Effectiveness of different sampling techniques for the collection of FT-IR spectra of proteins adsorbed on nano-hydroxyapatite

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The importance of understanding interaction of nanomaterials with biomolecules is shared by medicine, microbiology, and toxicology because nanoparticles introduced into biological media initially bind proteins. [1]

FT-IR spectroscopy is a powerful technique to obtain a relevant information for understanding protein adsorption on surfaces. [2]

When adsorbed or in proximity of metal particles or films, molecules can exhibit more intense IR absorptions . This effect is called Surface Enhancement of Infrared Adsorption (SEIRA), which results in a more sensitive technique to detect surface species in vicinity of metal surface promoting plasmon resonance when interacting with IR radiation [3].

Issues picked up for the current study include quantitative features of proteins adsorbed on the surface of nanohydroxyapatite (nano-HA) and the assessment of the possible occurrence of a SEIRA effect (by external reflection on gold substrates), evaluated with respect to different spectroscopic operational modes (transmission, internal reflection).

To this aim, infrared spectra of bovine serum albumin (BSA) adsorbed in known amount on Hydroxyapatite nanoparticles were collected at different conditions: on SEIRA-active rough gold surface and on flat gold support in external reflection mode, on inert surface in transmittance mode (CaF_2 support), and in single-reflection ATR mode (diamond crystal).

Quantification of adsorbed protein was carried out by UV-spectroscopy and evidence for the formation of protein multilayers were obtained by CD-UV spectroscopy.

It was concluded that some SEIRA effect occurred for samples in contact with nanostructured gold substrates, but likely limited to the part of protein multilayers adsorbed on nano-HA within the effective field for enhancement from the gold surface (ca. 10 nm [3]).

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References

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