Binding of BSA-conjugated gold nanoparticles to APTES self-assembled monolayer : an imaging ellipsometry study

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Introduction and motivation

- **Serum albumin in the most abundant protein in the blood plasma**
- **Human (HSA) or bovine (BSA) serum albumin acts as reservoir for small molecules : competition in adsorption processes and time-controlled release of drugs**
- **Immobilization of nanoparticles (NPs) on solid surfaces is influenced by the environmental conditions of the surrounding biological fluids and by the soft core-shell structure of the NPs when organic molecules (and competing proteins) are absorbed at the NP surface.**
- **Bound NPs exhibit optical resonances in the absorption/transmission spectra due the surface plasmon excitation**

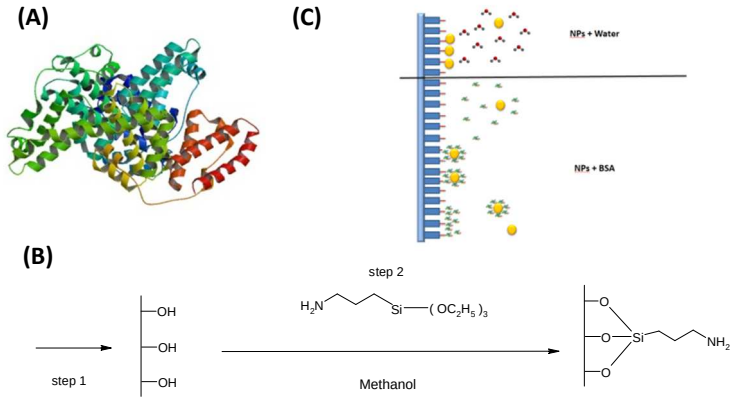

Figure 1 : (A) BSA molecule (B) APTES molecule grafting scheme (C) Binding scheme of the (conjugated) NPs to the functionalized surface

Results and discussion

Imaging ellipsometry : Non-destructive **LOCAL** optical analysis technique based on the relative change of polarization of the *p*- and *s*- components of the light at the interface between two media characterized by different optical properties (**Resolution : 1 µm / pixel** – Magn. 10x)

$$\rho = \frac{R_p}{R_s} = \tan\Psi\, e^{i\Delta}$$

with $\tan\Psi = \frac{|R_p|}{|R_s|}$ ***and*** $\Delta = \delta_p - \delta_s$

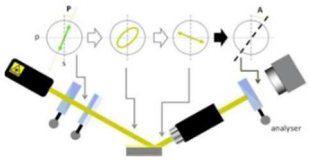

Figure 2 : Accurion EP3-sw imaging ellipsometer -- Polarized light reflected on the substrate gives light polarized in another direction in the *s* and *p* space. The analyzer and the polarizer rotate to extinguish the beam.

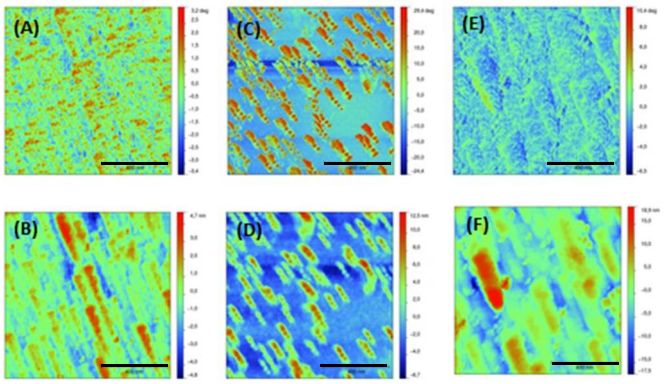

Figure 3 : AFM topography and phase images . (A,B) APTES-functionalized glass surface (C,D) Same with gold BSA-cNPs from PBS buffer bound to the APTES molecules (24 hrs) (E,F) Same with gold NPs electrostatically bound from water (24hrs) [scale bar : 400 nm]

Advantage of IE :
- Local information on optical properties and thickness
- Allows a fast statistical analysis with a large number of significant data
- Metal on Insulator (MOI) : relevant ellipsometric angle is Ψ

	APTES	cNPs / PBS	NPs / H2O
Ra (nm)	0.84	2.21	3.32
Rq (nm)	1.11	2.82	4.37
Kurtosis	0.91	0.80	148

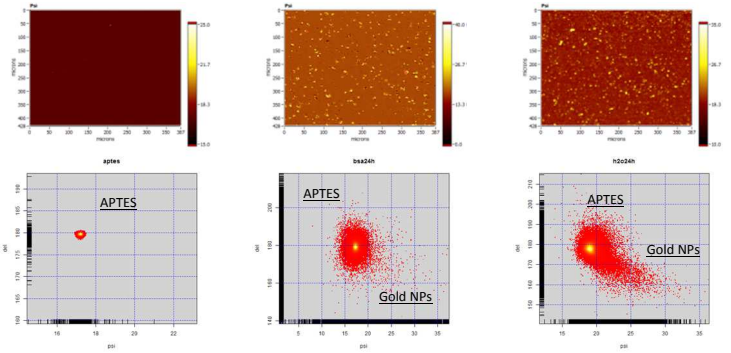

Figure 4 : (top panel) Ψ maps – (bottom panel) 2D Ψ–Δ histograms showing the (lack of) adsorption of the NPs. From left to right : APTES surface, BSA-cNPs from PBS solution, NPs from water (citrate stabilized).

Strong differences in adsorption for citrate stabilized (water) and BSA conjugated NPs. Obvious from the number of outliers in the statistical distributions. To be analyzed by cluster analysis methods and Gaussian mixture models (GMM).

Conclusion and acknowledgements

- **IE is an appropriate technique to monitor the (conjugated) NPs binding process to surfaces**
- **In solution : conjugation of the BSA to the NPs shifts the resonance from 519 nm to 524 nm**
- **Systematic study between AFM images and IE images to be continued, including kinematics effects**
- **Cluster analysis : a powerful tool to detect early stages of adsorption processes**

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