher heat treatment temperatures led to a decrease in the hardness of the pure Cu. Therefore, the hardness of Cu may vary depending on heat treatment. In the present study, Cu re-casting has a hardness of 74.54 HV.

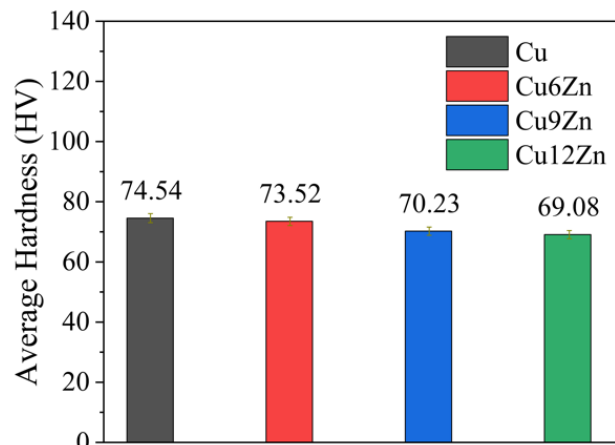


Fig. 2. Average hardness of various casting samples.

According to Figure 2, it can be seen that an increase in Zn content led to a decrease in the hardness. Another research study found that Cu (as-cast) has a hardness of around 100 HV, while Cu15 Zn (as-cast) has a hardness of 75 HV (Ezequiel et al., 2024). Nnakwo et al. also found that increased Zn content in the Cu alloy leads to decreased hardness due to increased in grain size and solid solution region (Nnakwo et al., 2021). According to a study by Qu et al. and García-Mintegui et al., pure Zn hardness is between 41-42 HV (García-Mintegui et al., 2021; Qu et al., 2020). Therefore, it could be concluded that Zn in the alloy leads to decreased hardness due to Zn hardness less than Cu.

Several researchers correlated measured hardness to crystallite size (Augustin et al., 2016; Syamsuir et al., 2023). Augustin et al. have found an increase in Cu's crystallite size, promoting a decrease in scratch and micro-hardness (Augustin et al., 2016). Syamsuir et al. found a decrease in Cu's crystallite size, leading to an increase in hardness (Syamsuir et al., 2023). Comparing Table 2 with Figure 2, it can be seen that an increase in the Zn content led to an increase in crystallite size and a decrease in the hardness. On the contrary, while as-cast samples do not form an alloy (Cu), the resulting hardness is not aligned with the crystallite size. It seems that it cannot compare the crystallite size were found with measured hardness between alloy (CuZn) and un-alloy (Cu) material.

Several transportation sectors, such as ambulance equipment, are made from Al alloy (Blanco et al., 2022; Vandersluis et al., 2020). According to Hajizadeh et al., Al alloy hardness is between 32-52 HV (Hajizadeh et al., 2017). Therefore, all specimens have hardness still higher than Al alloy.

4.3 OCP

Figure 3 shows the OCP measurement result of various casting samples in 0.9 % NaCl at room temperature. Generally, increased Zn content in the alloy promoted more negative potential,

which perfectly agrees with the Cocco et al. study (Cocco et al., 2016). Dridi et al. have found that E_{OCP} CuZn30 and CuZn39 are -0.578 and -0.604 V/MSE at 3 % NaCl, which means an increase in the Zn promoted to more negative potential (Dridi et al., 2020). Cu, Cu6Zn, Cu9Zn, and Cu12Zn samples have E_{OCP} potential at 1200 s measurement -0.014, -0.023, -0.027, and -0.032 V vs Ag/AgCl, respectively.

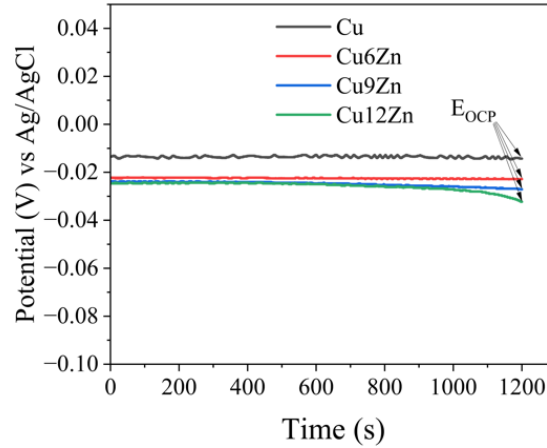


Fig. 3. OCP measurement of various casting samples.

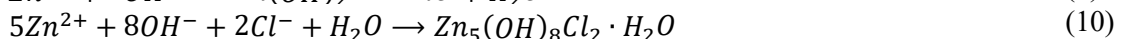
According to Figure 3, Cu, Cu6Zn, and Cu9Zn samples are steady at initial measurements until 1200 s, indicating that the protective layer formed has good protection. In contrast, the Cu12Zn sample is steady at initial measurements until 600 s, then moves in a more negative direction. These phenomena indicated that the formed protective layer had initially dissolved at 600 s; therefore, the measurement continuously moved forward in a negative direction until the measurement reached 1200 s.

4.4 LSV

Eccrine sweat and saline-infused for humans are nearly 0.9 % of NaCl (Bond & Lieu, 2014; Tayyab et al., 2021). Therefore, LSV measurement was conducted in 0.9 % NaCl at room temperature. Luo et al. found Cu₂O crystalline growth on the Cu surface when exposed to 0.9 % NaCl, while when exposed to pure water, Cu₂O crystalline was not seen (Luo et al., 2020). Commonly, Cu₂O crystallines are formed, which is preceded by the formation of CuCl when the specimen is tested in a chloride solution. Moreover, Zhang et al., in their study, found ZnO and Zn₅Cl₂(OH)₈·H₂O as corrosion products on top of Cu40Zn surfaces in a chloride environment (X. Zhang et al., 2016). The reaction of Cu in chloride solution is as follows (Milošev et al., 2024).



The reaction of Zn in chloride solution is as follows (Milošev et al., 2024).



LSV measurement results in 0.9 % NaCl can be seen in Figure 4.

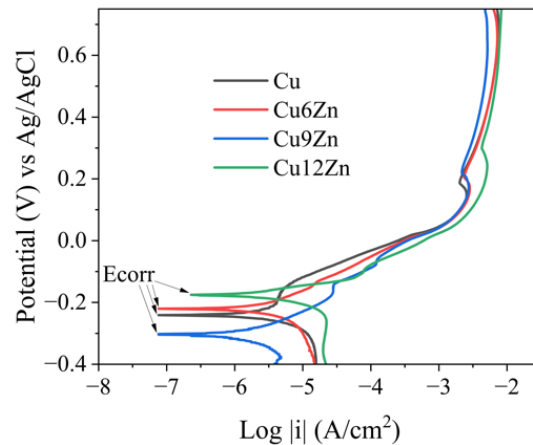


Fig. 4. LSV scans result of various casting samples.

According to Figure 4, corrosion E_{corr} and i_{corr} can be found using the Tafel extrapolation method. Moreover, the corrosion rate could be calculated by inserting i_{corr} into expression (1). Table 3 presented E_{corr} , i_{corr} , and the corrosion rate of various casting samples. It appears E_{corr} is independent of Zn addition; however, Zn is dependent on i_{corr} and corrosion rate (except for the Cu12Zn sample). This behavior is probably due to a protective layer that was formed. Compared to the OCP result, it can be seen that the Cu12Zn sample continuously moves forward in a negative direction from 600 until 1200 s of measurement. Therefore, that sample has a higher i_{corr} and corrosion rate.

Table 3 - Corrosion parameters of various casting samples.

Sample name	E_{corr} (V) vs Ag/AgCl	i_{corr} (A/cm ²)	Corrosion rate (mmpy)
Cu	-0.241	4.42×10^{-6}	5.13×10^{-2}
Cu6Zn	-0.220	3.33×10^{-6}	3.86×10^{-2}
Cu9Zn	-0.304	2.15×10^{-6}	2.49×10^{-2}
Cu12Zn	-0.175	6.19×10^{-6}	7.18×10^{-2}

Qu et al. have found that an increase in Zn content led to an increase in corrosion resistance, which perfectly agrees with the present study (except for Cu12Zn) (Qu et al., 2020). Milošev et al. investigated Cu, Cu10Zn, Cu40Zn, and Zn in 3 % NaCl and found i_{corr} after stabilized at 1 hour around 1.573, 1.456, and 2.114, and 5.21 $\mu\text{A}/\text{cm}^2$ respectively (Milošev et al., 2024). According to equation (1), i_{corr} strongly influences the corrosion rate. The more i_{corr} , the higher the corrosion rate. Moreover, a limitation in Zn content in the Cu alloy could influence the corrosion resistance. Presenting the Zn content ≤ 11 wt.% in the alloy could enhance the corrosion resistance; however, Zn of more than 10 wt.% could decrease corrosion resistance (Milošev et al., 2024).

According to Table 2, the Cu9Zn sample has the lowest microstrain than others. The measured microstrain could be associated with the sample's crystal defect (Soegijono et al., 2020). Based on Table 2, the Cu9Zn sample has the lowest microstrain, which confirms that the sample has the lowest corrosion rate. Moreover, the FCC sample with the preferred orientation of the (111) plane could offer a lower corrosion rate due to the highest surface atomic density (Jinlong et al., 2016; Soegijono et al., 2020). Compared to other samples, the Cu9Zn sample has the higher preferred orientation of the (111) plane. Even though the (111) plane of the Cu sample is the highest. Unfortunately, the (220) and (200) planes are still present and relatively high.

4.5 Antibacterial Activity

Figure 5 shows the direct contact kill of *Staphylococcus aureus* and *Escherichia coli* after 24 hours of incubation. The present study focused on *Staphylococcus aureus*, but *Escherichia coli* was also used for comparison. There is no diffusion in the sample; therefore, the inhibition zone could not be seen.

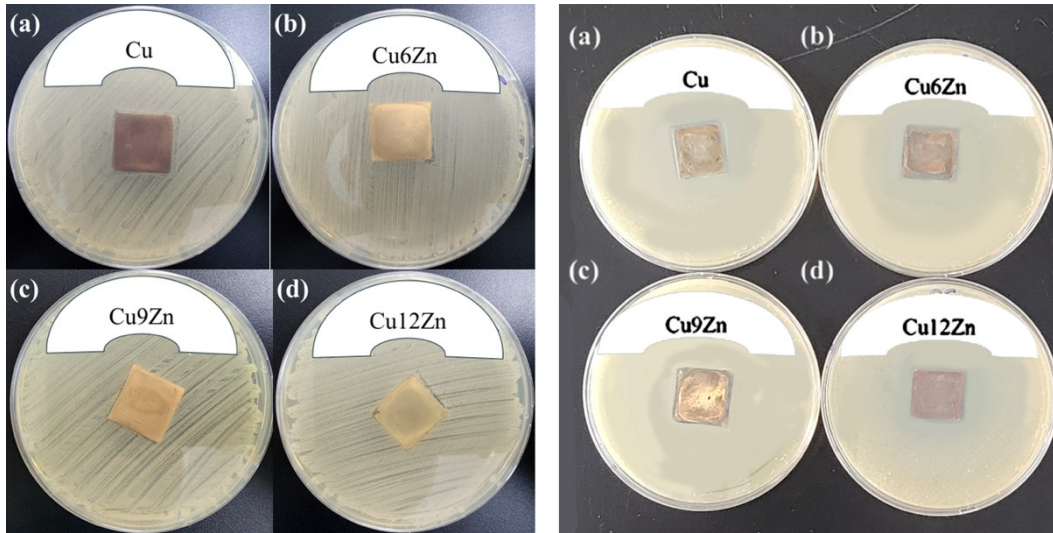


Fig. 5. Antibacterial activity test using *Staphylococcus aureus* (left) and *Escherichia coli* (right) (a) Cu, (b) Cu6Zn, (c) Cu9Zn, and (d) Cu12Zn.

Antibacterial activity after 24 hours of post-contact with various casting samples using *Staphylococcus aureus* and *Escherichia coli* can be seen in Figure 6. The removed sample places remain clear (with no regrowth) from bacterial activity. The antibacterial behavior was significantly influenced by Cu or Zn ions (Qu et al., 2020). Villapún et al. found that releasing Cu ions leads to the highest killing activity of *Staphylococcus aureus* (Villapún et al., 2016). Excess in the Cu ion could be bacteriostatic (Sabbouh et al., 2023). Moreover, Cu ions could be adsorbed on the cytoplasmic membrane surfaces, then penetrate the bacteria, react with sulfhydryl groups and cause the cell to die (Zeng et al., 2022). Furthermore, Cu ions could form hydroxyl groups in the presence of oxygen in nature, which could destroy cell membranes (Dou et al., 2022). Hutchings et al. stated that Zn^{2+} successfully inhibits the growth of *S. epidermidis* (Hutchings et al., 2021). This behavior is associated with the generation of reactive oxygen or the formation of (E. Zhang et al., 2021).

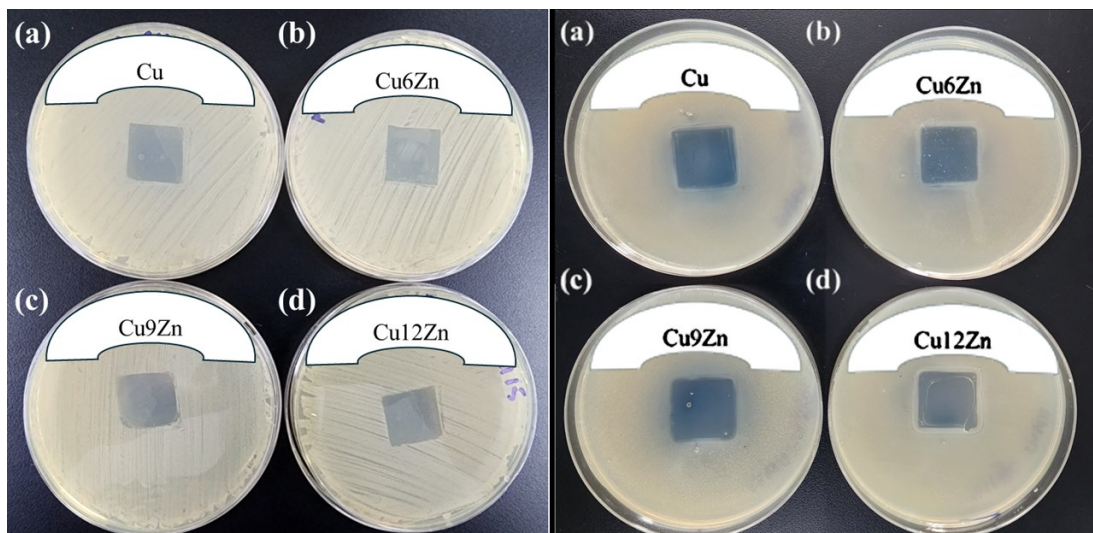


Fig. 6. Antibacterial activity after 24 hours of post-contact (regrowth assessment) towards *Staphylococcus aureus* (left) and *Escherichia coli* (right) (a) Cu, (b) Cu6Zn, (c) Cu9Zn, and (d) Cu12Zn

Figure 7 shows the fluid contact test of *Staphylococcus aureus* and *Escherichia coli*. The orientation of the test materials is mapped within the yellow box. For the Cu6Zn sample, *Escherichia coli* was killed on the 3rd hour. However, there is no significant reduction within the

fluid because there are no diffusible materials. Also, there is no visible growth after 3rd hour for the fluid in contact with the metal.

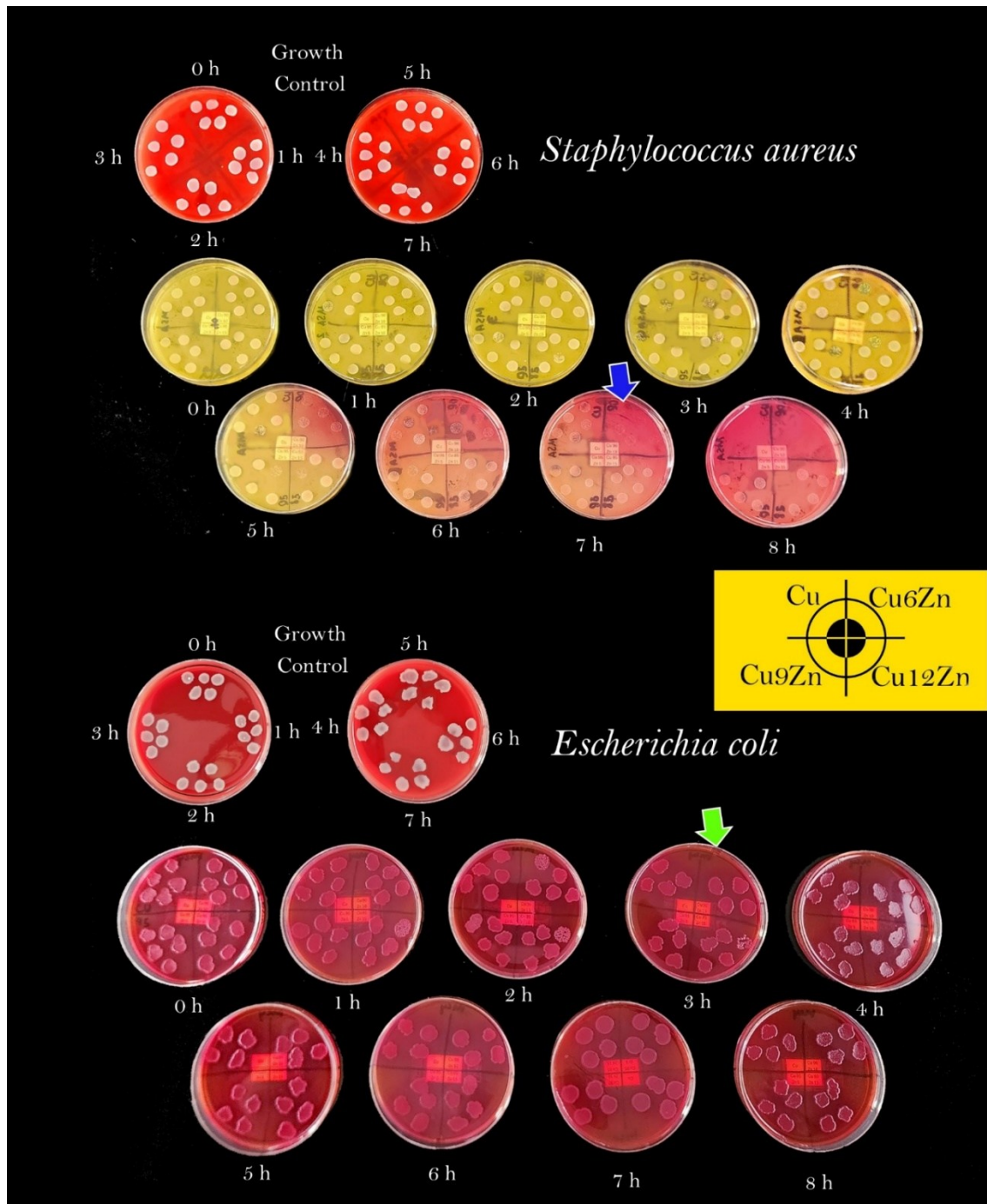


Fig. 7. Fluid contact test of *Staphylococcus aureus* and *Escherichia coli*

Moreover, it should be noted that the reduction of the colony is significant in the 7th hour for *Staphylococcus aureus* (blue arrow), but for *Escherichia coli*, the inhibition of *Escherichia coli* growth is shown within the 3rd hour of contact (green arrow), for Cu6Zn sample. The reduction of colonies is significant on the surface of the metal. While on the remaining fluid, the reduction is insignificant until 8 hours. This behavior is because *Staphylococcus aureus* and *Escherichia coli* have different membrane structures and thick cell walls, therefore could inhibit ion exchange and restrain the antibacterial effect of Cu and or Zn ions (Di et al., 2022). Cu killing is more effective in Gram-negative bacteria (e.g., *Escherichia coli*) because peptidoglycan affects the cell's susceptibility. The thicker the peptidoglycan layer, the harder it became for the Cu ions

to reach the membrane (Soltani et al., 2020; Xhafa et al., 2023). Therefore, *Escherichia coli* was killed in the 3rd hour. Another reason Cu6Zn has better antibacterial performance than others is probably due to its smaller crystallite size. Researchers found that smaller crystallite sizes promote the enhancement of antibacterial effects (Syamsuir et al., 2023). This behavior is attributed to an increase in surface area due to the crystallite size (Sangeetha et al., 2015). The Cu6Zn sample could be used as an alternative material for medical equipment in ambulances.

5. Conclusion

CuZn has been successfully fabricated. XRD confirmed that CuZn alloy has a single alpha phase with an FCC crystal structure. The rise of the Zn content in the alloy led to a decrease in the hardness due to an increase in crystallite size and led to a shift to more negative OCP potential at 1200 s measurement. Moreover, the rise of the Zn content to 9 wt.% decreased the corrosion rate. It appears there is a limitation in Zn content in the copper alloy that influences the corrosion rate, as shown when Zn content around 12 wt. % is promoted to increase the corrosion rate. Antibacterial activity observation found that all samples had no diffusion. Moreover, 24-hour post-contact observation found that sample places removed from the sample remained clear of bacteria. The Cu6Zn has better antibacterial performance than others due to the smallest crystallite size. According to the fluid contact test, the reduction of the colony of *Staphylococcus aureus* is significant in the 7th hour. The inhibition of *Escherichia coli* growth is also shown within the 3rd hour of contact. This behavior is because *Staphylococcus aureus* and *Escherichia coli* have different membrane structures and thick cell walls, therefore, could inhibit ion exchange and restrain the antibacterial effect of Cu and or Zn ions.

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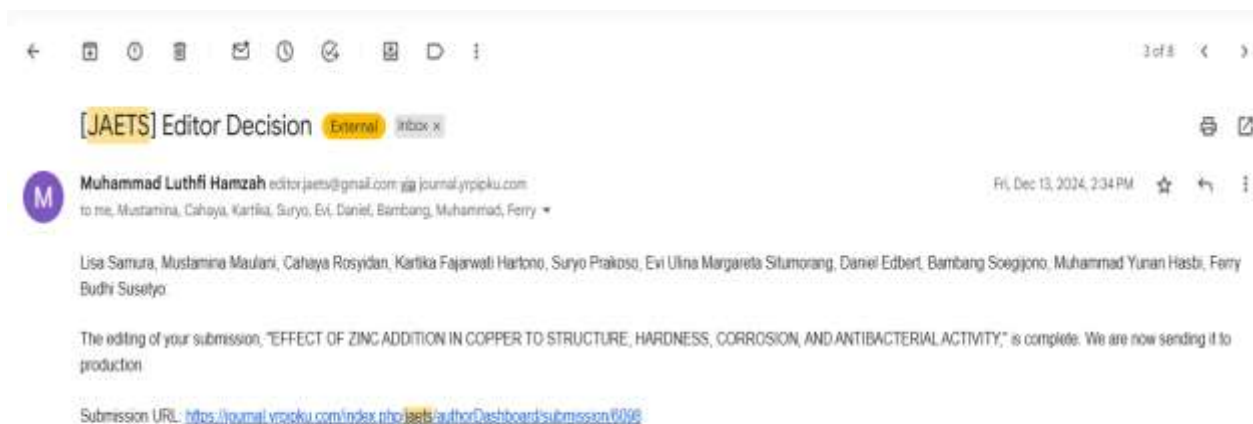
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4. BUKTI PROSES PRODUCTION

(13 Desember 2024)

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5. BUKTI PUBLISH

(Desember 2024)

EFFECT OF ZINC ADDITION IN COPPER TO STRUCTURE, HARDNESS, CORROSION, AND ANTIBACTERIAL ACTIVITY

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ABSTRACT

Brass (CuZn) is widely used today due to better mechanical, thermal, and chemical properties. The present research fabricated CuZn alloy by adding various Zn (6, 9, and 12 wt.%) to the Cu using gravity casting. Casts CuZn alloy by adding various Zn to the Cu to investigate optimum composition were resulting highest inhibited of bacterial activity. In addition, the structure, hardness, and electrochemical behavior of the alloy were also investigated using XRD, Vickers hardness, and potentiostat equipment. XRD confirmed that CuZn alloy has an alpha phase, and a FCC crystal structure. The rise of the Zn content in the alloy led to an increase in crystallite size, a decrease in the hardness and a shift to a more negative OCP potential at 1200 s measurement. Enhancing the Zn content to 9 wt.% in the alloy lead to decrease the corrosion rate. Moreover, 24-hour post-contact observation found that the sample places removed remained clear of bacteria. The Cu6Zn sample successfully inhibited the growth of *Escherichia coli* in the 3rd hour, while *Staphylococcus aureus* was 100 % reduced in the 7th hour. The Cu6Zn sample could be used as an alternative material for medical equipment in ambulances.

Keywords: XRD, Vickers, Electrochemical Measurement, *Staphylococcus Aureus*, *Escherichia Coli*

1. Introduction

Brass (CuZn) is an alloy widely used in the national defense, oil and gas industries, and health because it has better mechanical properties, thermal conductivity, and corrosion resistance (Bhavsar & Bali, 2023; Wang et al., 2023; Widyastuti et al., 2023). CuZn alloys for medical equipment in transportation such as ambulances need to consider two parameters: corrosion resistance and antibacterial characteristics. Commonly NaCl media was used to investigate corrosion for ambulance equipment. This condition due to ambulance equipment commonly exposure from medical patients eccrine sweat and saline-infused (Baker & Wolfe, 2020; Tayyab et al., 2021). Furthermore, some studies found that after cleaning the ambulance, 35.37 % of bacterial contaminants were still seen (Syamsuir et al., 2023).

Viegas et al. found *Staphylococcus aureus* was detectable on firefighter's ambulance equipment (Viegas et al., 2021). *Staphylococcus aureus* is a type of bacteria that could cause skin disease and is hard to treat with traditional antibiotics. *Staphylococcus aureus* bacteria tend to have methicillin resistance. According to Tajik et al., 38.4 % of *Staphylococcus aureus* methicillin resistant was found in Tehran community (Tajik et al., 2020). Moreover, this bacteria also could contaminate orthopedic implants and cause serious infections (Pietrocola et al., 2022).

Several researchers were interested in investigating the corrosion behavior of CuZn alloy in NaCl medium (Abed & Dawood, 2022; Chen et al., 2024; Gao et al., 2021; Yin et al., 2021). Abed and Dawood investigated the corrosion behavior of Cu40Zn alloy in 3.5% NaCl and found a corrosion rate of around 0.037 mmpy (Abed & Dawood, 2022). Yin et al. investigated the corrosion behavior of Cu alloy in NaCl medium were immersed in different times. More

time is immersed, resulting in more corrosion resistance of Cu (Yin et al., 2021). Chen et al. investigated Cu alloy in NaCl medium and found Cu potential around -0.305 V vs SCE and Zn potential around -1.165 V vs SCE (Chen et al., 2024). Gao et al. found that a reduction in thickness (50 to 60 %) of CuZn using cold rolling resulted in a significant decrease in corrosion current from 4.824 to 1.804 $\mu\text{A}/\text{cm}^2$ (investigating in 3.5 % NaCl) (Gao et al., 2021). Moreover, aluminum (Al) alloy widely used in transportation sector such as ambulance (Blanco et al., 2022; Vandersluis et al., 2020). Liu et al. have found Al alloy corrosion current between 4.868-5.251 A/cm^2 in a 3.5% NaCl medium (Liu et al., 2020). Comparing the studies of Liu et al. and Gao et al., Al alloy has a higher corrosion current than CuZn (Gao et al., 2021; Liu et al., 2020). Corrosion current significantly influences the corrosion rate, and a rise in the corrosion current would enhance the corrosion rate.

Recently, researchers have been interested in investigating CuZn alloy for medical applications (Azizian et al., 2024; Riaz et al., 2024; Sabbouh et al., 2023). Azizian et al. investigated CuZn alloys microstructure, mechanical properties and cytotoxicity for cardiovascular applications (Azizian et al., 2024). Riaz et al. investigated the structural and biological properties of CuZn alloy for orthopedic applications (Riaz et al., 2024). Moreover, Sabbouh et al. did the sonification of CuZn in an alkali solution to enhance the antibacterial inhibition zone (Sabbouh et al., 2023). Moreover, Syamsuir et al. have investigated the antibacterial activity of *Staphylococcus aureus* by presenting a Cu layer for ambulance equipment (Syamsuir et al., 2023).

The killing mechanism of bacterial activity inseparable from the ions released by the alloy (Qu et al., 2020). Cu^{2+} ions could be adsorbed on the cytoplasmic membrane surfaces, then penetrate the bacteria, react with sulfhydryl groups, and cause the cell to die (Zeng et al., 2022). The released Zn^{2+} ions could penetrate the cell membrane and cause cell death (Du et al., 2021). Zhang et al. have stated that Cu^{2+} and Zn^{2+} ions could act as antibacterial agents and inhibit *Staphylococcus aureus* growth (Zhang et al., 2021).

According to the literature review, research on CuZn alloys with Zn compositions in the range of 6–12 wt.% for medical transportation purposes has not been thoroughly investigated. As mentioned above, the killing mechanism of bacterial activity depend on Cu and Zn ions. CuZn alloy can transform into Cu and Zn ions. Therefore, the present research casts CuZn alloy by adding various Zn to the Cu to investigate optimum composition, resulting in a higher killing mechanism of bacterial activity. Moreover, different alloy compositions would result in different electrochemical behavior and mechanical properties. The present study investigated structure, hardness, electrochemical behavior, and antibacterial activity using X-ray diffraction (XRD), Vickers hardness equipment, potentiostat, and digital camera.

2. Literature Review

Many techniques are used to make CuZn alloys, including gravity and investment casting (Hendrawan et al., 2021; Ziat et al., 2020). Gravity casting is simple, inexpensive, and can rapidly fill complex geometry (Huang et al., 2024; Nuryadi et al., 2020). Moreover, in the fabrication of CuZn alloys, one thing needs to be considered to produce specific properties, namely alloy composition. Researchers focused on adding various Zn compositions onto Cu for different purposes. Strzpek et al. investigated the mechanical properties of Cu and alpha brass (Cu2.5Zn and Cu6.5Zn) wire (\varnothing 3.8 mm). Increased Zn content causes increases in ultimate tensile strength, yield strength and hardness (Strzpek et al., 2019). Situmorang et al. fabricated Cu with various Zn additions (10, 20, 38, and 45 wt.%) and found that the higher the Zn composition, the higher the antibacterial properties (Situmorang et al., 2019). Iqbal et al. melted Cu28.7Zn using a furnace and cast to investigate hardness and morphology (Iqbal et al., 2021). Shahriyari et al. added Zn (5, 15, 20, and 30 wt.%), increasing the hardness due to the alloy's rise in the Zn content (Shahriyari et al., 2022). Akhyar et al. melted Cu28.7Zn using a gas furnace and cast to investigate the tensile strength (Akhyar et al., 2023). Morath et al. have created Zn0.8Cu and Zn1.5Cu using casting methods to investigate the biological aspect of arterial implants (Morath et al., 2024). Azizian et al. have added Cu with compositions 1, 2, and 5 wt.% to CuZn alloy by melting in the induction furnace to investigate microstructure,

mechanical properties, and cytotoxicity for cardiovascular application (Azizian et al., 2024). Generally, a CuZn alloy with a Zn composition of less than 37 wt.% would produce a single alpha phase with an FCC crystal structure (Clement & Auger, 2023; Mousavi et al., 2020).

3. Research Methods

3.1 Material Preparation

Cu ingot (98.798 %) and Zn powder (99 %) were melted and cast with the alloy composition Cu_xZn ($x=0, 6, 9$, and 12 wt.%, namely as Cu, Cu6Zn, Cu9Zn, and Cu12Zn, respectively) and then confirmed the formed alloy using XRF (Table 1). Before melting was conducted, the ingot and the apparatus, such as the crucible, were cleaned using water to avoid impurities and then dried. The Cu ingot was first filled into a silicon carbide crucible (3kg) and then inserted into a muffle furnace. Melting was carried out in a crucible at 1100°C under atmospheric pressure. After the Cu has melted, remove it from the muffle furnace, mix it with Zn powder, stir it manually, and pour it into a permanent mold. For comparison, Cu ingot was melted and poured into a permanent mold without Zn addition. The as-cast ingots were cut for further characterization, such as XRD, hardness, electrochemical measurement, and antibacterial activity observation.

Table 1 – Chemical composition of various casting samples.

Sample	Element (wt.%)					
	Cu	Zn	Al	Si	P	Fe
Cu	98.798	-	0.135	0.444	0.384	0.238
Cu6Zn	92.308	6.384	0.135	0.444	0.384	0.345
Cu9Zn	89.228	9.497	0.135	0.444	0.384	0.312
Cu12Zn	87.075	11.557	0.135	0.444	0.384	0.405

3.2 XRD Measurement

XRD was measured using the PANalytical (Cu $K_{\alpha 1}$ $\lambda=1.5405980$) apparatus. XRD was scanned from 20 to 100° , using step size 0.0217° . The Highscore software was used to refine and collect peak, phase and crystallographic parameters of as-cast samples. By using that software, full width at half maximum (FWHM) are also found. FWHM data is used to calculate crystallite size.

3.3 Hardness Measurement

Before testing, samples with $20 \times 20 \times 6$ mm dimensions were polished using silicon carbide up to #3000 grit. Afterward, the polished sample was cleaned using water, followed by alcohol, and then dried using drier equipment. The hardness test was conducted using the Vickers method. An FV-300e hardness test was performed on top of various samples using 1 kg of load. Ten repeatable measurements were conducted.

3.4. Electrochemical Measurement

Two electrochemical measurements were conducted in the present research, such as open circuit potential (OCP) and Linear sweep voltammetry (LSV), using Digi-Ivy (DY2311) potentiostat in 0.9 % NaCl at room temperature. OCP was scanned until 1200 s using a sampling scan rate of 0.02 s, while LSV was conducted using a scan rate of 1 mV/s. Cu/CuZn samples are used as the working electrode, platinum wire as the counter electrode, and Ag/AgCl as the reference electrode. LSV data was examined using the Tafel extrapolation method to see corrosion potential (E_{corr}) and current density (i_{corr}). The corrosion rate could be found by inserting i_{corr} in the following equation (Soegijono et al., 2020).

$$\text{Corrosion rate (mmpy)} = C \frac{M \times i_{\text{corr}}}{\rho \times n} \quad (1)$$

Where C is corrosion constant (3.27 mmpy), M is atomic weight (g/mol), ρ is material density (g/cm^3), and n is the number of electrons involved.

3.5 Antibacterial Activity Observation

The sample dimension used for antibacterial activity is 20×20×6 mm. Before testing, samples were polished using silicon carbide up to #3000 grit. The experimental procedure for antibacterial activity is similar to that of the previous report (Syamsuir et al., 2023). Moreover, the recent study uses *Staphylococcus aureus* ATCC 25923 for Direct contact kill assay (24 hours) and Fluid contact assay (8 hours). In comparison, *Escherichia coli* 25922 was also used in the present study. Afterward, documentation was captured using a digital camera. In addition, 24-hour post-contact observation and Fluid contact test were also captured using a digital camera.

4. Results and Discussions

4.1 XRD

The diffraction pattern of Cu/CuZn with (111), (200), (311), and (222) planes can be seen in Figure 1. The four diffraction patterns match the alpha phase, which is shown to align with another study (Heidarzadeh et al., 2022). The acquired XRD data was then analyzed using Highscore software, and the parameters are listed in Table 2. All samples had a face-centered cubic (FCC) crystal structure, indicating that the Cu and Zn atoms dissolve into one another. According to Jinlong et al., the FCC quantity of surface energy is (111)>(001) >(110), and the FCC sample with the (111) plane has the lowest corrosion rate (Jinlong et al., 2016). Moreover, the FCC sample with the preferred orientation of the (111) plane has the highest surface atomic density (Soegijono et al., 2020).

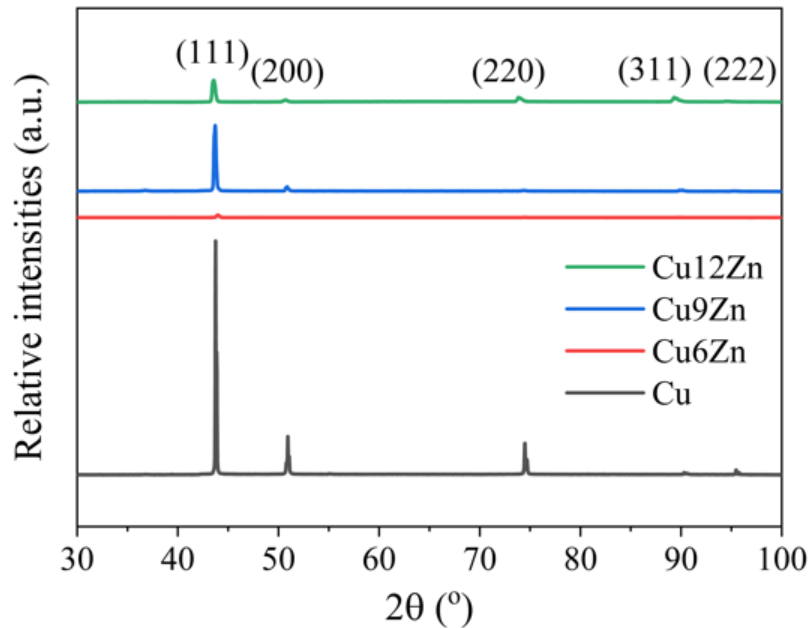


Fig. 1. XRD of various casting samples.

Table 2 - Crystallographic parameters of various casting samples.

Parameter	Sample			
	Cu	Cu6Zn	Cu9Zn	Cu12Zn
Crystal structure			FCC	
Lattice constant $a = b = c$ (Å)	2.964	8.929	2.981	2.964
Cell volume (Å ³)	26.03	90	26.49	26.05
d-spacing (Å)	1.69	1.18	1.69	2.07
Crystallite Size (nm)	227.20	109.95	204.95	316.26
Micro strain	0.37	0.53	0.16	0.32

Presenting Zn in the alloy reduces crystallite size from 227.20 to 109.95 nm and increases Zn content from 6 to 12 wt.%, leading to an increase in crystallite size from 109.95 to 316.26 nm. This behavior is similar to Özdemir and Karahan's study that showed Zn in the alloy leads to decreased crystallite size, and an increase in Zn content in the alloy leads to increased

crystallite size (Özdemir & Karahan, 2014). Moreover, the microstrains of the as-cast sample are independent of Zn content, which perfectly agrees with Karahan and Özdemir's study ((Karahan & Özdemir, 2014). The smallest microstrain is seen in the Cu9Zn sample.

4.2 Hardness

Figure 2 shows the average hardness of various casting samples. Nikhil et al. have found that pure Cu has a hardness of 140 HV when treated at 400 and 600 °C and then held for two hours, followed by quenching in tap water, resulting in a hardness of 100 and 60 HV (Nikhil et al., 2021). The higher heat treatment temperatures led to a decrease in the hardness of the pure Cu. Therefore, the hardness of Cu may vary depending on heat treatment. In the present study, Cu re-casting has a hardness of 74.54 HV.

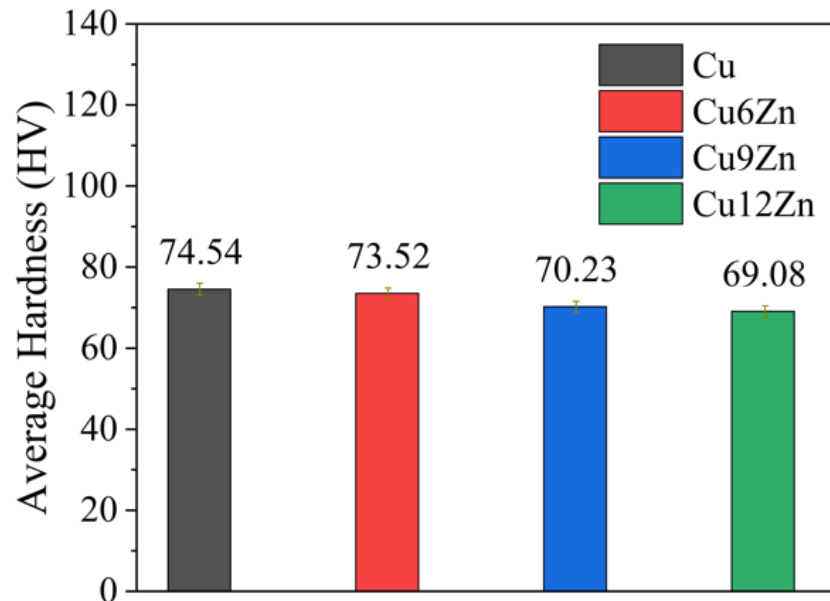


Fig. 2. Average hardness of various casting samples.

According to Figure 2, it can be seen that an increase in Zn content led to a decrease in the hardness. Another research study found that Cu (as-cast) has a hardness of around 100 HV, while Cu15 Zn (as-cast) has a hardness of 75 HV (Ezequiel et al., 2024). Nnakwo et al. also found that increased Zn content in the Cu alloy leads to decreased hardness due to increased grain size and solid solution region (Nnakwo et al., 2021). According to a study by Qu et al. and García-Mintegui et al., pure Zn hardness is between 41-42 HV (García-Mintegui et al., 2021; Qu et al., 2020). Therefore, it could be concluded that Zn in the alloy leads to decreased hardness due to Zn hardness less than Cu.

Several researchers correlated measured hardness to crystallite size (Augustin et al., 2016; Syamsuir et al., 2023). Augustin et al. have found an increase in Cu's crystallite size, promoting a decrease in scratch and micro-hardness (Augustin et al., 2016). Syamsuir et al. found a decrease in Cu's crystallite size, leading to an increase in hardness (Syamsuir et al., 2023). Comparing Table 2 with Figure 2, it can be seen that an increase in the Zn content led to an increase in crystallite size and a decrease in the hardness. On the contrary, while as-cast samples do not form an alloy (Cu), the resulting hardness is not aligned with the crystallite size. It seems that it cannot compare the crystallite size were found with measured hardness between alloy (CuZn) and un-alloy (Cu) material.

Several transportation sectors, such as ambulance equipment, are made from Al alloy (Blanco et al., 2022; Vandersluis et al., 2020). According to Hajizadeh et al., Al alloy hardness is between 32-52 HV (Hajizadeh et al., 2017). Therefore, all specimens have hardness still higher than Al alloy.

4.3 OCP

Figure 3 shows the OCP measurement result of various casting samples in 0.9 % NaCl at room temperature. Generally, increased Zn content in the alloy promoted more negative potential, which perfectly agrees with the Cocco et al. study (Cocco et al., 2016). Dridi et al. have found that E_{OCP} CuZn30 and CuZn39 are -0.578 and -0.604 V/MSE at 3 % NaCl, which means an increase in the Zn promoted to more negative potential (Dridi et al., 2020). Cu, Cu6Zn, Cu9Zn, and Cu12Zn samples have E_{OCP} potential at 1200 s measurement -0.014, -0.023, -0.027, and -0.032 V vs Ag/AgCl, respectively.

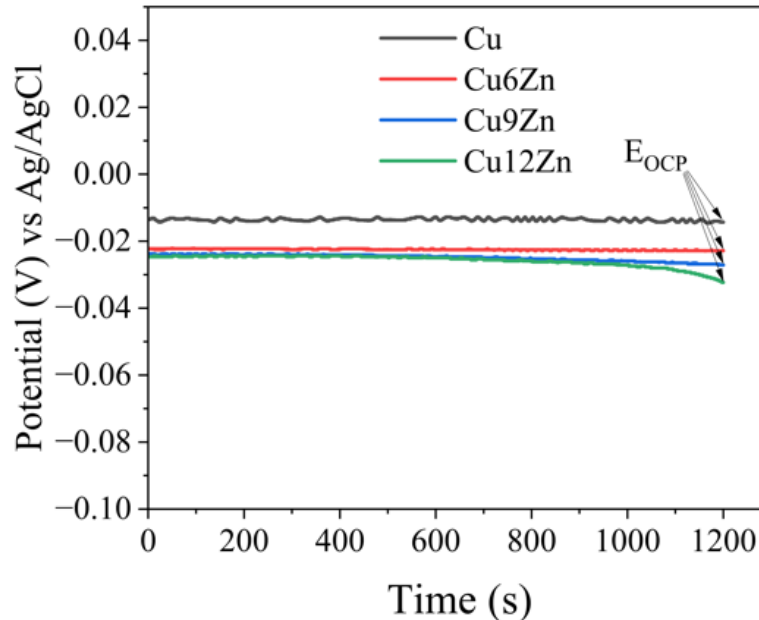
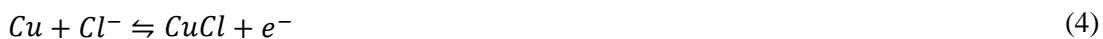


Fig. 3. OCP measurement of various casting samples.

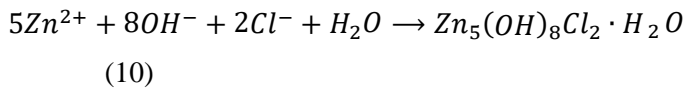
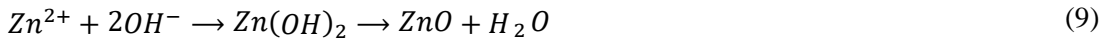
According to Figure 3, Cu, Cu6Zn, and Cu9Zn samples are steady at initial measurements until 1200 s, indicating that the protective layer formed has good protection. In contrast, the Cu12Zn sample is steady at initial measurements until 600 s, then moves in a more negative direction. These phenomena indicated that the formed protective layer had initially dissolved at 600 s; therefore, the measurement continuously moved forward in a negative direction until the measurement reached 1200 s.

4.4 LSV

Eccrine sweat and saline-infused for humans are nearly 0.9 % of NaCl (Bond & Lieu, 2014; Tayyab et al., 2021). Therefore, LSV measurement was conducted in 0.9 % NaCl at room temperature. Luo et al. found Cu₂O crystalline growth on the Cu surface when exposed to 0.9 % NaCl, while when exposed to pure water, Cu₂O crystalline was not seen (Luo et al., 2020). Commonly, Cu₂O crystallines are formed, which is preceded by the formation of CuCl when the specimen is tested in a chloride solution. Moreover, Zhang et al., in their study, found ZnO and Zn₅Cl₂(OH)₈·H₂O as corrosion products on top of Cu40Zn surfaces in a chloride environment (Zhang et al., 2016). The reaction of Cu in chloride solution is as follows (Milošev et al., 2024).



The reaction of Zn in chloride solution is as follows (Milošev et al., 2024).



LSV measurement results in 0.9 % NaCl can be seen in Figure 4.

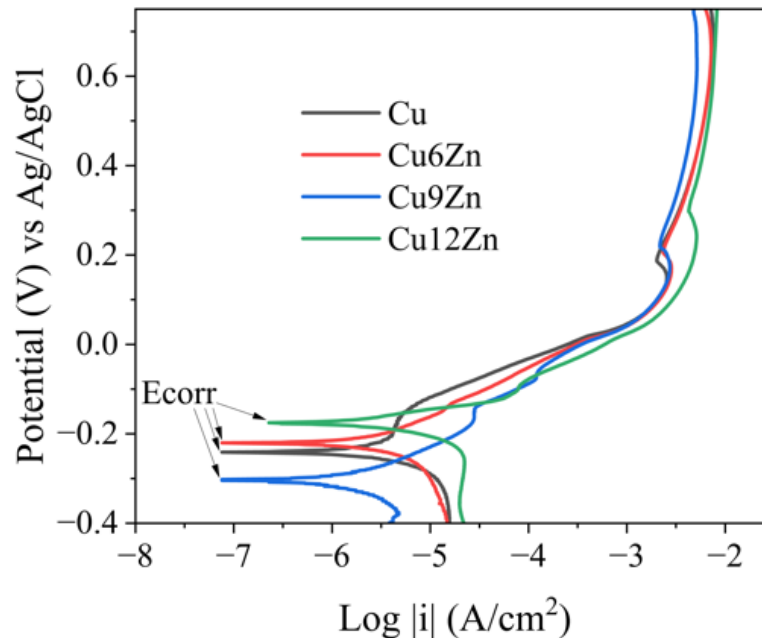


Fig. 4. LSV scans result of various casting samples.

According to Figure 4, corrosion E_{corr} and i_{corr} can be found using the Tafel extrapolation method. Moreover, the corrosion rate could be calculated by inserting i_{corr} into expression (1). Table 3 presented E_{corr} , i_{corr} , and the corrosion rate of various casting samples. It appears E_{corr} is independent of Zn addition; however, Zn is dependent on i_{corr} and corrosion rate (except for the Cu12Zn sample). This behavior is probably due to a protective layer that was formed. Compared to the OCP result, it can be seen that the Cu12Zn sample continuously moves forward in a negative direction from 600 until 1200 s of measurement. Therefore, that sample has a higher i_{corr} and corrosion rate.

Table 3 - Corrosion parameters of various casting samples.

Sample name	E_{corr} (V) vs Ag/AgCl	i_{corr} (A/cm ²)	Corrosion rate (mmpy)
Cu	-0.241	4.42×10^{-6}	5.13×10^{-2}
Cu6Zn	-0.220	3.33×10^{-6}	3.86×10^{-2}
Cu9Zn	-0.304	2.15×10^{-6}	2.49×10^{-2}
Cu12Zn	-0.175	6.19×10^{-6}	7.18×10^{-2}

Qu et al. have found that an increase in Zn content led to an increase in corrosion resistance, which perfectly agrees with the present study (except for Cu12Zn) (Qu et al., 2020). Milošev et al. investigated Cu, Cu10Zn, Cu40Zn, and Zn in 3 % NaCl and found i_{corr} after stabilized at 1 hour around 1.573, 1.456, and 2.114, and 5.21 $\mu\text{A}/\text{cm}^2$ respectively (Milošev et al., 2024). According to equation (1), i_{corr} strongly influences the corrosion rate. The more i_{corr} , the higher the corrosion rate. Moreover, a limitation in Zn content in the Cu alloy could influence the corrosion resistance. Presenting the Zn content ≤ 11 wt.% in the alloy could enhance the corrosion resistance; however, Zn of more than 10 wt.% could decrease corrosion resistance (Milošev et al., 2024).

According to Table 2, the Cu9Zn sample has the lowest microstrain than others. The measured microstrain could be associated with the sample's crystal defect (Soegijono et al.,

2020). Based on Table 2, the Cu9Zn sample has the lowest microstrain, which confirms that the sample has the lowest corrosion rate. Moreover, the FCC sample with the preferred orientation of the (111) plane could offer a lower corrosion rate due to the highest surface atomic density (Jinlong et al., 2016; Soegijono et al., 2020). Compared to other samples, the Cu9Zn sample has the higher preferred orientation of the (111) plane. Even though the (111) plane of the Cu sample is the highest. Unfortunately, the (220) and (200) planes are still present and relatively high.

4.5 Antibacterial Activity

Figure 5 shows the direct contact kill of *Staphylococcus aureus* and *Escherichia coli* after 24 hours of incubation. The present study focused on *Staphylococcus aureus*, but *Escherichia coli* was also used for comparison. There is no diffusion in the sample; therefore, the inhibition zone could not be seen.

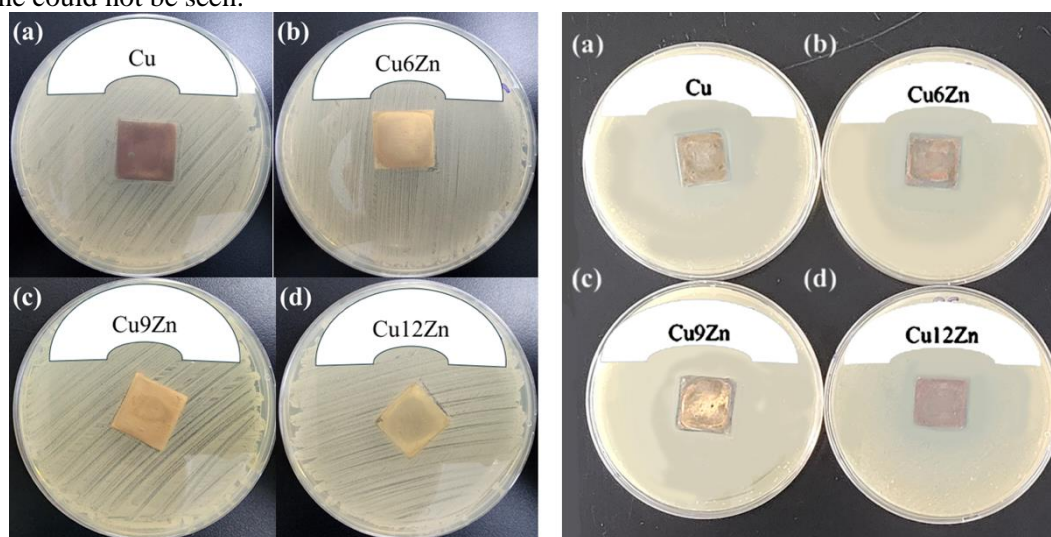


Fig. 5. Antibacterial activity test using *Staphylococcus aureus* (left) and *Escherichia coli* (right) (a) Cu, (b) Cu6Zn, (c) Cu9Zn, and (d) Cu12Zn.

Antibacterial activity after 24 hours of post-contact with various casting samples using *Staphylococcus aureus* and *Escherichia coli* can be seen in Figure 6. The removed sample places remain clear (with no regrowth) from bacterial activity. The antibacterial behavior was significantly influenced by Cu or Zn ions (Qu et al., 2020). Villapún et al. found that releasing Cu ions leads to the highest killing activity of *Staphylococcus aureus* (Villapún et al., 2016). Excess in the Cu ion could be bacteriostatic (Sabbouh et al., 2023). Moreover, Cu ions could be adsorbed on the cytoplasmic membrane surfaces, then penetrate the bacteria, react with sulfhydryl groups and cause the cell to die (Zeng et al., 2022). Furthermore, Cu ions could form hydroxyl groups in the presence of oxygen in nature, which could destroy cell membranes (Dou et al., 2022). Hutchings et al. stated that Zn^{2+} successfully inhibits the growth of *S. epidermidis* (Hutchings et al., 2021). This behavior is associated with the generation of reactive oxygen or the formation of (Zhang et al., 2021).

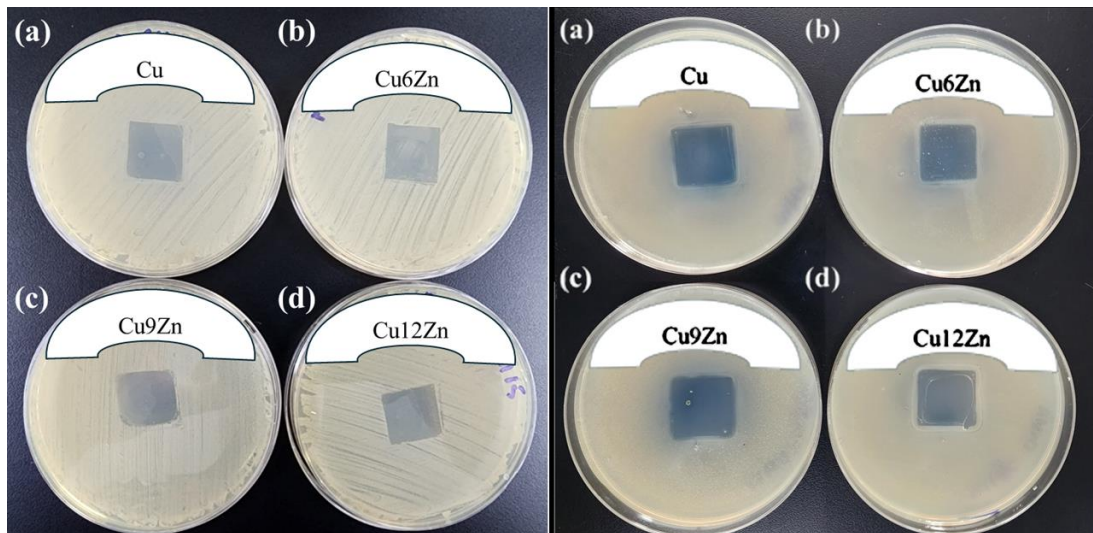


Fig. 6. Antibacterial activity after 24 hours of post-contact (regrowth assessment) towards *Staphylococcus aureus* (left) and *Escherichia coli* (right) (a) Cu, (b) Cu6Zn, (c) Cu9Zn, and (d) Cu12Zn

Figure 7 shows the fluid contact test of *Staphylococcus aureus* and *Escherichia coli*. The orientation of the test materials is mapped within the yellow box. For the Cu6Zn sample, *Escherichia coli* was killed on the 3rd hour. However, there is no significant reduction within the fluid because there are no diffusible materials. Also, there is no visible growth after 3rd hour for the fluid in contact with the metal.

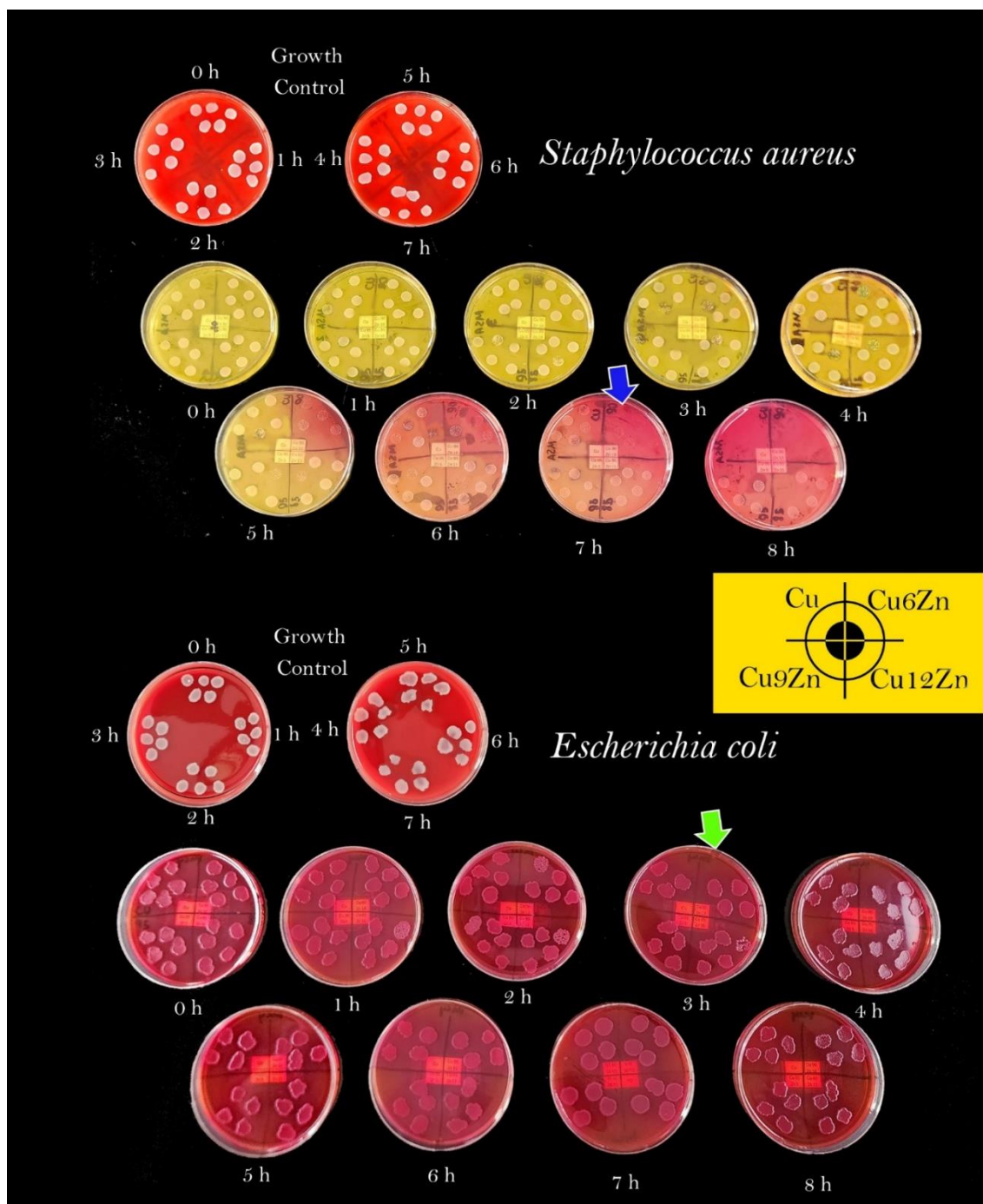


Fig. 7. Fluid contact test of *Staphylococcus aureus* and *Escherichia coli*

Moreover, it should be noted that the reduction of the colony is significant in the 7th hour for *Staphylococcus aureus* (blue arrow), but for *Escherichia coli*, the inhibition of *Escherichia coli* growth is shown within the 3rd hour of contact (green arrow), for Cu6Zn sample. The reduction of colonies is significant on the surface of the metal. While on the remaining fluid, the reduction is insignificant until 8 hours. This behavior is because *Staphylococcus aureus* and *Escherichia coli* have different membrane structures and thick cell walls, therefore could inhibit ion exchange and restrain the antibacterial effect of Cu and or Zn ions (Di et al., 2022). Cu killing is more effective in Gram-negative bacteria (e.g., *Escherichia coli*) because peptidoglycan affects the cell's susceptibility. The thicker the peptidoglycan layer, the harder it became for the Cu ions to reach the membrane (Soltani et al., 2020; Xhafa et al., 2023). Therefore, *Escherichia coli* was killed in the 3rd hour. Another reason Cu6Zn has better antibacterial performance than others is probably due to its smaller crystallite size. Researchers found that smaller crystallite sizes promote the enhancement of antibacterial effects (Syamsuir

et al., 2023). This behavior is attributed to an increase in surface area due to the crystallite size (Sangeetha et al., 2015). The Cu6Zn sample could be used as an alternative material for medical equipment in ambulances.

5. Conclusion

CuZn has been successfully fabricated. XRD confirmed that CuZn alloy has a single alpha phase with an FCC crystal structure. The rise of the Zn content in the alloy led to a decrease in the hardness due to an increase in crystallite size and led to a shift to more negative OCP potential at 1200 s measurement. Moreover, the rise of the Zn content to 9 wt.% decreased the corrosion rate. It appears there is a limitation in Zn content in the copper alloy that influences the corrosion rate, as shown when Zn content around 12 wt. % is promoted to increase the corrosion rate. Antibacterial activity observation found that all samples had no diffusion. Moreover, 24-hour post-contact observation found that sample places removed from the sample remained clear of bacteria. The Cu6Zn has better antibacterial performance than others due to the smallest crystallite size. According to the fluid contact test, the reduction of the colony of *Staphylococcus aureus* is significant in the 7th hour. The inhibition of *Escherichia coli* growth is also shown within the 3rd hour of contact. This behavior is because *Staphylococcus aureus* and *Escherichia coli* have different membrane structures and thick cell walls, therefore, could inhibit ion exchange and restrain the antibacterial effect of Cu and or Zn ions.

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