

**Cu Later: Balancing Copper Ion  
Release and Antibacterial  
Performance of Copper Alloys  
Against *E. coli* k-12**

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# Research Questions

- Do alloys with **higher copper content** show **stronger antibacterial effect** on *E. coli* K-12?
- How does **bacterial presence** influence **copper ion ( $\text{Cu}^{2+}$ ) release** over time?

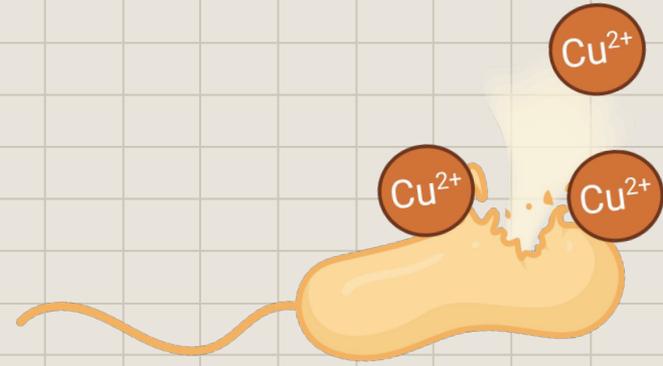
## Purpose

**Evaluate the trade-off between antimicrobial performance and environmental toxicity.**

# Key Concepts

- Copper kills bacteria by:
  - Damaging cell membranes
  - Disrupting proteins (thiol binding)
  - Generating reactive oxygen species
- Antibacterial activity depends on **Cu<sup>2+</sup> ion release**
- Excess copper ions can cause **environmental toxicity**, especially **aquatic ecosystems**

Copper ions interacting with *E. coli*



**Gap I Found!**

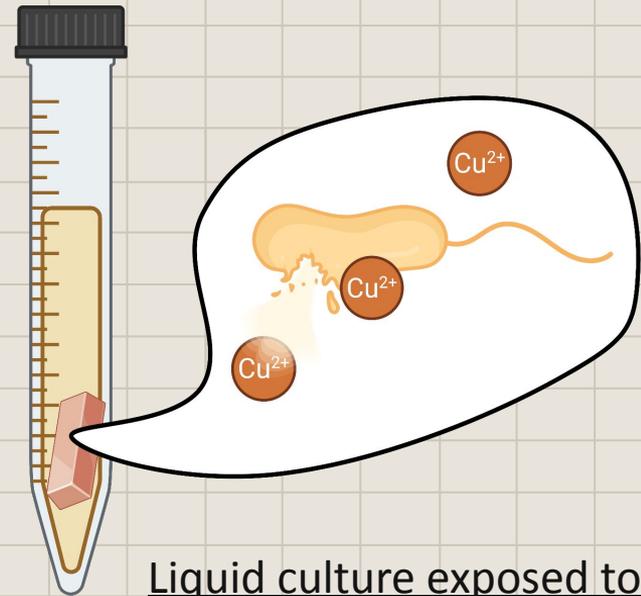
Limited research compares **antibacterial effectiveness vs. ion release simultaneously**

# Hypothesis

- **Increasing copper content** → **increased antibacterial activity** due to enhanced  $\text{Cu}^{2+}$  release

# Prediction

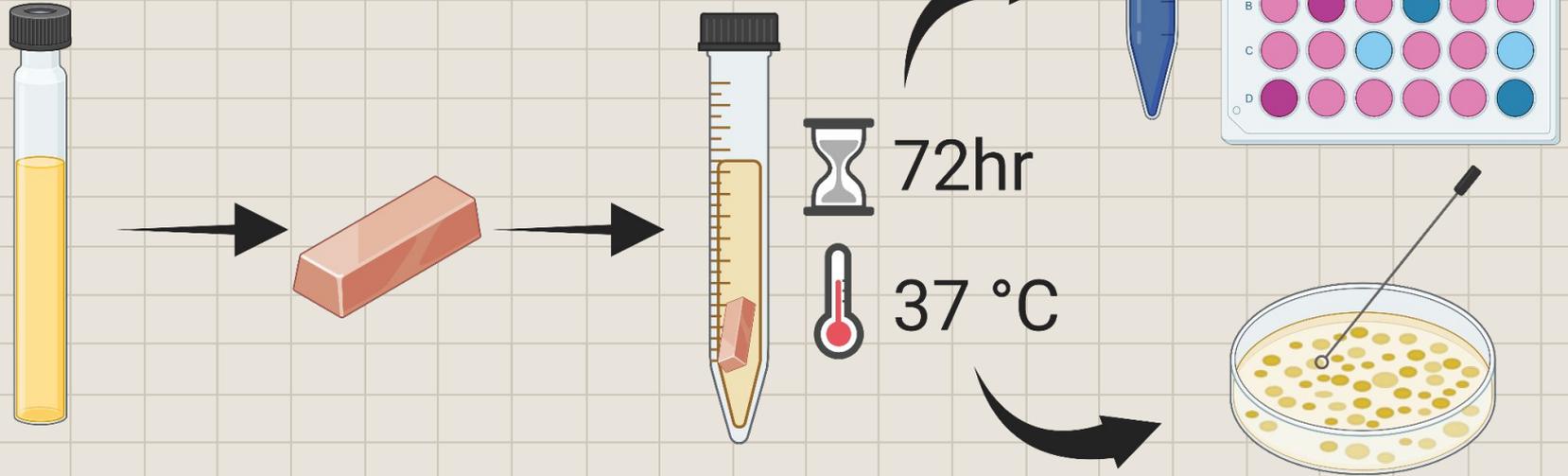
- **Brass** will show strong antibacterial effects compared to **pure copper**
- **Brass** will release fewer  $\text{Cu}^{2+}$  than **pure copper**



Liquid culture exposed to copper alloy, releasing  $\text{Cu}^{2+}$

<b>Independent Variable:</b>	Percent of copper in metal Zinc (0%), brass (65%), bronze (88%), copper (100%)
<b>Dependent Variable:</b>	Bacteria viability ( <b>CFU/mL</b> ), Metabolic activity ( <b>resazurin</b> ), and $\text{Cu}^{2+}$ concentration (mg/L)
<b>Biological Control:</b>	E. coli + LB broth
<b>Chemical Control:</b>	Metal + LB broth

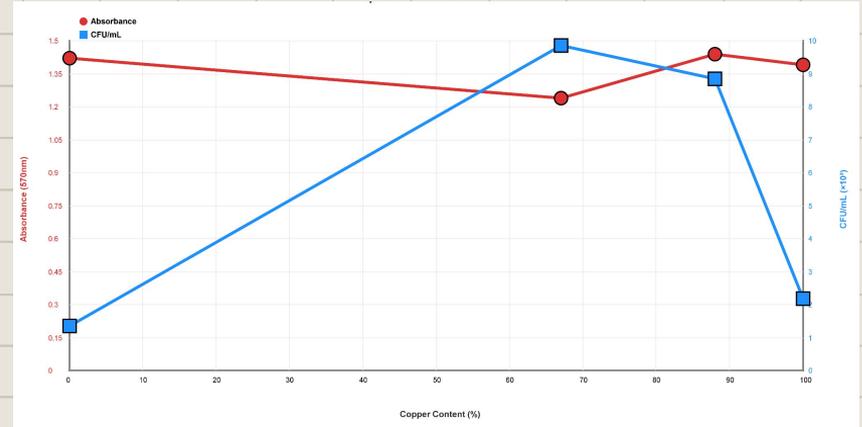
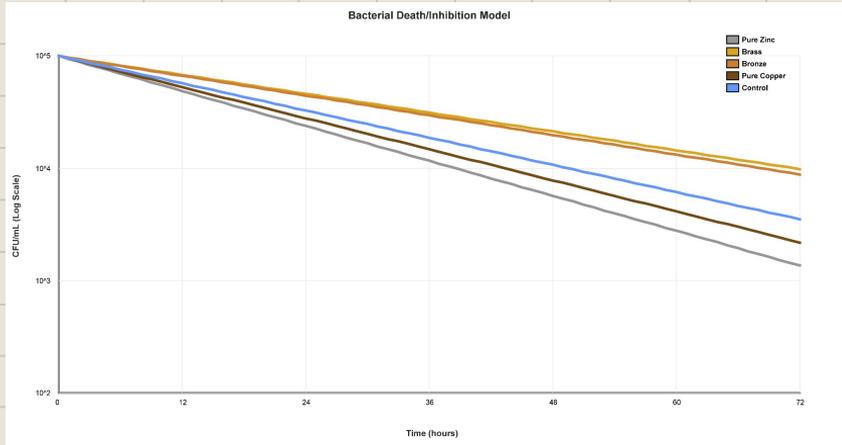
# Method Overview



1. Grow *E. coli* K-12 liquid culture
2. Standardize to **OD600 = 0.1**
3. Add metal alloys to treatments
4. Incubate for 72 hours at 37 °C (**n=3**)

5. Measure: **Resazurin absorbance** every 24, 48, 72 hrs (570 nm), CFU plate counts (**3 plates per tube**), and Copper ion concentrations

# Results



- Copper ion release increased with copper percentage
- Pure copper released the highest Cu<sup>2+</sup> followed by bronze then brass, predictably
- Antibacterial activity was **not linear**
- Strongest metabolic inhibition occurred at 80%-100% copper
- **Optimized antibacterial window** showcases range of copper ion release that **maximizes antibacterial activity with limited ion release** compared to pure copper

# Statistical Analysis

## One-way ANOVA

- $F = 78.30$
- $P = 0.00005$

## Interpretation

- Alloy composition **significantly** affects bacterial viability

## Additional Observations

- CFU data confirmed strong antibacterial effects in **zinc** and **copper**
- $\text{Cu}^{2+}$  release **did not directly** predict antibacterial activity

# Summarized Conclusions

- Antibacterial activity does **not increase linearly** with copper content due to **multi-mechanism variables** confirmed by the use of **Zinc**
- An **optimized antibacterial window** exists
- Maximum  $\text{Cu}^{2+}$  release is not required for strong bacterial inhibition
- Multiple mechanisms contribute to antimicrobial effects, rather than a **“more copper = more killing” outlook**

# Applications & Future Research

## Applications

- **Healthcare:** physical antimicrobial surfaces
- **Public infrastructure:** high-touch handrails
- **Environmental toxicology:** safer metal engineering + use

## Future Research

- Test pathogenic bacteria
- Analyze surface roughness effects
- Investigate additional alloy compositions

