Wire Arc Spray Copper Coatings in Medical Applications

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Abstract

Infection of medical devices and treatment rooms can cause significant morbidity and mortality. Having antibacterial surfaces such as silver and copper coated areas reduces the risk of bacteria growth considerably. In the current study, wire arc spraying technique has been utilized to produce an ultra fine microstructure Antibacterial copper coating on stainless steel substrate. The chemical composition, microstructure, and surface morphology of copper coatings were characterized with X-ray diffraction (XRD) and scanning electron microscope. Determination of thickness and adhesion of the coating were investigated. The antibacterial property of copper coatings was analyzed by both gram negative Escherichia coli NCTC 10418 and gram positive Staphylococcus aureus NCTC 11047. The antibacterial performance of coatings was compared to stainless steel 316 and a micro grain structure of the commercially available copper. Results indicated that as-sprayed copper coatings have an excellent antibacterial behavior compared to stainless steel and micro grain copper which can be contributed to the fine grain size and existing of defects and micro pores in the microstructure.

Introduction

In modern life with scientific and technological advances, much attention is paid to the safety, sanitation, and health of environments. Therefore, daily appliances are increasingly being designed with antibacterial features (Ref 1).Bacterial infection is one of the major clinical complications. Prevention of bacterial-related infection remains a major challenge for the delivery of quality of medical care and the problem results in a high rate of mortality and morbidity resulting in increasing health care costs significantly (Ref 2-4).

Silver and copper have been widely utilized as effective materials for sterilizing liquids, and also human tissues for centuries. Some advantages of copper materials including their bactericidal activity were well known even since the time of ancient civilizations (Ref 5). Today copper sometimes is utilized as an agent for purification of water and inactivation of some microorganisms and bacterias (Ref 6). In spite of the negligible responsiveness of human tissues to copper, microorganisms show high sensitive to copper (Ref 7, 8). Recently, due to development of some resistant bacteria strains against the antibiotics, the antibacterial activity of nanostructure or ultra fine materials such as silver and copper has been received great attentions (Ref 9,10).For instance silver nanoparticles and nanostructures with a high bactericidal activity have been widely applied in medicine to prohibit colonization of bacteria on prostheses, dental materials, wound dressing, and to reduce infections in burn treatment (Ref 11-13). Antibacterial activity of copper nanostructures and ultrafine grain has not been studied, due to fast oxidation of metallic copper and both chemical and physical instability of the copper oxides formed at temperature below 200^0C , particularly if Cu^{2+} are formed (Ref 14,15).

Determining an appropriate copper coating with high antibacterial properties is inevitable and essential (Ref 12).Twin wire arc spray is known to be one of the less expensive thermal spraying processes with an ability to produce dense coatings with a wide rate of material deposition (Ref 16).It is proven that the microstructure of these coatings mainly depends on the spray parameters. In the current research, wire arc spraying technique has been applied to deposit copper coatings on stainless steel substrates. The microstructure and antibacterial properties of the coatings were characterized and investigated comprehensively.

Materials and Methods

Materials and Preparation

In the current study, wire arc spraying technique has been utilized to deposition copper coating on stainless steel 316 substrates. Two consumable copper wires with 1.6 millimeter in diameter (Sulzer Metco, USA) were fed automatically to meet at a point in an atomizing gas stream. In the present work, the wire arc spray gun was a Value Arc (Sulzer Metco Dewsbury, NY) used with a converging nozzle. Wire arc spray process parameters were tabulated in Table 1.

Table 1: Wire arc spray parameters

Gun	Value arc
Feed rate	82 g/min
Spray distance	100 mm
Current	200 A
Voltage	33V

Commercially available Copper anode sheet with 99.99 % purity according to ASTM-B115 standard was used for comparison studies. Stainless steel 316 also was utilized as a reference. All samples were cut into 10×10 mm^2 pieces with 1 mm thickness.

Characterization of Surfaces

The phase composition of the coating surface were analyzed using a X pert Philips XRD instrument with Cu Kα (λ=1.542 nm) radiation. The XRD patterns were recorded in the 2θ range of 20-80° (step size: 0.05° and time per step: 1 s).

The etchant (1.50 mL HNO_3, 0.5 g $AgNO_3$ and 50 mL H_2O) was used to reveal the microstructure of the copper coating and commercially copper anode (Ref 20). Microstructural observations by scanning electron microscopy (SEM, Philips XL 30) were performed on the polished sample cross-sections for the coating thickness and porosity evaluation. Energy dispersive spectroscopy (EDS) was also used to examine the composition along the coating thickness.

Antibacterial Behavior

The antibacterial activity of the copper coatings, copper anode and stainless steel sheets were tested using a standard procedure based on the Japanese JIS Z 2801:2000 spread plate method (Ref 17). Two bacterial species, gram negative Escherichia coli NCTC 10418 and gram positive Staphylococcus aureus NCTC 11047 were selected as tests bacteria. The bacteria were cultured in Tryptone soya agar (Oxoid, UK) overnight and then diluted in Tryptone soya broth (Oxoid, UK) to an optical density OD600 nm of 0.05, which is equivalent to 107 cells ml-1. Bacterial suspension with 10^5 cell/ml was prepared. Samples were sterilized by autoclaving, following by placing on sterile filter papers saturated with distilled water inside sterile 90 mm diameter Petri dishes to maintain ambient humidity, 20 ml of the diluted bacterial suspension was pipette onto each sample, which was then covered with a sterile glass cover slip, in order to maintain the same contact area of suspension on each tested sample surface. All Petri dishes were incubated at room temperature for the designated duration. Times of 60, 120, 240 and 360 min were used to monitor the relation between the percentage of cells killed and contact time (reduction rate). After the designated time the sample was transferred to 10 ml of sterile phosphate-buffered saline (PBS) (Sigma) in a sterile container. The contents were vortex mixed for 10 s to dislodge the cover slips and suspend the surviving bacteria in the PBS. Serial dilutions of the bacterial suspension were made and 100 µl aliquots of each dilution pipetted onto Tryptone soya agar plates, which were incubated overnight at 37 ^{0}C. The number of colony forming units (CFU) resulting from the growth of the viable bacterial at 37 ^{0}C after 24 h represents the initial viability of bacteria that survived in the suspension. The percentage reduction was calculated according to the following equation:

$$\text{reduction\%}\ [(N_0 – N_R)/N_0] \times 100 = (\text{CFU ml}^{-1}) \qquad \text{(Eq 1)}$$

where Nt is the mean CFU ml^{-1} from a test sample after a designated contact time and N0 is the mean CFU ml^{-1} for the same material sample at time zero. Three specimens of each test material and the controls were analyzed and three plates were spread from the PBS suspension resulting from each sample. Results are expressed as means ± SD of the measurements.

Results and Discussion

Investigation of X-Ray Diffraction test

The XRD pattern of the copper coatings is shown in Fig. 1. Main phase composition in the XRD pattern is contributed to pure copper (shown with stars). Some minor peaks belong to copper dioxide (shown with circle) can also be detected.

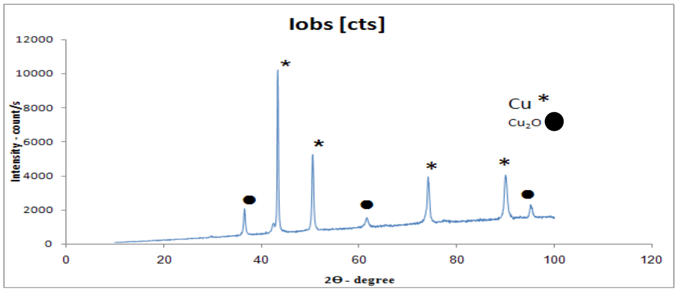

Figure 1: X-ray diffraction pattern from the surface of the copper coating.

The existing of copper dioxide is due to oxidation of the molten copper particles during spraying by atomizing air and cooling after impact (Ref 18).

Microstructural Analysis

Microstructure of the commercial copper anode is shown in Fig. 2a with average grain size around 30µm, which could be defined as a micro grain size structure. The coating microstructure presented in with good interface and adhesion bonding. The thickness of the copper coating was around 500 µm.

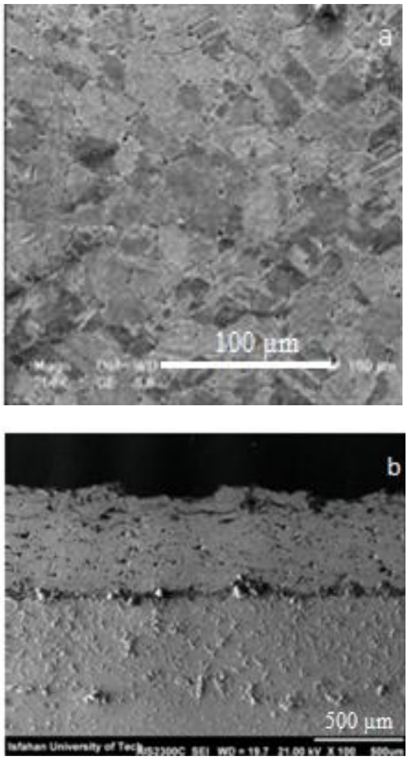

Figure 2: SEM micrographs from cross section of (a) commercial copper anode (b) Wire arc copper coating.

The etched copper coating microstructure is shown in Fig. 3a and 3b. The grains columnar structure inside of the individual splats indicated the heat transfer direction during solidification which was perpendicular to the substrate. The average grain width measured around 260 nm by image analysis software (Ref 19). It can be noted that there are some pores in the copper coating, especially between splats.

Surface morphology of the copper coating is illustrated in Fig. 4. The diameter of splats was estimated to be around 100 µm. One of the features of wire arc spray is that most of the sprayed particles are molten completely, resulting in formation of this sort of surface morphology.

Evaluation of Oxidation during Deposition Process

Thin oxide layers (mainly, copper oxides) were formed between individual splats inside of the copper coating as results of EDS mapping show in Fig. 5. Our studies have shown that decrease in particle size leads to increase of the level of its oxidation. The smaller particles have oxides distributed over the entire particle whereas with increase in particle size, it is found that the particle develops an oxide coating or shell. Thus, the oxidation of an in-flight particle proceeds from particle surface towards the center. Oxide inclusions outlining the grain or splat boundaries have also been noted (Ref 22).The particle with more surface oxidation was observed in a wire arc sprayed deposits.

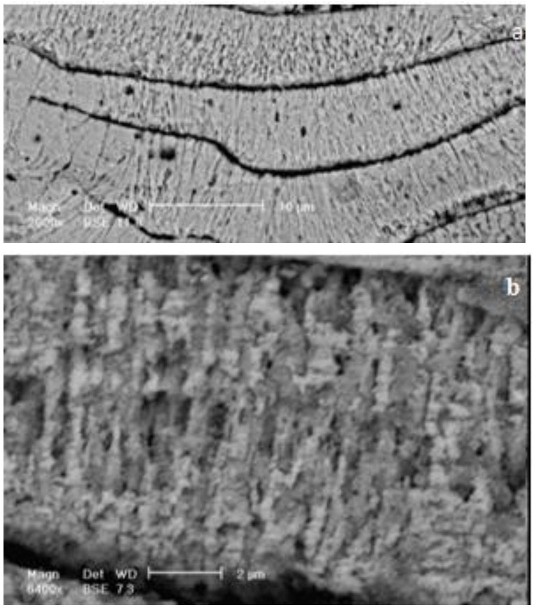

Figure 3: SEM micrograph of (a) microstructure of splats in copper coating (b) microstructure of grains in copper coating.

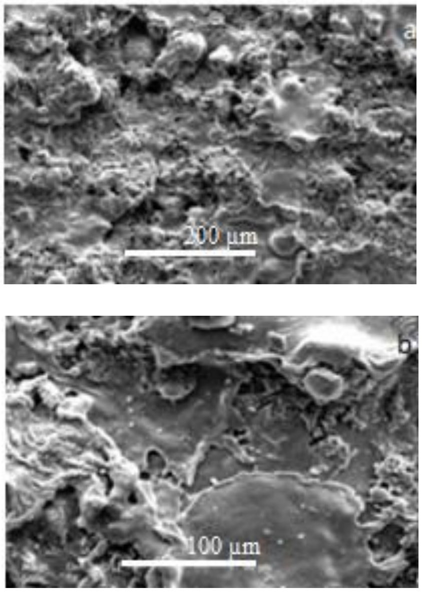

Figure 4: SEM micrographs of surface morphology of copper coating with two different magnifications (a) 125X (b) 500X.

As Fig. 6 illustrates, an EDS line scan through about 14 µm of the cross section of the as-sprayed structure which consist of five individual splats, verifies the distribution of oxygen in the lamellae boundaries. The coating microstructure in Fig. 6a shows splats on top of each other separated by alternating oxide layers.

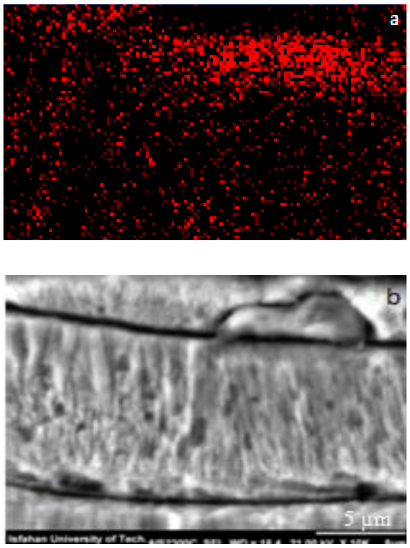

Figure 5: EDS mapping of as-sprayed copper coating showing oxide layer; (a) oxygen mapping and (b) SEM image.

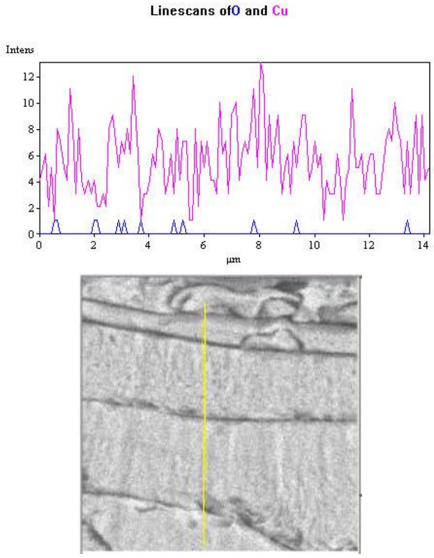

Figure 6: Line scan from a cross section of the as-sprayed copper coating.

Antibacterial Susceptibility Evaluation

Reduction in bacterial numbers of E. coli NCTC 10418 and Staphylococcus aureus NCTC 11047 after contact with the three different surfaces is indicated in Fig. 7. E. coli NCTC 10418 was more sensitive to the inhibitory action of the copper coating (97% after 6 h), whereas S. aureus needs longer time to reach a similar percentage of reduction. The killing rate for stainless steel is in the range 8–24 h. In comparison with the copper anode, antibacterial activity of the copper coating was much better than that of copper sheet viable bacteria on the copper coating have been killed in a shorter period of time. The mechanism of the copper toxicity to bacteria remains to be fully understood, but it has been reported that it associated with a bacteria interaction with protein thiol groups. More recently Lansdown et al. (Ref 20) demonstrated that silver binds to bacterial DNA and RNA and inhibits bacterial replication. These interactions could explain the non-selective biocidal activity of copper on E. coli and S. aureus.

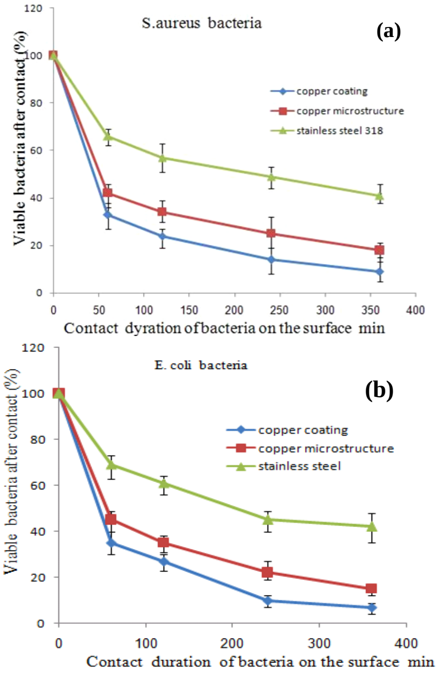

Figure 7: Reduction in bacterial numbers after contact with the surfaces (a) S. aureus NCTC 11047 (b) E. coli NCTc 10418.

Defects of the coating structure such as incoherent twin boundaries, micro pores and micro cracks are energetically favorable as segregation points for solute atoms such as oxygen. More importantly, these sites contribute to differences in energy states on the copper coating and, hence, provide the necessary driving force for sustained dissolution of metal ions from the surface. The physical nature of the copper crystallites, in the form of nanocrystals, and their association with Cu–O super oxides and defects acting as preferential dissolution sites in aqueous media, are crucial to providing enhanced antibacterial activity (Ref 25).

Summary and Conclusion

Copper coatings were successfully deposited on stainless steel substrate by wire arc spraying for antibacterial applications. The coating microstructure showed splats on top of each other separated by alternating oxide layers. Viable bacteria on the copper coating have been killed in a shorter period of time in comparison to copper sheet and stainless steel 316. E. coli NCTC 10418 was more sensitive to the inhibitory action of the copper coating (97% after 6 h), whereas S. aureus needs longer time to reach a similar percentage of reduction. In summary, the results presented in this paper suggested that the bactericide properties of the twin wire arc sprayed copper coating could be function of the coating grain size and lattice defects, as well as the presence of specific oxygen species.

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