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Functionalized coatings by Cold Spray: an *In Vitro* Study of micro- and nanocrystalline Hydroxyapatite compared to Porous Titanium

A.M. Vilardell*¹, N. Cinca¹, S.Dosta¹, I.G.Cano¹, J. M. Guilemany¹

X.Nogués², N.Garcia-Giralt²

¹Centre de Projecció Tèrmica (CPT). Dpt. Ciència dels Materials i Enginyeria Metal.lúrgica. Universitat de Barcelona Martí i Franquès 1, 08028 Barcelona, Spain.

²IMIM (Institut Hospital del Mar d'Investigacions Mèdiques), RETICEF, CIBERFES, ISCIII. Doctor Aiguader 80, 08003 Barcelona, Spain

*Corresponding author: amvilardell.research@gmail.com

ABSTRACT

Three different surface treatments on a Ti6Al4V alloy have been *in vitro* tested for possible application in cementless joint prosthesis; all of them involve the novelty of using the Cold Spray technology for their deposition: (i) an as-sprayed highly rough titanium and, followed by the deposition of a thin hydroxyapatite layer with (ii) microcrystalline or (iii) nanocrystalline structure. Primary human osteoblasts extracted from knee were seeded onto surfaces and cell viability using MTS and LIFE/DEAD assays, osteoblasts differentiation by alkaline phosphatase (ALP) quantification as well as cell morphology were tested at 1, 7 and 14 days of cell culture.

Different cell morphologies between titanium and hydroxyapatite surfaces were exhibited; at 1 day of cell culture, cells on the titanium coating were spread and flattened, expanding the filopodia actin filaments in all directions, while cells on the hydroxyapatite coatings showed round like-shape morphology due to a slower attachment. Higher cell viability was detected at all times of cell culture on titanium coating due to a better attachment at 1 day. However, from 7 days of cell culture, cells on hydroxyapatite showed good attachment onto surfaces and highly increased their proliferation, mostly on nanocrystalline, achieving similar cell viability levels than titanium coatings. ALP levels were significantly higher in titanium, in part, because of greatest cell number. Overall, the best cell functional results were obtained on titanium coatings whereas microcrystalline hydroxyapatite presented the worst cellular parameters. However, results indicate that nanocrystalline hydroxyapatite coatings may achieve promising results for the faster cell proliferation once cells are attached on the surface.

Keywords: Titanium; Hydroxyapatite; Cold Spray; in-vitro testing;

1. INTRODUCTION

The surface treatments for cementless joint prosthesis have been the subject of exhaustive study during the last decades. These have been performed with the intention to improve the osseointegration of titanium aiming at modify the microstructure of the surface by texturing or changing the chemical composition [1]; textures have played an important role leading to an increase of free surface and cell differentiation [2]. On the micron scale, roughness is clearly important and it has been shown to be important for bone bonding. However, those textures should be in a range between 100-500 microns to enable the bone ingrowth for good mechanical fixation [3]. Moreover, textures are also important at

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