Antimicrobial activity of different silver-containing nanostructured materials

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Silver compounds have been used as antibacterial agents for centuries, from the use of coins to preserve water in ancient Greece and Rome, to the XIX century use of silver nitrate to treat a variety of ailments, from typhoid to post-partum infections. [1] Today a variety of applications are contemplated, including Ag-coated medical devices, dressings for chronic wounds and burns, cosmetics, food preservation and water treatment. [1], [2]. The silver mechanism of action is far from understood: the biochemical explanations proposed include (i) inactivation of proteins, enzymes and DNA by Ag attachment to groups containing sulfur or phosphorus, (ii) cell membrane and mitochondria damage, (iii) inhibition of respiratory processes and (iv) generation of reactive oxygen molecules and free radicals.

In this work, we describe the bactericidal ability of different forms of silver on highly adherent slime producing *Staphylococcus aureus* strain 9213 [3]. We show that silver-exchanged zeolites (ZSM5) at low Ag loadings are more effective against *S.aureus* than other materials with higher amount of silver such as silver (I) oxide and nanoparticulated (< 100 nm) silver.

NH_4-ZSM-5 microparticles with a Si/Al ratio of 20 were purchased from Zeolyst Int.; other commercial Ag-exchanged zeolites with different silver loadings (i.e.,15-20 wt. % and 35 wt. %) and geometries (pellets and granular, respectively) were purchased from Sigma-Aldrich as well as silver (I) oxide and nanoparticulated (< 100 nm) silver. The ammonium form of the zeolite was calcined to obtain its protonic form and ion-exchanged by using 1 wt. % $AgNO_3$ solution (atomic absorption standard solution 1 wt. % (HNO_3), Sigma Aldrich) under stirring, for up to 24 hours. In this way we obtained Ag loadings around 0.2 wt. %. [4].

For the antibacterial assays, 2 mL of an overnight stationary growth phase of TSB bacterial culture were added under sterile conditions to glass tubes, each of which contained the 60 mg of Ag-ZSM5, H-ZSM5 (as control), silver (I) oxide, commercial pellets, commercial granular particles and nanoparticulated silver. Samples were subsequently incubated at 37 ºC for 4, 6 or 24 hours in the dark. After incubation tubes were placed in a 50 Hz ultrasonic bath for 15 minutes (tubes at 4 and 6 hours) or 30 minutes (tubes at 24 hours). Seven 1:10 dilutions of the contents of each ultrasonicated tube were made and then three 25 μL drops of each suspension were spread on Triptone Soy Agar (TSA) plates. Bacterial colonies were counted after incubation overnight at 37º C. Silver content on the released media was determined using atomic absorption spectrophotometry (AAS) in a Varian Spectra A110. Media were centrifuged using exclusion centrifugal filters (5 nm cut–off) to obtain supernatants free of any lose nanoparticles. [4].

The bactericidal action when using the same amount of material added to the stationary grown bacteria (10^9 CFU/mL) was found maxima for the Ag-exchanged zeolite compared to the other commercial silver-exchanged zeolites (pellets and granular) (Figure 1).

Silver exchanged zeolites showed nanosized clusters on their surfaces indicating that in those materials silver is present as cation in the zeolite network as well as metallic silver as clusters or nanoparticles on their surfaces. The higher the bioavailability of cationic silver, the higher the bactericidal action of the silver-carrier material. Silver (I) oxide showed the highest bactericidal action of all the materials tested. Ionic silver content, particle geometry, silver solubility, particle size and crystallinity are some of the variables controlling the bactericidal effect of silver.

References

[1] D. K. W. Brett, ***Ostomy Wound Manage***., (2006), 52, 3.
[2] X. Chen and H. J. Schluesener, ***Toxicol. Lett***., (2008), 176, 1.
[3] M. Monzón et al. ***J. Orthopaedic Res.*** (2001), 19, 280.
[4] P.Lalueza, M.Monzón, M.Arruebo, J.Santamaria. ***Chem. Comm.*** (2011), 47, 680.

Figures

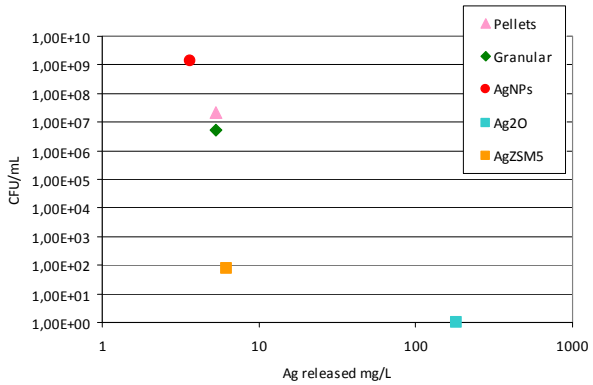

Figure 1. *Bactericidal action of the different silver-containing materials.*

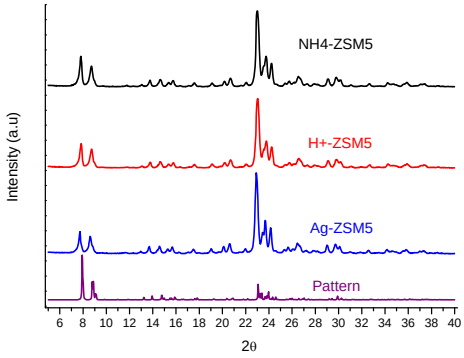

Figure 2. *XRD ZSM5 before and after calcination and ion-exchange.*

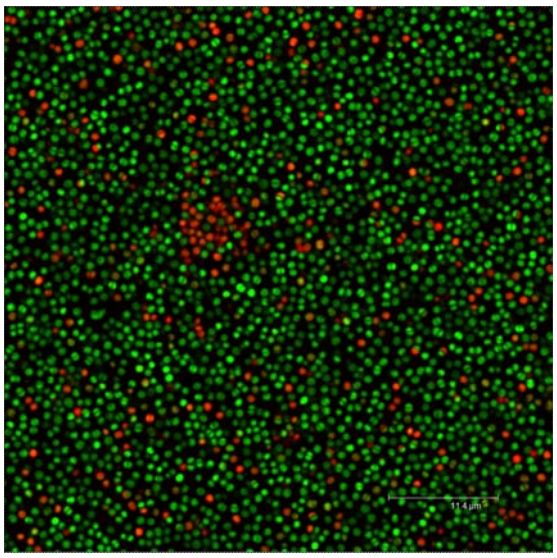

Figure 3. *S.aureus by CLMS.*

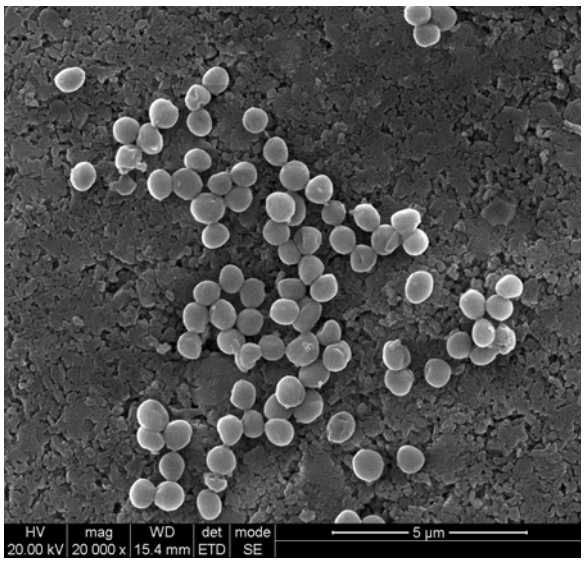

Figure 4. *S.aures by SEM.*