

**Usefulness of bronze containers for decontamination of
Gram - negative and Gram - positive bacteria from water**

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Bronze containers of different sizes with different composition of copper have beneficial effects in decontaminating different bacteria from water at different durations. Experiments revealed that Gram- negative bacteria in water were no longer detected after storage in bronze containers within 24 hours. The experiment was conducted by using bronze containers of different capacities (50 ml-500 ml). Sterilized distilled water was seeded with 10^5 - 10^7 organisms per ml each of *Escherichia coli* (ATCC 25922), *Salmonella typhi*, *Proteus morganii*, *Vibrio cholerae* *Staphylococcus aureus* and *Bacillus subtilis* from overnight broth cultures. Bacteria count was done hourly by using respective selective media plates such as MacConkey agar for *E.coli* and *S .typhi* and TCBS agar for *V. cholerae* and nutrient agar for *S. aureus* and *B. subtilis* by the streak method. The plates were incubated at 37°C overnight and colony count was calculated. It was observed that all Gram-negative bacteria tested were not recovered after 4 hours of storage. The growth of bacteria depends on the type of bronze used.

INTRODUCTION

It has been known since a long time ago that silver, copper, and zinc are commonly known elements with antibacterial and antifungal properties. Since silver coated materials have been found inappropriate in some applications, attempts have been made to investigate the antibacterial properties of copper-vermiculite [1].

Antibacterial and antifungal materials have attracted great interest in recent years because of their ability to inhibit the growth of microbes. Potential uses of antibacterial and antifungal materials include those for household, industrial and public facilities. Appropriate release of antibacterial metallic ions from composite can effectively inhibit growth of harmful microbes. Compared to the organic antibacterial material, ideal inorganic antibacterial material may have advantages such as long lasting action, chemical stability, thermal resistance,

safety, protection from drug resistance, and effectiveness against a broad spectrum of bacteria or fungi [2].

In Myanmar, bronze containers are used as household utensils in many ways and among them bronze cups are used as drinking cups, which are placed on top of earthen pots used for storage of drinking water in both rural and urban areas. Thus, this research aimed to investigate the decontamination of bacteria from water by using bronze containers.

MATERIALS AND METHODS

Types of bronze containers used

Four different kinds of Myanmar bronze containers were used.

Other containers

Stainless steel containers, glass beakers, porcelain basins and chinaware were used.

Bacteria used as contaminants

- (1) *Escherichia coli* ATCC25922
- (2) *Proteus morganii* (BIKEN)
- (3) *Salmonella typhi* (BIKEN)
- (4) *Vibrio parahaemolyticus*
- (5) *Staphylococcus aureus*
- (6) *Bacillus subtilis*

Size of inoculum

Different amounts of water were used with respect to the size of containers. The number of bacteria in an overnight broth culture was calculated from its density by using the spectrophotometer at the wavelength of 450 nm as well as by plate dilution method. The water was allowed to contaminate with respective bacteria from overnight broth culture to obtain the concentration of 10^6 to 10^7 CFU/ml.

Determination of the rate of decontamination

The contaminated water in different containers was kept at 25°C (RT) with an aluminium foil cover. One milliliter of water from each tested container was collected hourly up to six hours and again at 24 hours (one day) up to 5 days. Plating of samples was done on respective media and incubated at 37°C overnight. The colony forming units per ml were then calculated.

RESULTS AND DISCUSSION

As shown in Table 1, Gram-negative bacteria count in water from bronze containers decreased hourly. In contrast, the rate of reduction of Gram-positive bacteria is slower than that of the Gram-negative bacteria. In experiments of using glass beakers, porcelain basins and stainless steel containers, the growth of bacteria after 24 hours was not significant and bacteria count was not decreased from the initial count. However, there was a 10-fold decrease in bacterial count when using earthenware (chinaware), and 20-fold

Table 1. Decontamination rate of bacteria after storage in bronze containers

Time (hour)	Gram-negative bacteria				Gram-positive bacteria	
	<i>E. coli</i>	<i>S. typhi</i>	<i>P. morganii</i>	<i>V. parahaemolyticus</i>	<i>S. aureus</i>	<i>B. subtilis</i>
0	10^6	10^6	10^7	120	10^6	10^6
1	150	20	50	100	1250	10^6
2	100	18	40	0	10^3	10^6
3	0	9	20	0	10^3	10^6
4	0	1	0	0	10^3	10^6
5	0	0	0	0	10^3	10^6
6	0	0	0	0	99	10^6
24	0	0	0	0	55	0

decrease when a bronze spoon was dipped in the glassware container for 4-8 hours (depending on the type of spoon). Moreover, the bacteria in water using stainless steel containers survived up to 45 days. As shown in Table 1, the bacterial count decreased hourly for Gram-negative bacteria. Thus, bronze containers are effective for the reduction of bacteria. However, Gram-positive cocci *Staphylococcus aureus* and *Bacillus subtilis* (saprophyte) took longer time than 4 hours, but the bacteria count decreased ten-fold. At the same time, when the contaminated organisms were higher than 10^6 organisms per ml it took more than 24 hours to be killed. Similarly, the decontamination effect of bronze depends on type of different utensils. Experiments also show that bronze could eliminate Gram-negative bacteria within 4 hours compared to Gram-positive bacteria. It will be the best container for using water storage as it could inhibit the growth of bacteria. Thus, this experiment shows that bronze containers are very useful to decontaminate pathogenic Gram-negative bacteria (water and food borne infectious agents). It has been reported by other workers [3,4] that bronze has antibacterial activity, and the benefits obtained from bronze materials may be useful tools for preventing water-borne infections in many ways for mankind. Copper is a naturally hygienic metal that slows down the growth of coliforms [5,6].

It has important potential application in food preparation, hospitals, making of coins (biocidal copper), door knobs and plumbing systems.

Bronze=Copper+ Zinc

Bronze=Copper+ Tin

Cupro nickel=Copper+Nickel

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