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*Full Length Research Paper*

# Antibacterial properties of some metals and alloys in combating coliforms in contaminated water

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**An investigation into the antibacterial properties of silver, copper, aluminium, zinc, brass, bronze, tin and silicon, in combating coliform bacteria in contaminated water has been carried out. The metals, in the form of thin plates, were immersed in water samples at various time intervals at room temperature (~20°C). Total coliform and *Escherichia coli (E. coli*) enumeration was done on the samples, before and after immersion, using 3 M Petri films. The results show that copper, silver and zinc in group I and II of the periodic table of elements have the highest rate of destroying the bacteria, followed by aluminium in group III, and the least reaction was in tin and silicon, both in group IV. These results indicate that the antibacterial activity of materials depend on the group they belong to in the periodic table; the higher the group the lower their efficacy. No direct relationship has been found between the metal activity and ionisation energy. An interesting observation was that zinc was as effective as copper in destroying the bacteria. It was also found that the more the surface area of the metals, the faster they destroyed the coliforms. Practical applications of the results are outlined.**

**Key words:** Metals, alloys, coliforms, *Escherichia coli.*

## INTRODUCTION

Water is an essential resource for life and good health. Supply of adequate clean water to communities is a problem worldwide, especially in developing countries where rural population depends on water from rivers, dams and streams for domestic use. It is estimated that around 1.2 billion people or almost one-fifth of the world's population live in areas of physical water scarcity, and another 1.6 billion people, or almost one quarter of the world's population face shortage of clean water (United Nations, 2007). Lack of water to meet the daily needs is a reality today for one in three people around the world. Globally, the problem is getting worse as cities and population grow (World Health Organization, 2009). Nearly 2.2 million people die each year from consumption of contaminated water, and 9,500 children alone die every day (Sciperio, 2009). More than 300 million lack clean water in Africa alone and approximately 2.5 billion have no access to adequate sanitation (Water for Africa, 2010). The majority of these cases occur in rural areas of developing nations where the water supply remains polluted and adequate sanitation is unavailable.

The major cause of shortage of clean water is pollution due to sewage and industrial waste discharge into rivers. Industrial discharge is responsible for the presence of heavy metals and toxic chemicals. Exposure to these metals can cause serious health problems to human beings and animals. Sewage discharge into rivers and dams is the major cause of water – borne diseases in human beings caused by pathogenic microorganisms, mainly the coliforms. They are present in the environment and in the faeces of all warm-blooded animals and humans. Among the many types of coliforms found in untreated water, *Eschericia coli* is the most common bacterium that lives in human and animal intestines in large numbers. There are hundreds of *E. coli* strains and most of them are relatively harmless, except the *E. coli* strain 0157:H7 that produces a powerful toxin which can cause severe illness like diarrhoea, kidney damage, and occasionally death, whereby it becomes a major health hazard. In the recent times, a great deal of effort is being made to find ways of eliminating these bacteria in water to make it portable.

Taking into account the importance of making clean water available to all human beings, researchers have been exploring ways to develop simple and affordable methods of water disinfection for use in places where fresh water resources are non-existent and the available water contains large amounts of pathogens. Such systems should be stand-alone, use local expertise for fabrication and maintenance and should be operational without power source. More importantly, they should be able to eliminate pathogens completely to ensure safety against diseases.

For centuries copper, silver and brass are known to have the property of destroying bacteria on contact with them, and a lot of information is now available on their practical use for food safety and hygiene, for disinfection of water in swimming pools and hospitals, wound healing, air disinfection and surface sanitation (Beer et al., 1999; Keevil, 2000; Dresher, 2004; Rogers et al., 1994a,b; Domek et al., 1984; Landeen et al.,1989; Santo et al., 2008; Wilks et al., 2005; Lin et al., 1996, 1998; Blanc, et al. 2005). It is believed that the germicidal property of metals, especially heavy metals, is due to oligodynamic effect in which metal and metal compounds, when introduced into the interior of bacterial cells, have ability to change and finally kill them in a characteristic way (Charles et al., 1948). Copper and silver are the most studied metals for oligodynamic action. Data from silver suggest that its ions denature proteins in the bacterial cells (Gram- positive and Gram-negative) by binding to reactive groups resulting in their inactivation (Mikihoro et al., 2005; Shrestha et al., 2009). In disinfection systems, which contain both copper and silver, copper ions penetrate the cell wall and as a result, they will create an entrance for silver ions, which penetrate the core of the microorganism. The silver ions bind to various parts of the cell, such as the DNA and RNA, cellular proteins and respiratory enzymes, causing all life support systems in the cell to be immobilized (Lenntech, 2010). A recent study on copper surfaces suggests that hydroxyl radicals in the solution are responsible for their lethal action (Santo et al., 2008). Whatever causes the inactivation, whether ions or radicals, the exact mechanism of the antibacterial action that takes place within the cells is not yet known. Therefore, one of the objectives of this project was to study the germicidal action of metals having different electronic and chemical properties to see if the results can lead to some clue.

In the past, there have been several reports on the use of antibacterial property of metals such as copper, silver and their alloys for water purification. Generally, this technique is used in large scale, commercial water disinfection units in which metal ions are generated by electrolytic or chemical means, and the technique is found to be quite effective (Dresher, 2004). However, there seems to be no much effort made to use this as a means of water purification for small-scale stand - alone systems for use by rural communities and in emergencies. Such a system should be operational without any power source and should avoid use of chemicals to safeguard against recontamination by the by-products. A disadvantage of electrolytic systems is that it needs a power source for operation. Use of chemical for ion generation is also not welcome because it requires an additional filter unit to remove the by-products. A simple point-of-use (POU) water treatment unit that utilises the antibacterial property of metals might be a solution to the current water crisis in rural areas of underdeveloped countries. Coliform bacteria, especially *E. coli strain 0157:H7* is a major disease causing pathogen commonly found in contaminated water in rivers and dams. Removal of these bacteria is necessary in water disinfection processes. Therefore, this study was also intended to find the extent to which some selected metals and alloys could destroy coliform bacteria present in contaminated water by simply immersing them for various time intervals without the use of any external agency to produce the metal ions.

## MATERIALS AND METHODS

Materials were selected from groups I to IV in the periodic table. They were silver, copper, aluminium, zinc, tin and silicon. Two alloys, bronze, brass, were also included in the study. All materials were in the form of plates having thickness of 0.5, and 50 x 50 mm size. They were all 99.5% pure. Brass had a composition of 63% copper and 37% zinc, while bronze had 94% copper and 6% tin. Metal plates were cleaned using abrasive, washed in distilled water, and finally dried in hot air. They were immersed in 200 mL raw water samples collected from a nearby river, for specific time intervals of 30, 60, 90, 120 and 150 min at room temperature (20 ± 1°C). 3M Petri films were used to enumerate total coliform and *E.coli* in the samples before and after immersion of the metals. For this purpose, 1 mL water was spread in to the Petri films using a dropper and the plates were placed in an incubator for 48 h at 35°C. Coliform counts in the plates were done using a light source and a magnifier. Three sets of readings were taken for each metal using different raw samples and the average counts were used for the analysis. Subsequently, copper and aluminium plates with varying surface area were immersed in 200 mL raw water and coliform counts were done after 3 h to study the effect of surface area of the metal on the survival of the coliforms. Here also, average of three tests was used for analysis. Though only two metals were used in this test, it is anticipated that the similar trend would follow in other materials as well.

## RESULTS

### Rate of metal reaction with coliforms

Figures 1 and 2 show the rate at which the materials under study can eliminate total coliform and *E.coli* from contaminated water samples. It is clearly seen that among the metals silver, copper in group IB and zinc in group IIB have the fastest reaction rates. Silver could bring down the total coliform and *E.coli* content by more than 98% in about 90 min, while copper and zinc reduced them to zero levels within the same time interval. Copper is slightly more reactive than zinc. Group IV elements tin

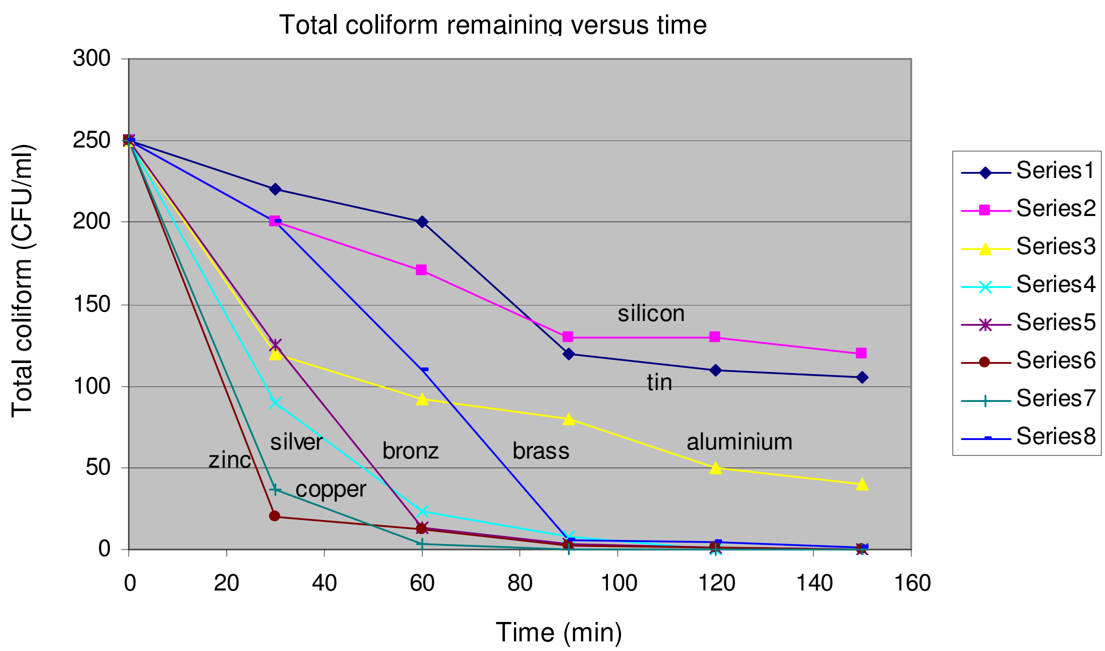


**Figure 1.** Total coliform count remaining in 200 mL of contaminated water after immersion of the metal plates for various time intervals.

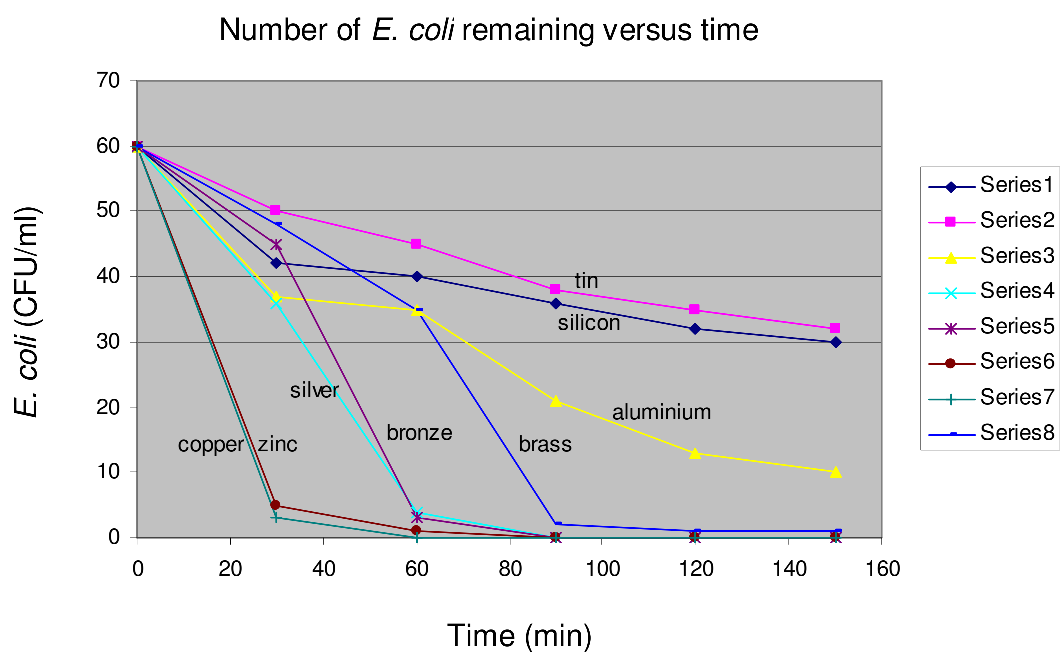


**Figure 2.** *E. coli* count remaining in 200 mL of contaminated water after immersion of the metal plates for various time intervals.

and silicon were the least reactive. Aluminium in group III lies in the middle range of the activity. Antibacterial activity of the alloys, brass and bronze was slower than that of copper and zinc. Bronze is slightly faster than brass in destroying the coliforms.

### Effect of surface area of metals on the antibacterial activity

Figure 3 shows the antibacterial effect of copper and aluminium as a function of the surface area of the metal

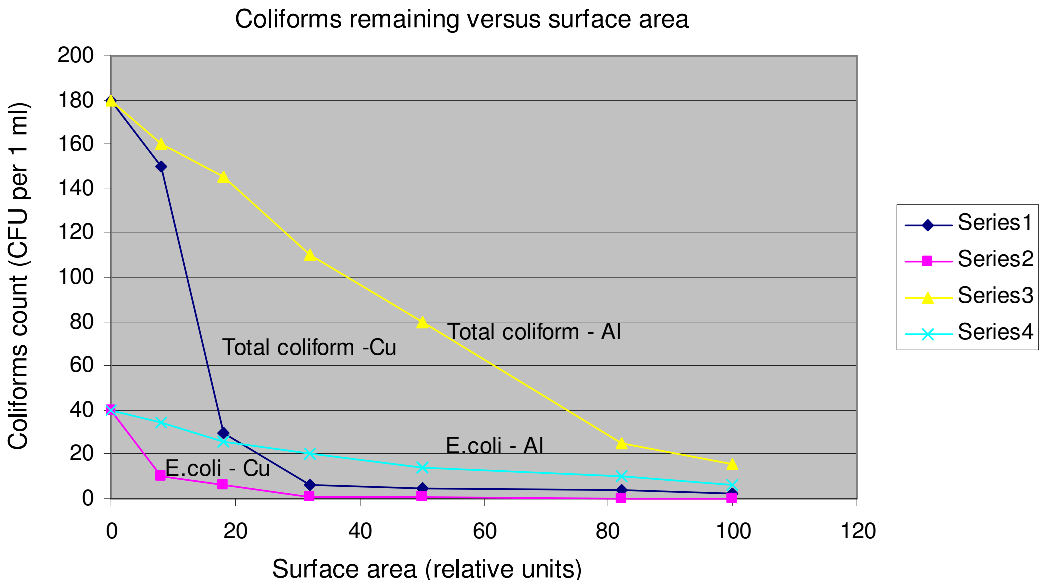


**Figure 3.** Total coliform and *E. coli* counts remaining in 200 mL of contaminated water after 3 h of immersion of copper and aluminium plates having different surface area.

plates. The reaction was monitored after 3 h of immersion in 200 mL water. Both total and *E. coli* were found to get destroyed much faster on larger surfaces.

## DISCUSSION

Oligodynamic effect has been known for decades, and there are several reports on the effective use of this for various applications. Previous studies on the antibacterial activity of copper and silver have confirmed that metal ions are responsible for the inactivation of the bacteria. However, there seems to be no clear-cut explanation of the inactivation mechanism taking place within the cells of these microorganisms. Some reports suggest that metal ions bind to DNA, enzymes and cellular proteins in the bacteria causing cell damage and death (Mikihoro et al., 2005). A recent study on copper surfaces suggests that hydroxyl radicals in the solution are responsible for their lethal action (Santo et al., 2008). This is in agreement with the observation that hydroxyl ions destroy *E.Coli* bacteria when water is passed through $TiO_2$ films under UV light (Cho et al., 2004). However, it is not known how they contribute to the actual detoxification mechanism.

One of the objectives of this project was to get some insight into the kinematics of the germicidal action of metals with varying degrees of electronic and chemical properties. Figures 1 and 2 show that both total coliform and *E.coli* are destroyed with time of immersion of the metals in water. This should be as a result of these organisms coming in contact with the metals as they move about due to Brownian motion. Because metal ions are assumed to be responsible for germicidal action, it was expected that ionisation energy of the metals would be related to their toxicity towards coliforms; the less the ionisation energy, the faster the reaction. However, the results do not show any such relationship. For instance, the ionisation energy of silver is 7.57 eV whereas that of aluminium is 5.98 eV, yet results show that copper is more reactive than aluminium. Similar observations are made in terms of the chemical activity of these metals, as well. Tin lies much higher in the activity series than copper and silver, though it is almost inactive with coliforms. A reasonable trend observed in this study is that the germicidal activity of metals depends on the group to which they belong in the periodic table. Reaction rate was the highest in copper and silver group IB and zinc in group II B. This was followed by aluminium in group III and least reaction was found in tin and silicon, both group IV. Thus, a correlation between germicidal activity and valence could be established, whereby the activity decreases with increase in the number of valance electrons in the material. The significance of this result is that it might help identifying a metal as far as its germicidal effect is concerned. It has not been possible to confirm whether it is the metal ions or the hydroxyl radicals that are responsible for the inactivation of the coliforms; neither does it give any clue to the actual mechanism taking place within the cell that leads to its death.

Antibacterial activity of the alloys, brass and bronze was lower than that of copper and zinc. Bronze is slightly faster than brass in destroying the coliforms. This is expected since bronze had 94% copper compared to 63% in brass. This observation is in agreement with previous reports that the percentage of copper determines the antibacterial capacity of copper alloys (Noyce et al., 2006).

The second objective of the study was to see if the germicidal property of metals could be used for simple, stand-alone water purification systems for use in rural communities and homes. Commercial water treatment devices using copper and silver are available today (Aqualyse, 2010; Doulton USA, 2010). It is of interest to note that in such devices metals ions are generated through electrolytic or chemical processes. Therefore these devices are not feasible in places where electrical power in not available. In addition, use of chemicals is not recommended as it might add by-products into the water, removal of which requires additional treatment making the whole process very complex. Since the germicidal properties of metals are known, it is worthwhile utilising them for point-of-use water treatment systems in remote areas where portable water is not available. The results of this study show that such systems are feasible. As shown in Figures 1 and 2, copper, zinc and silver could eliminate both total colifom and *E. coli* completely within a few hours. The data could be used as a guideline to estimate the size of the metal needed for treatment of a specific quantity of water, depending on the initial coliform concentration. It is also of importance to note that mere presence of the metals in the water could destroy the coliforms. A possible explanation of this is that the metal ions could have been produced by reaction with other ions present in the contaminated water. Further investigation is necessary to confirm this.

Figure 3 shows how the reaction rate of the two metals, copper and aluminium depends on their surface area. Larger area can eliminate the bacteria in a shorter time. This is expected since more bacteria come in contact with the metal as the area increases. Though only two metals were used in this study it is expected that other metals would also follow the same trend. From practical point of view, such results can be used to estimate the metal size against the type of metal used. The choice of the metal type and quantity should be made after a thorough investigation into the acceptable levels of concentration of ions remaining in the water after treatment. This is because if excess ions remain, they can be toxic. In addition, for drinking purpose, there is need to eliminate other hazardous physical and chemical pollutants usually present in water from rivers and dams, using other means such as a porous medium to filter out such contaminants. It is expected that combined with the use of appropriate metals or alloys, it would provide a safe, simple and cost-effective means of water purification for use in rural communities.

## Conclusion

The key findings of this study are:

a. Some metals and alloys have antibacterial property to eliminate coliforms from contaminated water by simple immersion.
b. Among the materials studied, antibacterial effect is highest in copper, zinc and silver, least in silicon and tin, and that of aluminium lies somewhere in between these two extremes.
c. Zinc is as effective as copper in eliminating coliforms.
d. The antibacterial property of a material depends on the element group it belongs to in the periodic table. The higher the group, the lower is its ability to react with coliforms.
e. No definite correlation is found between the antibacterial property of metals and their ionisation energy or their position in the reactivity series.
f. Among the alloys, bronze, with a higher copper concentration has faster reaction rate than brass.
g. One of the factors that control the rate of reaction with coliforms is the surface area of the metal. Larger area destroys coliforms at a faster rate.

These results can be useful for designing stand-alone water purification systems for household and communal use.

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