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Wet Biochemical Synthesis of Copper Oxide Nanoparticles Coated on Titanium Dental Implants

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ABSTRACT: Titanium-based materials have been used for dental implants due to their excellent biological compatibility, superior mechanical strength and high corrosion resistance. The osseointegration of titanium dental implants is related to their composition and surface treatment. A better anti-bacterial performance of the abutment seated in the prosthetic crown is beneficial for the osseointegration and for avoiding the microbial infection after implantation surgery. Copper oxide nanoparticles are normally considered to be of a size no greater than 100 nm, and the exploitation of their unique attributes to combat infection has increased markedly over the past decade. The potential of copper oxide nanoparticles to control the formation of bio films within the oral cavity, as a function of their biocidal, anti-adhesive, and delivery capabilities, is now coming under close scrutiny. The latest insights into the application of copper oxide nanoparticles in the control of oral infections. This study was focused on importance of copper oxide nanoparticles as an antimicrobial agent. In present study copper oxide nanoparticles coated dental implants were studied by using Standard slurry dipping technique and wet chemical synthesis. Coating titanium surfaces of dental implants with copper oxide nanoparticles should lead to an increased rate of implant success.

KEYWORDS: Copper oxide, Nanoparticles, Anti-adhesive, Bio film, Titanium.

I. INTRODUCTION

For centuries, people have used copper for its antibacterial qualities. However, copper nanoparticles have showed antibacterial activities more than copper. Minuscule amounts of copper nanoparticles can lend antimicrobial effects to hundreds of square meters of its host material. This is more effective and proved to have antibacterial activity in the formulation of micro scale to nano scale sized particles. Also Guanog Ren said that the copper nanoparticles have widespread antibacterial activity against *E.coli* and *S.aureus*. Copperhas potent biocidal properties. Copper ions, either alone or in copper complexes, have been used for centuries to disinfect liquids, solids and human tissue. This manuscript reviews the biocidal mechanisms of copper and the current usages of copper and copper compounds as antibacterial, antifungal and antiviral agents, with emphasis on novel health related applications. Dental implants are manufactured from titanium, a metal that is known to be compatible with body tissues and able to bond with adjacent bone during healing. There is a now a wealth of evidence from studies and from clinical experience to show that dental implants are a safe and convenient way to replace lost teeth with natural-looking results. Pure titanium (Ti) is commonly used as artificial joints and implants in both dental clinics because of its biocompatibility and mechanical properties. The excellent biocompatibility of titanium surfaces as an implant materials results from its surface properties [4]. While problems in the osseous healing of implants appear to be resolved, the adsorption of bimolecular pellicles and the subsequent accumulation of bacteria on these surfaces are still the main stimulus for the induction of inflammatory processes. Although the biocompatibility of Ti has been confirmed it is still difficult to meet all the requirements, such as antibacterial ability, biocompatibility, osseointegration, and mechanical properties [14]. Good biocompatibility and rapid osseointegration are essential factors of prolonged stability of the implant. Especially for dental implants, some of the studies indicated that both the quality and quantity of plaque adhesives on the implant abutment surface, which contact with gingival tissue, influence the long-term implant success [6]. The initial adhesion

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and the colonization of bacteria to an implant surface are considered to play a key role in the pathogenesis of infections related biomaterials [5]. Because Ti does not exhibit antimicrobial properties, one approach to achieve better disinfection with biocompatibility is to modify the surface material of the Ti-based implant [9]. Bacterial attachment plays a significant role in determining the outcome and success of a Ti-based implant [7]. Therefore, surface modification of titanium by coating or adding antibacterial properties of metals or alloys to reduce the number of bacteria and microbial adhesion seems an efficient way to increase the benefit of clinical treatment. An in vitro study by B. Groessner-Schreiber et al showed that TiN and ZrN coatings possessed antibacterial performance to the oral microflora and Streptococcus [13]. Ag and Cu are known to be efficient antibacterial agents because of their specific antimicrobial activity and nontoxicity of the active Ag and Cu ions to human cells [10]. Recently, it was reported that Ag-doped TaN and Cu-doped TaN with nanoparticles can decrease the multiplication of Escherichia coli bacteria, and showed improved antibacterial effect [12]. In addition, Ta alloys are known to have excellent biocompatibility which makes TaN an excellent protective coatings in biomedical applications [3]. Ag, as a doping element, is proved not miscible with TaN, which makes the synthesis of TaN-Ag nano composite coatings possible. Even though TaN-Ag has been confirmed that it can improve the antibacterial efficiency of *E. coli*, no study has been investigated the effects on the bacterial ability when TaN-Ag is used as a coating material for dental or even orthopaedic implants. In this study, copper oxide nanoparticles coatings with titanium dental implants to control the dental infections. The objective of this work is to study wet biochemical synthesis of copper oxide nanoparticles coated on titanium dental implants for the dental applications. *Staphylococcus aureus* and *E.coli*, a major pathogen frequently found in the percutaneous and dental implant-associated infections was chosen as the model for this in vitro study.

II. MATERIALS AND METHODS

The bacterial test cultures *Staphylococcus aureus* and *E.coli* were isolated from the biological specimens collected from a diagnostic laboratory at Coimbatore, Tamil Nadu, and India.

Synthesis of copper oxide nanoparticles

Chemical reduction method:

About 6.9 g of copper sulphate pentahydarate was dissolved in 100 ml of distilled water (Solution-1). Solution-2 was prepared by dissolving 34.6 g of sodium potassium tartarate and 12 g of sodium hydroxide in 100 ml of distilled water. About 50ml of solution-1 and 50 ml of solution-2 was mixed together with vigorous stirring. Reducing agent, glucose of about 5 g was added to the mixture was stirred for 10min. The mixture was kept in a boiling water bath at 60°C for 10min. The final mixture was centrifuged and washed with distilled water and ethanol twice at regular intervals. The solution was air dried to obtain a powdery substance for further analysis [8].

Preparation of antibacterial drug and coating titanium dental implants by slurry-dipping technique

The technique started with the preparation of stable slurry with specific amount of copper oxide nanoparticles in molten poly ethylene glycol. Polyethylene glycol 2g mixed with copper oxide nanoparticles in glass vial. The mixture was heated in a water bath to obtain homogenous slurry. The resulting slurry was homogenized in a magnetic stirrer for 5 to 10 min. Each piece of dental implant alloys was dip coated twice with intermediate dying (suspension coating method) in the copper nanoparticles slurry mixture. The dip coat procedure was carried out in sterile glass beaker on a shaker 120 rpm for 30 minutes. With a drying period of about 15 minutes the two coating procedure was carried out under aseptic condition. Subsequently in order to slow down the release rate of copper oxide nanoparticles from poly ethylene glycol coating and mitigate friction effect between implant surface and mucosa. Second layer was formed on the implant surface. PVA was dissolved in DMSO to acquire a 10w/w % solution. PEG coated sample were submerged into PVA solution three times for 1 minute each. And they are stored in 0°C in the deep freezer to implement one freeze than cycle and physically crosslink the sample. Then it is dried in room temperature to remove residual DMSO. The antibacterial activity of this coated implant was checked against the dental micro organism which is grown in the nutrient agar medium [1].

Assessing the qualitative antibacterial activity of titanium dental implants

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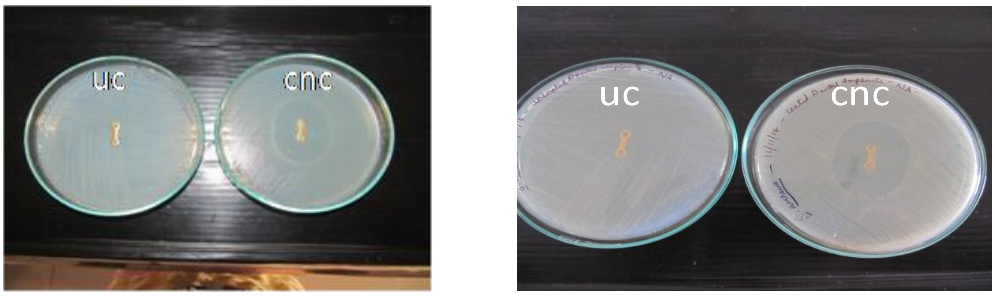
The method was performed for analysing the antibacterial activity of IVCs after slurry dip-coating with copper oxide nanoparticles and polyethylene glycol. In this qualitative method, the titanium dental implants were tested from each preparation [copper oxide nanoparticles coated and uncoated titanium dental implants]. The titanium dental implants were all rinsed twice in phosphate buffered saline (PBS) before testing to remove any surface accumulation of copper oxide nanoparticles. All titanium dental implants were placed on the surface of Mueller-Hinton agar (MHA) plate which had previously been seeded with an overnight broth culture of the test organisms and incubated at 37°c for 24 to 48h. The experiments were carried out in triplicate. Anti bacterial activity was expressed as the diameter of the zone of inhibition [11].

III. RESULTS AND DISCUSSION

The diffusing ability of the antibacterial drugs from the copper oxide nanoparticles coated titanium dental implants to retard the growth of test bacteria seeded on MHA plate was calculated based on the zone of inhibition. The zone of inhibition measured in millimetres for each copper oxide nanoparticles combinations (tested in triplicates) were calculated to obtain the mean value. In Table-1, reveal the antibacterial activity of copper oxide nanoparticles coated titanium dental implants for all the test organisms. No inhibitory zones were observed for all uncoated materials. In contrast, all the copper oxide nanoparticles coated titanium dental implants showed significant inhibitory zones ranged from 31.6mm to 38.6mm against all the test organisms. The antibacterial activity of Ti has been somewhat controversial. While some studies had shown no influence of Ti on various oral bacteria in vitro, others had found some antibacterial activity [17], [18]. Ti has also been suggested to have antimicrobial and anti-inflammatory effects due to the formation of peroxides at the Ti surface in vitro [19]. A previous study by X. Wang et al. showed that smooth Ti possessed low *S. aureus* bacterial adherence that resulted in low probability of infection [16]. The antibacterial action of Ag may be explained that Ag nanoparticles on the surface of TaN-Ag coatings attached to the bacteria and resulted in bacterial wall pitting. The inhibitory effect of Ag ions, released from Ag nanoparticles under a complex physiological condition, is believed to be due to its sorption to the negatively charged bacterial cell wall, deactivating cellular enzymes, disrupting membrane permeability, and finally leading to bacterialysis and death [20] of dental implants. The biocompatibility of Ti is attributed to surface oxide spontaneously forming in air and/or other surface treatments (e.g. thermal oxidation or anodic oxidation). It is believed that the cellular behaviour including proliferation, adhesion, and spreading is greatly influenced by this oxide layer of Ti [15]. In the dental use of Ti implants, where the implant components, such as abutments, contact not only bone but also the gingival, and are partially exposed to the oral cavity that includes bacteria, it is especially important to fabricate a coating material with both antimicrobial capacity and biocompatibility so as to increase the likelihood of implant success. TaN-Ag nanocomposite coatings possessed good HGF cell viability and improved the antibacterial effect to *S. aureus* for dental implant applications.

Table-1: The qualitative antibacterial activity of dip-coated Titanium dental implants

S .no	Sample	Organisms	Zone of inhibition (mm)	
			Coated titanium dental implant	Uncoated titanium dental implant
1.	Titanium dental implants	*Staphylococcus aureus*	38.6	0
2.		*E. coli*	31.6	0



(a) (b)

Fig.1. (a) *E.coli* (b) *Staphylococcus aureus* (cnc: copper nanoparticles coated samples, uc: uncoated samples).

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IV. CONCLUSION

Dental infection is an *infection* that originates within a *tooth* or in the closely surrounding tissues, which is caused by bacteria easily. Prevention of dental infection is one of the significant scientific efforts. In this study one such approach was reported to prevent the dental infection. Qualitative antibacterial activity of copper oxide nanoparticles coated titanium dental implants showed promising results against the entire varied test cultures used in the study. Thus the obtained results showed that copper oxide nanoparticles can hold promise for the chance of suppression of dental infections. Copper oxide nanoparticles analysis from the titanium dental implants surface and the biocompatibility of the surface modified titanium dental implants need to be assessed for the approval in human trails.

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