Effect of silver ion incorporation into a bioactive glass surface on the adsorption of albumin

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Effect of silver ion incorporation into a bioactive glass surface on the adsorption of albumin

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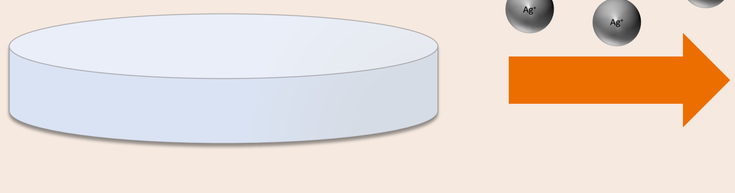
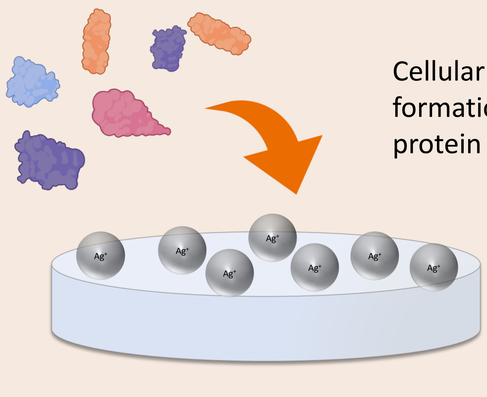
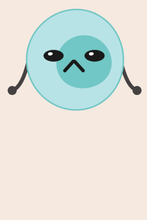
Bioactive glasses are able to promote tissue integration, such as osteointegration, and hydroxyapatite precipitation. Unfortunately, also biofiml can form on their surfaces.
Thus, silver ions are incorporated in bioactive glasses surfaces for achieving antimicrofouling properties.



How these surface treatments change protein adsorption?

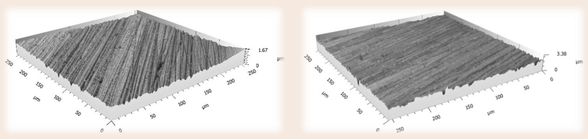


Cellular response and biofilm formation are driven by the protein layer on biomaterials



The topography is unchanged by the surface treatments

SBA2: S_a= 81 nm Ag-SBA2: S_a = 120 nm

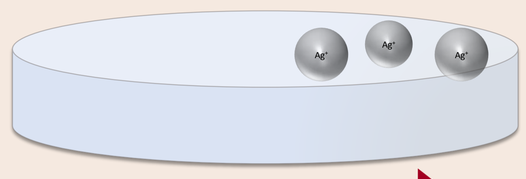


Surface 3D images by confocal microscopy and surface roughness

Silver is succesfully incorporated into the glass

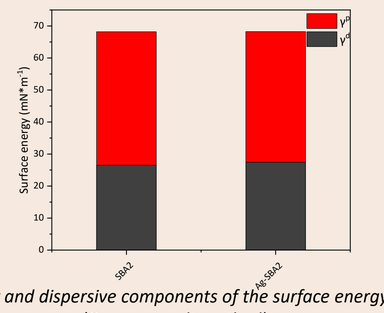
Atomic %	Si	O	Ca	Na	Ag	C&Others
SBA2	5.08	37.61	7.67	2.88	n.d.	46.76
AgSBA2	11.96	41.8	2.91	2.95	7.03	33.35

Surface chemical composition (X-ray photoelevtron spectroscopy)



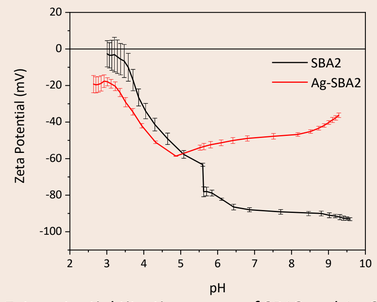
Both surface energy, regarding dispersive and polar components, and the water contact angle are unaffected

	Contact angle
SBA2	22°
Ag-SBA2	22.33°

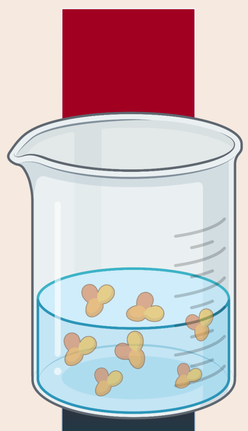


Polar and dispersive components of the surface energy (Owens-wendt method)

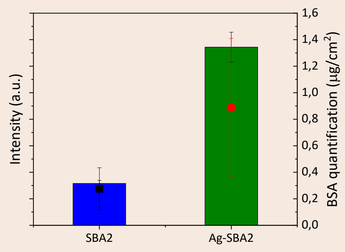
Ag-SBA2 has a more positive surface at basic pH thanks to Ag^+ ions



Zeta potential titration curves of SBA2 and Ag-SBA2

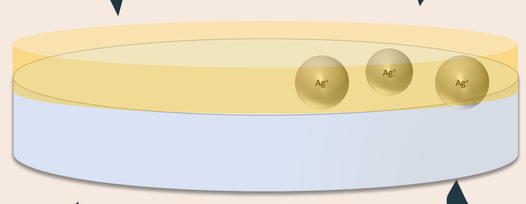


Quantification of the adsorbed albumin by different methods (chemical assay, fuorescence and XPS) shows that Ag-SBA2 can adsorb a slightly higher amount of protein

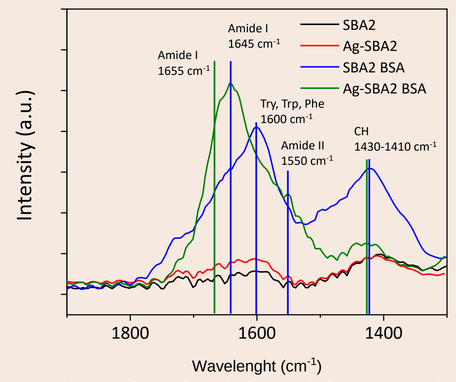


	N%
SBA2	2.01
Ag-SBA2	9.01

Protein quantification by fluorescent intensity (rodhamine-conjugated BSA, bars) or bichinconininc acid assay (BCA, dots)



The redshift of the Amide I peak on Ag-SBA2 indicates a partial unordering of the α-helical structure with respect to the protein adsorbed on SBA2



FTIR spectra in the Amide I region for surfaces before and after protein adsorption. Relevant deconvoluted peak positions are highlighted

After BSA adsorption, the shift of the IEP towards the one of albumin confirms the presence of the protein on both surfaces

Zeta potential titration curves of SBA2 and Ag-SBA2

Both surfaces showed similar coverage of the protein layer, which is homogeneous

Fluorescent images (200x) of SBA2 (left) and Ag-SBA2 (right) after BSA adsorption

Surface potential immagine (Kelvin Probe – AFM) can highlight both the formation of the protein layer (lighter areas) and the one of the silica gel (darker areas)

Surface potential images of SBA2 (left) and Ag-SBA2 (right) partially covered by BSA

Materials

Glass substrates (disks, ϕ = 1cm; grit with SiC paper up to 1000) :

- SBA2 (mol %: 48% SiO2, 18% Na2O, 30% CaO, 3% P2O5, 0.43% B2O3, 0.57% Al2O3)
- Ag-SBA2: SBA2 soaked in 0.03M $AgNO_3$ for 1h at 37°C

Protein solution: bovine serum albumin (BSA) 7in PBS 20 mg/ml, pH 7.4.

Bibliography:

- K. Zheng et al. Applied Materials Today 15 (2019) 350–371
- M. Miola et al. Biomedical Materials 10 (2015) 055014

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Conclusion

Silver doping of a bioactive glass does not affect much some surface properties which are pivotal for protein adsorption, such as topography, wettability and surface energy. Still, at physiological pH, the surface is less negatively charged. As consequence, the Ag-SBA2 can adsorb more albumin thanks to interactions between the negatively charged protein surface (-COO^- groups) and the positive metal ions, Ag^+. The stronger interaction results also in a partial denaturation of the protein on the surface.
Knowing how antibacterial surface modifications alter the formation of the protein layer can improve the optimization of such treatments in order to elicit a proper biological response.