

## Raman mapping investigation of functionalized hydroxyapatite ceramics surface for bone engineering

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### ABSTRACT

The development of osteoinductive bone substitutes for large bone defects under mechanical stress requires the functionalization of their surface by biomolecules involved in cell adhesion, angiogenesis or osteogenesis in order to improve their biointegration. Among the proteins of interest, vascular endothelial growth factor (VEGF) is involved in vascularization and bone formation.<sup>[1]</sup> Protein functionalization of bioceramics can be achieved in a multi-step protocol using cross-linker agents such as organosilanes. In this approach, each step is strongly influenced by the previous one, *i.e.* the distribution of the protein is related to that of the intermediate agents.

Two calcium phosphate ceramics were chosen as substrates because their chemical composition and structure are close to the mineral phase of bone: stoichiometric hydroxyapatite (HA) which is widely used as a bone substitute and carbonate hydroxyapatite (CHA) which is very similar to natural biological apatite.<sup>[2-4]</sup> VEGF was grafted onto the substrates surface using intermediate molecules: silane (APTES) and NHS-PEG6-maleimide (PEG6) used for silanization and pegylation steps respectively. A chemical analysis of the same area was recorded by Raman spectroscopy after each step of the functionalization to assess the spatial distribution of intermediate molecules and that of VEGF. Raman mappings were reconstructed on the specific bands of grafting intermediates and proteins (Fig. 1) which highlighted the impact of the functionalization steps on the final distribution of the VEGF.

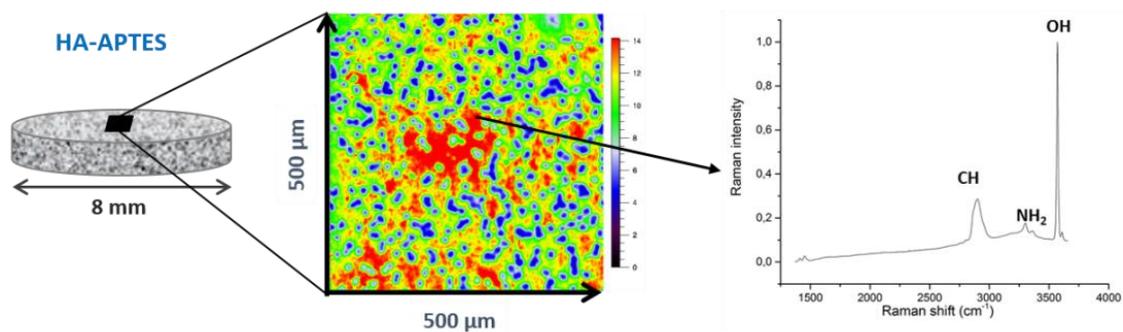


Fig. 1: 2D Raman maps were performed using an InViaReflex (Renishaw) spectrometer at a wavelength of 532 nm over wide areas ( $500 \times 500 \mu\text{m}^2$ ) and reconstructed on the vibrational mode of aliphatic C-H bonds between  $2814$  and  $3006 \text{ cm}^{-1}$ .

### References:

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