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Antimicrobial Surfaces from Incorporated Nano-agents

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Abstract: Biofilm development is a survival strategy for majority of bacteria to adapt to their living environment. These biofilms are prevalent on most surfaces in nature. Microbial cells in biofilm exhibit significantly enhanced tolerance and resistance to antimicrobial challenge and host defenses. Therefore, the control and eradication of biofilm-associated diseases present a great challenge. Recently, there is considerable biomedical incentive in the development of nanoparticles with self-antibacterial activity or as antibiotic carriers. The advantages of coatings incorporating different types of nano-agents for antimicrobial surface generation are here reviewed. Although the main coatings currently used or investigated and the associated synthesis processes are individually described, emphasis is made on silver-loaded mesoporous thin coatings. Recent developments suggest that mesoporous oxide thin films (especially titania) incorporating metal nanoparticles is becoming a prime candidate for antimicrobial coatings.

Keywords: Antimicrobial coating, self-antibacterial nanoparticles, antibiotic carriers, nanocomposite, mesoporous films, biofilms.

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1. INTRODUCTION

The vast majority of microorganisms on Earth live within a self-produced matrix consisting of a mixture of polymeric compounds, primarily polysaccharides, generally referred to as extracellular polymeric substance. Bacteria can develop these biofilms on a great number of different surfaces, such as natural aquatic and soil environments, living tissues, medical devices or industrial or potable water piping systems [1]. Within biofilms microorganisms are generally well-protected against the influence of environmental stresses [2]. This is why the formation of biofilms in natural and industrial environments allows bacteria to develop resistance to bacteriophages, chemically biocides and antibiotics [3] and as a consequence biofilms are extremely difficult to eradicate. As such, they cause major problems in medicine, agriculture, (food) industry and the household environment. Many studies approach strategies to eliminate or reduce biofilm formation has been encouraged as a result of the significant resistant of biofilms to conventional antibiotic therapy or to disinfectants [5].

Antimicrobial coatings are of great interest to prevent alterations or damage on diverse surfaces, for the fabrication of antimicrobial packaging films, in finishing of textiles to protect from odor-generating microorganisms and in variety of biomedical applications like implants and wound dressing. A way to generate this antimicrobial surface is to incorporate antimicrobial agents. There are many different antimicrobial agents, and there are several classifications in the literature according to the nature, mechanism of antimicrobial activity and efficiency, among others. These antibiotic agents can be incorporated to surfaces in many ways.

Nanoparticles offer unique chemical, physical, electrical properties and biomedical implications activity determined by their size, shape, composition and crystallinity. In this context, a variety of novels methods have been developed to synthesize nanomaterials in predictable and reproducible shapes and sizes, including spheres, pyramids, cubes and rods. In the last years, the synthesis of nanoparticles with self-antibacterial activity has received greatly attention. These nanoparticles exhibit the ability to kill bacteria themselves, derived from its reactive oxygen species generation although some are effective due to their physical structure and metal ion release [6, 7]. On the other hand, the development of nanoparticles as drug carriers in which an antibiotic is loaded and released from the nanoparticle and intercepts bacteria in the vicinity was also widely explored [8]. With this in mind, one novel way to incorporate antibiotic agents to surfaces consists of using these type of nanoparticles that present antibacterial activity themselves or as drug carriers. The incorporation of these nanoparticles into surfaces can bring many additional advantages like improvements in the washing resistance, the release control and the biocompatibility; it can also improve the material desired degradability or stability, together with biomechanical and physical properties. In this mini-review,

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we present an overview of different syntheses of coatings with incorporated antimicrobial nanoparticles and their antibacterial activities, emphasizing on silver nanoparticle-loaded mesoporous oxide coatings.

2. COATINGS INCORPORATING NANOPARTICLES WITH ANTIMICROBIAL DRUGS

The use of polymer nanocomposite materials where the drugs are associated with nanoparticles dispersed in the polymer matrix has attracted increasing attention and, in the case of antimicrobial nanocomposites, they also include an antibiotic or antifungal drug attached to the nanoparticles. These polymer nanocomposites are being intensively used as antimicrobial materials for the development of implants which are able to prevent post-operative infections. A common characteristic of the antimicrobial nanocomposites summarized in this mini-review is that they all present a long term and controlled release of the entrapped drugs compared to the nanoparticles alone or the polymer matrix without any filler. Moreover, in most cases these nanocomposites present lower toxicity towards different mammalian cell cultures.

Nanosize Hydroxyapatite (HA) results very popular in the field of biomaterials, principally for application in bone regeneration. As an evolution towards antimicrobial medical coating, HA nanoparticles in a crosslinked-gelatin matrix as a porous drug-delivery carrier for tissue-regeneration and wound-healing treatments was addressed. The obtained bodies were crosslinked with carbodiimide derivatives to retain chemical and thermal integrity. In this case, tetracycline was entrapped within the scaffold, and the drug-release profile was examined showing the gelatin-HA nanocomposites had lower drug releases compared to pure gelatin because of their lower water uptake and degradation. All the nanocomposite scaffolds released drugs in proportion to the initial drug addition, suggesting their capacity to deliver drugs in a controlled manner [9]. In a recent example, nano-HA particles as carriers for the antimicrobial drug vancomycin and the mixture of oxidized sodium alginate and gelatin as the polymer matrix where fabricated. In this work, the antibiotic vancomycin was loaded efficiently on the surface of these particles and could be released slowly, which endowed the composite with a continuous antimicrobial performance [10]. In addition to nano-HA, there are different types of nanoparticles which can also be used as fillers for the synthesis of nanocomposites. Among them, silica nanoparticles can be mentioned. These nanoparticles were successfully incorporated in a silica–collagen type I nanocomposite hydrogel in a single step to be used as medicated dressings to prevent infection in chronic wounds. The loading process of two antibiotics, gentamicin and rifamycin, was performed using a single step procedure allowing the easy tuning of the particle size, the drug content and, ultimately, the dose and kinetics of the antibiotic release from the nanocomposites over 1 week (Fig. **1**). Gentamicin was incorporated with high efficiency while rifamycin showed low yields as their electrostatic interaction with the negatively charged nanoparticles was different. The gentamycin-loaded particles could be immobilized at high silica dose in concentrated collagen hydrogels without modifying their fibrillar structure or impacting on their rheological behavior and increasing their stability to proteolysis. Gentamicin release from the nanocomposites was sustained over 7 days and particle immobilization also decreased their cytotoxicity towards surface-seeded fibroblast cells. The synergy between the two components was evidenced by the concomitant increase in hydrogel mechanical and proteolytic stability and decrease in silica particle cytotoxicity towards fibroblast cells. The complex interplay of interactions between drugs, silica and collagen is a key factor regulating the properties of these composite hydrogels as antibiotic-delivering biological dressings and must be taken into account for future extension to other wound healing agents [11].

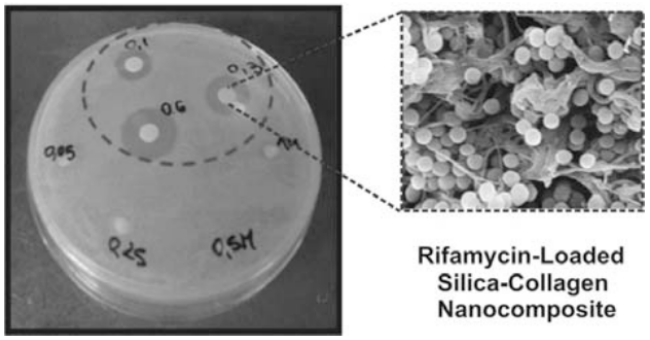

Fig. (1). Disk diffusion method evaluation of the antibacterial activity of antibiotic-loaded SiNPs silica–collagen nanocomposites after 1 day. (Reproduced with permission from Royal Society of Chemistry [11]).

The combination of the aforementioned nanoparticles can also be found in literature. For example, a mesoporous silica–hydroxyapatite composite material was used as drug vehicle for polymer matrices composed of poly(latic-co-glycolic acid) (PLGA) and microspheres sintering techniques. Drug delivery testing of these nanocomposites indicated a slow gentamycin release from the particles, which lasted for nearly one month. In addition, the scaffolds demonstrated low cytotoxicity, and supported mesenchymal stem cells growth more effectively than pure PLGA scaffolds [12].

Calcium phosphate ceramics can be mentioned as they are analogous to the mineral component of bones, and therefore their biocompatibility and osteoconductive properties make them desirable as implant materials. Nanoporous calcium phosphate ceramics with well-defined rod-like morphology can be obtained based on filling of calcium and phosphorus source into the nanospaces of rod-like ordered mesoporous carbon (CMK-3) and subsequent removal of carbon templates. As antibiotic carriers, these nanoporous bioceramics showed much higher charging capacity than that of commercially available bulk bioceramics without nanoporosity [13]. Alternatively, a composite material composed of a porous β-tricalcium phosphate ceramic and biodegradable polymers (poly-ε-caprolactone and polyethylene glycol) loaded with gatifloxacine for treating chronic osteomyelitis was developed. The composite was fabricated using a solvent-free process in which no toxic solvent was used, and

characterized in terms of its *in vitro* antibiotic release efficiency, bactericidal property as well as *in vivo* effectiveness and safety. This composite showed efficacy in controlling infection at the bone defect formed by debridement, and supported bone tissue reconstruction at the bone defect [14].

In a separate but related thread, magnetic nanoparticles and the drug Nystatin were included in a composite with chitosan which was highly active against *Pseudomonas aeruginosa*, *Candida albicans* and *Escherichia coli* [15]. Bovine serum albumin or polyallylamine nanoparticles adsorbing high cefamandole nafate amounts owing to their high surface/volume ratio were incorporated in a polymer matrix of carboxylated polyurethane to release the drug in a control manner. For these systems the drug delivery was at least of 50% with respect to nanoparticles alone with a prolonged antimicrobial activity up to 9 days [16].

As an interest alternative, chitosan nanoparticles loaded with ciprofloxacin were applied as a coating onto titanium substrates to obtain an orthopaedic implant surface able to *in situ* release the antibiotic for the prevention of post-operative infections. The novelty of this system was the incorporation of two cyclodextrins (CD), which are both approved by FDA for parenteral administration, a negatively-charged CD was employed to crosslink the positively-charged chitosan whereas an uncharged CD was used to increase drug loading in the chitosan network [17].

3. MATERIALS INCORPORATING SELF-ANTIMICROBIAL NANOPARTICLES

For many thousands of years, various metals, mainly silver and cooper, have been used owing to their antimicrobial properties. The existence of metal resistant bacteria reported since the 1960s reflects the human antimicrobial use in infectious disease and microbial exposure to these metals from industry and agriculture. The mechanisms of resistant include: efflux systems; alteration of solubility and toxicity by changes in the redox state of the metal ions; extracellular complexation or precipitation of metals; and the lack of specific metal transport systems. Bacteria have specific genes for resistances to the toxic ions of heavy metal elements including Ag^{+}, Cd^{2+}, Co^{2+}, Cu^{2+}, Hg^{2+} and Ni^{2+}. Mostly, these resistance systems have been found on plasmids, but frequently related systems are subsequently found determined by chromosomal genes [18].

Silver and copper nanomaterials, with the above reservations, show even stronger antibacterial properties since their small particle size and high specific surface area enable a large increase in the metal ions released from the particle surface [19]. In addition, these metal nanoparticles show great promise in having likewise efficacy against the most resistant spores [20]. Ag-nanoparticles are particularly attractive as these are non-toxic to human body at low concentrations and have a broad-spectrum antibacterial nature. Ag nanoparticles inhibit bacterial growth at very lower concentration than antibiotics. These nanostructured metals have been successfully incorporated into polymer matrices or hydrogel scaffolds. The nanocomposite networks in which nanoparticles are embedded, control the release of metal ions and improve the release time and resistance of the material.

As an example of antibacterial coating from a polymer including nanoparticles, an *in situ* synthesis of oil-based polymer-silver nanocomposites was achieved by using photoinduced free radical polymerization processes in which silver nanoparticles were formed by electron transfer reaction. An oil-based macromonomer was prepared and then copolymerized with styrene in the presence of $AgNO_3$. Copolymerization was started with free radicals formed by photolysis of 2,2-dimethoxy-2-phenyl acetophenone and simultaneously silver nitrate was reduced to metallic silver in nanosize by electron transfer reaction. The obtained polymer nanocomposite film was examined in respect of surface coating material that would have an antibacterial effect against Gram-positive, Gram-negative and spore-forming bacteria. It was demonstrated that suitably coated nanocomposite samples exhibited an antibacterial effect against these bacteria [21].

Hydrogels based on crosslinked poly(acrylamide) and carbohydrates, polyvinylalcohol in combination with the crosslinking agent polyvinyl pyridine has been studied [19]. Hydrogels based on polymers of alginate, poly(vinyl alcohol) (PVA) and poly(N-vinyl-2-pyrrolidone) (PVP) that can act as efficient donors of silver nanoparticles and silver ions has been studied for biomedical applications. As a practical alternative, silver nanoparticles-alginate nanocomposites on cotton were synthesized using a one-step synthetic route by reduction of silver nitrate using alkali hydrolyzed alginate solution that acts as reducing and capping agent. The treated cotton demonstrated an excellent antibacterial activity against the tested bacteria, *E. coli, S. aureus* and *P. aeruginosa*. A slight decrease in the antibacterial feature of the cotton fabrics was observed after successive washings. However, an efficient antibacterial activity still remained on the fabrics [22]. Novel wound dressing materials have attracted increasing attention, for example bacterial cellulose, with the issue of having no antimicrobial activity, which is one of critical skin-barrier functions in wound healing. To overcome such deficiency, in situ synthesis and deposition of uniform spherical (10–30 nm) silver nanoparticles into bacterial cellulose (BC) fibrous membrane has been successfully developed through the reaction of Tollens' reagent under ambient conditions (Fig. **2**). This stable hybrid nanostructure offered excellent and sustainable controllability of Ag+ release. Regardless the slow Ag+ release, Ag Nanoparticles-Bacteria Cellulose nanocomposites still exhibited significant antibacterial activities with more than 99% reductions in *E. coli, S. aureus* and *P. aeruginosa.* Moreover, these hybrid nanocomposites allowed attachment and growth of epidermal cells with no cytotoxicity emerged. The results demonstrated that Ag-nanoparticle loaded bacterial cellulose could reduce inflammation and promote wound healing [23].

Porous silica represents a valuable alternative as a matrix to host silver nanoparticles. For example, a facile and "green" method was proposed to introduce uniformly silver nanoparticles into a hierarchically porous silica monolith with an effective method by using ethanol as the solvent,

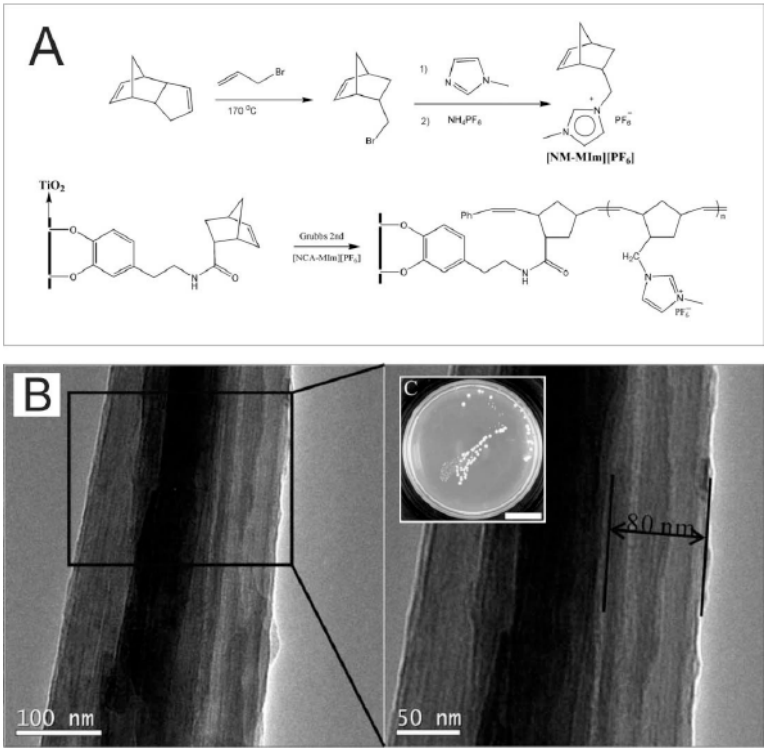

Fig. (2). (**A**) Synthetic procedure of poly(ionic liquid) brushes generated on TiO_2 nanowires (PNM-TiO_2) (**B**) Transmission electron microscopic morphology of PNM-TiO_2. (**C**) The growth inhibition of PNM-TiO_2 against S. aureus (Reproduced with permission from Royal Society of Chemistry [40]).

(3-aminopropyl)-triethoxysilane (APTES) as the modifier and ethylene glycol as the reductant [24]. APTES has also been bound to titanium dioxide, which offers antimicrobial properties derive from its photocatalytic killing of microbes, by a Ti-O-Si chemical bond. Dual antimicrobial action has also been obtained by incorporating alkyltrialkoxysilanes with pendant quaternary ammonium groups embedded with silver nanoparticles [19].

Qureshi *et al*., report a facile layer-by-layer method that can be repeated serially for creating an antimicrobial coating composed of silver nanoparticles on titanium orthopedic implant material via APTES crosslinker. Nanoscale silver nanoparticle layers were attached to the titanium orthopedic implant material via aminopropyltriethoxy silane crosslinker. A monolayer of silane, followed by a monolayer of silver nanoparticles would form one self-assembled layer and this process can be repeated serially, resulting in increased silver nanoparticles deposition. The release rate of silver ion increases with increasing numbers of layers and at appropriate thicknesses these coatings demonstrate high reduction of viable *E. coli* and *S. aureus* bacteria [25]. Silica metallic antibacterial coatings have also been synthesized by Akhavan and Ghaderi using sol-gel procedure on soda lime glass substrate. A series of hybrid titania thin films containing copper-based nanotube and nanopore oxide structures by one-pot anodizing of Ti- and TiAl-related alloys have shown strong antibacterial activity against *E. coli and S. aureus* [26] These materials have been studied in a wide range of fields. For example, in food industry, active packaging is an innovative area which helps food products to have better sensorial features and extended shelf-life, enhancing food quality and safety. To create an antibacterial food packaging films Sadeghnejada *et al*., generate a surface modification of polyethylene films by corona air plasma and coating of silver nanoparticles on the surface of treated film [27]. For biomedical application, the antibacterial property of silver oxides has also been studied. Coating endotracheal tubes with silver oxides reduces the incidence and mortality of ventilator-associated pneumonia in clinical trials. Also the use of silver alloy urinary catheters reduces the risk of nosocomial urinary tract infection [28]. Anodization has potential applications in the fields of antimicrobial materials, biomedical or implanting devices. It has shown an excellent capability in fabricating multiple kinds of oxide layers and nanostructures.

Nano-sized ZnO exhibits varying morphologies and shows significant antibacterial activity over a wide spectrum of bacterial species explored by a large body of researchers [29]. ZnO nanoparticles also cause significant inhibition in spore germination [30]. Interestingly, ZnO-NPs are reported by several studies as non-toxic to human cells [31]. A nano structured-hybrid coating formulation for corrosion and microbial prevention of mild steel based on tetra functional epoxy resin reinforced with surface functionalized nano zinc oxide was developed by Duraibabu *et al.* Zinc oxide has a photo-oxidizing and photo-catalytic ability against chemical and biological species and the activity of the nanoparticles is in part due to their electrostatic interaction with cell surfaces. Furthermore, the antimicrobial test indicated that F-ZnO-TGBAPB coating had strong antimicrobial activity against high concentration of *E. coli* (Gram-negative) bacteria [32]. Nanostructured zinc oxide, nanosilver and hybrid

nanostructured silver-zinc oxide surface decorations were studied in order to extend potential applications of wood–plastic composites in interiors with respect to hygienic requirements. Wood floor was modified by a microwave assisted solvothermal synthesis method starting from soluble precursors. They found that hybrid silver and zinc oxide combined flour have significantly higher efficiency than the single mode [33]. In textile, various approaches have been considered to introduce zinc oxide on the textile and polymer surfaces such as polyester, wool, cotton, and others. Zinc oxide had been included as nanoparticles on different substrates, including polyester, cotton fabrics, cotton-polyester and polyurethane composites through different methods such as through homogeneous phase reaction, wet chemical, and melt-spinning. For example, nano and bulk zinc oxide on cotton fabric was synthesized and applied, concluding that nano has a higher antimicrobial activity than bulk zinc oxide [34]. It has been reported a higher tensile strength on cotton and wool fabrics and more air permeability of cotton fabric treated with nano zinc oxide. The possibility that nanoparticles with small size enter to the polymer chains, leading to the generation of some bonds with them, acting similar to cross-linking agents inside the polymer structure and therefore resulting in the stiffness of the fabrics has been proposed [35]. A novel system using electrohydrodynamic atomisation (EHDA) that provides a uniform coating was employed to deposit zinc oxide nanoparticles as a coating material to inhibit bacterial adhesion and to promote osteoblast growth onto the surface of glass substrates. Also, a mixture of zinc oxide and nanohydroxyapatite was tried but the substrate coated with zinc oxide exclusively resulted more efficient in antimicrobial activity. Furthermore, osteoblast function was explored by exposing cells to nano-ZnO supernatants showed minimal toxicity [36].

Photocatalytic surfaces offer promise as innovative and cost-effective biocidal engineering solutions. In this sense, TiO_2-nanorods photocatalysts were prepared through hydrothermal synthesis. From these nanoparticles, transparent TiO_2 thin films of high quality on glass substrates were successfully fabricated by dip-coating method. The TiO_2-nanorods thin films irradiated with UV-A light exhibited high antibacterial on multiresistant *Klebsiella pneumoniae* strains [37]. Incorporation of silver to titanium dioxide is of great interest for photocatalytic bactericidal effect. The incorporation of silver to powdered Degussa P25 TiO_2 increases the antibacterial activity using *E. coli* as model microorganism [38]. As an alternative, a biocidal photocatalytic needle-like shape nanocomposite with a thickness of a few nanometers, composed of TiO_2 and multi-walled carbon nanotubes, was synthesized via wet chemistry followed by a heat treatment. Irradiating bacterial endospores (*Bacillus cereus*) with solar UV lamps in presence of this photocatalyst successfully inactivated the spores while solar UV Lamps with Degussa P25 only showed no significant inactivating behavior [39]. Polymer brushes provide an ideal platform for studying biofouling and screening anti-biofouling materials. In fact, high-density poly(ionic liquid) brushes based on imidazolium salts were successfully grafted to surfaces via surface-initiated ring-opening metathesis polymerizations from catecholic initiator and their anti-microbial activity was evaluated. It was found that poly(ionic liquid) brushes can resist adhesion of *Chlorella* spores and the counter-anions have a key impact on the anti-microbial property. The hexafluorophosphate anion poly(ionic liquid) coated TiO_2 nanoparticles have excellent anti-bacterial properties compared to pristine TiO_2 nanoparticles against both *E. coli* and *S. aureus* under both light irradiation and dark condition [40].

4. SILVER NANOPARTICLES-LOADED MESOPOROUS THIN COATING

Offering highly controlled confined spaces mesoporous materials are excellent host matrices for nanoparticles [41, 42]. In particular, mesopores constitute highly controlled nanoreactors for nanoparticles. The size-dependent nanoparticle properties, tuned by pore size, combined with an accessible and well-defined mesopore system, and the transparency and optimal transport properties of mesoporous thin films present an enormous potential in fields such as biomedicine.

In situ-dispersed Ag nanoparticles on a mesoporous $Al(OH)_3$ layer were fabricated by alkali surface modification. By means of the polyol process, *in situ* dispersion of the Ag nanoparticles was achieved on the mesoporous layer without using additional immobilizers. Among various substrates, the $Ag/Al(OH)_3$ mesoporous nanocomposite film showed excellent antibacterial properties [43].

In another study, transparent bilayered SiO_2/TiO_2 mesoporous coatings doped with silver nanoparticles were obtained by a sol–gel technique. Silver nanoparticles were alternatively embedded in SiO_2 or TiO_2 layers. All samples show good photocatalytic and bactericidal activities and both properties are enhanced by doping with Ag nanoparticles (Fig. **3**). All the coatings showed a strong bactericidal activity against planktonic forms of Gram-negative and Gram-possitive pathogens, as well as a strong germicidal effect against deadly spores of human gas gangrene- and anthrax-producing bacteria. The bactericidal and sporocidal activity was improved by doping the coatings with Ag nanoparticles. Further strengthening of the bactericidal properties was obtained when the Ag nanoparticles were placed into the TiO_2 layer, because they are more accessible to the environment. The increase in bactericidal activity was derived from the release of silver ions into the medium [44].

Mesoporous TiO_2 thin films of polycrystalline anatase containing silver nanoparticles were also prepared by the template sol–gel method on silicon substrates. Silver nanoparticles were uniformly distributed and strongly attached to the mesoporous TiO_2 matrix. The morphology of the composite film could be controlled by simply tuning the molar ratio of the silver nitrate aqueous solution. These composite films display excellent antibacterial activity. A similar strong antimicrobial property was observed even after the composite films were stored for long periods [45].

The antibacterial activity of self-accumulated Ag nanoparticles on mesoporous TiO_2 thin film was investigated against *E. coli* bacteria, in dark and also under exposure to UV light. Accumulation of the silver nanoparticles on the

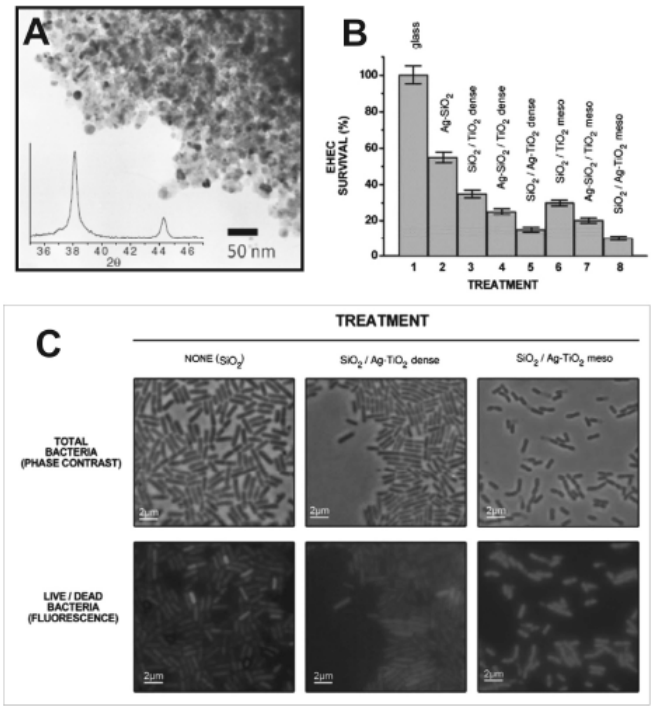

Fig. (3). (**A**) TEM image of mesoporous Ag-doped films, inset) GXRD patterns (**B**) Bactericidal activity of Ag and/or TiO_2 films against bacteria under UV irradiation. (**C**) Phase contrast and fluorescent microscopy of bacterial before and after treatments with SiO_2/Ag–TiO_2 composites. (Reproduced with permission from Elsevier [44]).

film surface let the mesoporous Ag–TiO_2 nanocomposite thin films have excellent antibacterial activity. The behavior of silver ion release showed that the dominant mechanism of the release process in long time was based on water diffusion through the capillary mesoporous of the TiO_2 layer, unlike the usual diffusion of water on the surface of silver-based bulk materials [46]. In other work, aligned and compact Ag nanorods capped by sol–gel mesoporous TiO_2 layer were grown on Ti/Si(100) by applying an electric field perpendicular to the surface of the Ag/Ti/Si thin film while it was being heat-treated at 700 °C in an Ar + H_2 ambient. Antibacterial activities of the TiO_2-capped Ag nanorods against *E. coli* bacteria were examined in the dark and under UV irradiation. The Ag^+ ion release measurements showed that the mesoporous TiO_2 layer can control the silver release process by inter-diffusion of water and silver ions through its capillary structures, completely different from the ion release behavior of the silver nanorods not capped by the TiO_2 layer. The results showed a strong and lasting antibacterial activity for the TiO_2-capped Ag nanorods with 2.34 h^{-1} for the relative reduction rate of the number of viable bacteria [47]. In another methodological approach, titanium oxide thin films containing silver (Ag) nanoparticles were synthesized on commercially pure titanium substrates using a reactive magnetron co-sputtering method. The goal of this work was to maximize bactericidal activity along with sustained biocompatibility [48].

Recently, TiO_2 mesoporous thin films on Ti substrates using anodization and sintering of a paste of TiO_2 nanoparticles were successfully fabricated. Due to the highly increased surface area, large quantities of antibacterial agents could be loaded. Four kinds of agents including silver nanoparticles and various antibiotics were loaded and each one showed varying antibacterial activity against five different bacterial strains. Additionally, two agents (silver nanoparticles and minocycline) were loaded together on the thin film and tested for antibacterial effectiveness. The combination of silver nanoparticles and minocycline was found to display a wide range of effectiveness against all tested bacteria [49].

Other silver nanoparticle incorporation scheme has been developed to take advantages of the photocatalytic behavior of titania using a fast simple procedure. Optically transparent Ag/TiO_2 composite films were prepared by incorporating Ag in the pores of anatase films with an impregnation method via photoreduction. After the doping of Ag, excellent antimicrobial ability was obtained [50]. To elucidate the roles of the photocatalytic effect and the doped silver in the antimicrobial activity, cells from a silver-resistant *E. coli* stain were used. These results indicated that Ag nanoparticles in the mesoporous functioned not only as an antimicrobial but also an intensifier for photocatalysis. Firmly immobilized Ag nanoparticles on mesoporous TiO_2 surfaces were evaluated using degradation of Rhodamine B (RhB) and disinfection of *E. coli* under ultraviolet irradiation. Photocatalytic degradation of RhB and inactivation of *E. coli* effectively occurred in an analogical trend. The deposited Ag slightly decreased adsorption of target pollutants, but greatly increased adsorption of molecular oxygen with the latter enhancing production of reactive oxygen species (ROS) with concomitant increase in contaminant photodegradation. The composite photocatalysts were stable and could be used repeatedly under UV irradiation [51].

5. CONCLUSION

Any newcomer to the production scene will, for obvious reasons, have a very hard time in displacing well-established materials and technologies. Table **1** illustrates the wide variety of novel antibacterial nanocoating reported. In this sense, the efficacies of antimicrobial surfaces from incorporated nano-agents with regards to timely disinfection of spores and metals resistant bacteria, will be required to reach their fullest potential. One should not forget that, in the case of silver nanoparticles, biomedical technology profits from the wide experience base of the ancestral silver utilization on infections. Silver-based nano-coatings dominate in terms of production, but copper and zinc oxide loaded surfaces have the potential to undercut costs.

As the research volume on antimicrobial coatings from inserted bacterial active nanoparticles continues to increase, a stage will soon be reached at which the availability of raw materials, production aspect, ecological considerations, and operational reliability (rather than laboratory performance) become the prime issues in selecting and promoting a given nano-coating. It is safe to assume that Ag-TiO_2 thin-films will play an significant role in the future antimicrobial-coating market. Antibacterial coatings incorporating self-antimicrobial nanoparticles cater to a very wide range of different requirements.

Table 1. Summary and comparison of different antimicrobial nanocoatings (Nano-agent, Matrix, Antibacterial Agent, Tested bacteria, Antimicrobial Effect).

	Composition		Antibacterial Agent	Tested bacteria	Antimicrobial Effect	Reference
	Nano-agent	**Matrix**				
Coatings incorporating nanoparticles with antimicrobial drugs	HA nanoparticles	Oxidized sodium alginate and gelatin	Vancomycin	*S. aureus*	The incorporation of vancomycin produced a significant decrease of the colony count.	Yu J. *et al.* [10]
	Silica nanoparticles	Collagen type I	Gentamicin Rifamycin	*P. aeruginos, S. aureus*	Prolonged antibacterial activity.	Alvarez GS *et al.* [11]
	Porous β-tricalcium phosphate ceramic	Poly-ε-caprolactone (PCL) and polyethylene glycol.	Gatifloxacine	*S. milleri B.fragilis*	This composite showed efficacy in controlling infection at the bone defect.	Miyai T. *et al.* [14]
	Magnetic nanoparticles	Chitosan	Nystatin	*P.aeruginosa, C.albicans, E.coli*	The nanocomposite exhibited high antimicrobial activity.	Hussein-Al-Ali SH *et al*. [15]
	Bovine serum albumin or polyallylamine nanoparticles	Carboxylated polyurethane	Cefamandole nafate	*S.epidermidis*	Prolonged antimicrobial activity up to 9 days.	Crisante F *et al.* [16]
	Chitosan nanoparticles	Cyclodextrins	Ciprofloxacin	*S. aureus, P. aeruginosa*	The system inhibited the growth of the bacteria for 48 h.	Mattioli-Belmonte M *et al.* [17]
Materials incorporating self-antimicrobial nanoparticles	Ag nanoparticles	Alginate	Ag ions	*E.coli, S. aureus, P. aeruginosa*	The composite deposited on the surface of cotton fabrics exhibit excellent antibacterial activity, even after 20 washing cycles.	Zahran MK *et al.* [22]
	Ag nanoparticles	Bacterial Cellulose	Ag ions	*E.coli, S. aureus, P. aeruginosa*	The system exhibited significant antibacterial rates with more than 99% reduction against the tested bacteria.	Wu J *et al.* [23]
	Ag nanoparticles	Silica incorporating alkyltri-alkoxysilanes	Ag ions	*E.coli, S.epidermidis*	The composite exhibited high antibacterial activity toward both waterborne and airborne bacteria.	Simoncic B *et al.* [19]
	Ag nanoparticles	Titanium materials modified layer-by-layer with APTES	Ag ions	*E.coli, S. aureus*	At an appropriate thickness the coatings demonstrate high reduction of viable bacteria.	Qureshi *et al.* [25]
	Ag nanoparticles	Polyethylene films	Ag ions	*E. coli, S. aureus*	The released silver from the coated film had antimicrobial activity against both bacteria.	Sadeghnejad A *et al.* [27]
	Wood flour modified by hybrid Ag/ZnO nanostructured particles.	PVC	Ag ions and Zn	*E.coli, S. aureus*	Excellent surface antibacterial activity was experienced testifying for total bactericidal effect against both used bacterial strains.	Bazant P *et al.* [33]
	ZnO nanoparticles	Cotton fabric	Zn	*E.coli, S. aureus*	ZnO nanoparticle-coated fabric had high antibacterial efficiency and was also able to withstand the antibacterial activity after five hand washes.	Subash AA *et al.* [34]

Table (1) contd….

	Composition		Antibacterial Agent	Tested bacteria	Antimicrobial Effect	Reference
	Nano-agent	**Matrix**				
	TiO_2 nanoparticles	Carbon nanotubes	Photocatalytic effect	*B. cereus* spores	TiO_2–MWNT nanocomposites are promising for rapid deactivation of spores.	Lee S-H *et al.* [39]
Silver Nanoparticles-loaded Mesoporous thin coating	Ag nanoparticles	Mesoporous $Al(OH)_3$ layer	Ag ions	*E. coli*	The $Ag/Al(OH)_3$ mesoporous nanocomposite film showed excellent antibacterial properties.	Seo Y I *et al.* [43]
	Ag nanoparticles	Mesoporous SiO_2/TiO_2 bilayered	Ag ions Photocatalytic effect	*E. coli, L. monocytogenes, B. anthracis* spores, *C. perfringens spores*	All the TiO_2 coatings showed a strong bactericidal activity. The bactericidal and sporocidal activity was improved by doping the coatings with Ag NPs.	Roldán MV *et al.* [44]
	Ag nanoparticles	Mesoporous TiO_2 thin film	Ag ions Photocatalytic effect	*E. coli*	The composite films display excellent antibacterial activity both in the dark and under UV illumination, even after it was stored for long periods.	Yu B *et al.* [45]
	Ag nanorods	Mesoporous TiO_2 layer	Ag ions Photocatalytic effect	*E. coli*	The TiO_2-capped Ag nanorods can be applied as a lasting and strong antibacterial material with controllability of the silver ion releasing through the mesoporous and aqueous cap layer.	Akhavan O *et al.* [47]
	Ag nanoparticles	Mesoporous TiO_2 thin film	Ag ions	*S. aureus*	The magnetron sputtered Ag nanoparticle-containing titanium oxide coatings create an efficient antibacterial layer with sustained biocompatibility.	Song D-H *et al.* [48]
	Ag nanoparticles	Mesoporous TiO_2 thin film	Ag ions Minocycline	*S.aureus, P.aeruginosa A.actinomycetemcomitans Y4, P.intermedia, P.gingivalis*	The combination of silver nanoparticles and minocycline was found to display a wide range of effectiveness against all tested bacteria.	Park S W *et al.* [49]
	Ag nanoparticles	Mesoporous anatase TiO_2 thin films	Ag ions Photocatalytic effect	*E. coli*	Mesoporous anatase TiO_2 materials modified with Ag nanoparticles displayed significant capacity for the degradation of RhB and disinfection of *E. coli* under UV light irradiation	Xiong Z *et al.* [51]

One can therefore expect, especially with future growth in production and market volume, that various different antimicrobial surfaces from incorporated nano-agents will coexist, each of them dedicated to different applications. In addition, the presence of many bacterial strains in biofilms calls for the simultaneous incorporation of diverse antimicrobial nanoparticles in nanocoatings to have broad spectrum range against bacterial species.

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

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