Laboratory tests

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Expression of SP7, RUNX1, DLX5, and CTNNB1 in human mesenchymal stem cells cultured on xenogeneic bone substitute as compared with machined titanium

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ABSTRACT

After tooth extraction, a negative bone remodelling occurs, with important dimensional changes. Consequently, one of the major clinical objectives is to prevent or limit the alveolar bone loss, maybe through an implant inserted immediately after one of the available augmentation techniques, such as the use of growth and differentiation factors, particulate and block grafting materials, distraction osteogenesis, and guided bone regeneration. Alveolar bone resorption and remodelling involve genes and a greater knowledge about factors expressed during bone repair could be useful in order to individuate novel therapeutic alternatives in this field. The aim of this research was to investigate the gene expression profile of 4 transcription factors in human mesenchymal stem cells (hMSC) cultured with an organic bone substitute consisted of cortical porcine bone (CPB) (OsteoBiol® *Lamina*, Tecnoss®, Giaveno, Italy) and a titanium surface consisted of Machined Titanium in the form of Disks (MTD) (Biotec, Vicenza, Italy). In vitro studies were performed on hMSC cells, which grew in contact with CPB and MTD for 10 days. RNA quantification for genes DLX5, CTNNB1, RUNX1, and SP7 was assessed by quantitative real-time polymerase chain reaction. For cells supported by titanium, immunocytochemistry of osteocalcin (OC) was also performed. In the osteoblast-induced cells (OIC), DLX5, CTNNB1, and RUNX1 were significantly upregulated (+2.38-, +3.51-, and +7.08- fold, respectively), whereas SP7 was downregulated (-26.32-fold). None of the genes seemed to be upregulated or downregulated by the cortico-cancellous porcine bone. In cells grown on titanium support, DLX5 and RUNX1 were respectively upregulated (+3.12-fold) and downregulated (-2.14-fold).

CONCLUSIONS

The 2 genes RUNX1 and SP7 resulted differently expressed in cells cultured on metallic supports if compared with their expression recorded for induced osteoblasts. An induction of the osteogenic phenotype was observed when cells were cultured on machined titanium, but not on xenogeneic material. Cortical porcine bone seemed to have minimal impact on gene expression. An induction of the osteogenic phenotype was observed when cells were cultured on machined titanium.

ORIGINAL ARTICLE
Implant Dentistry
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LABORATORY TESTS

Plasma of argon enhances the adhesion of murine osteoblasts on different graft materials

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ABSTRACT

It has been demonstrated that the amount of bone regeneration is strictly correlated to the interaction between the graft material and the osteoregenerative cells and that the process of new bone formation and graft material resorption depends on the physico-chemical properties of the material itself. As acceleration of osseointegration of graft particles may depend on the optimization of the biomaterial rather than on an actual increase in the rate of bone response, the alteration of the physical surface characteristics might positively affect early bone response. Plasma treatment demonstrated to activate surfaces at the atomic and molecular level, producing hydrophilic surfaces and enhancing their wettability. Thus, plasma application can lead to an improved adhesion of cells. Following these considerations, the aim of the present study was to test the effect of plasma treatment on different graft materials regarding a change in surface characteristics and assessing protein adsorption and osteoblast growth. Four different classes of graft materials, representing commonly used classes of bone substitute material, were used: synthetic pure hydroxyapatite discs (Sintlife, Finceramica, Faenza Italy) (Mg-HA); biphasic calcium phosphate (60% HA, 40% ß-TCP) discs (SUN- STAR Degradable Solutions AG, Schlieren, Switzerland) (BCP); cancellous xenogeneic (porcine) bone matrix discs (OsteoBiol® *Sp-Block,* Tecnoss®, Giaveno, Italy) (CaBM) and cortical xenogeneic (porcine) bone matrix discs (OsteoBiol® *Cortical Lamina*) (CoBM). All specimens were manufactured for the purpose of the study and were non-commercial products. Fifty serially numbered disks with a 10 mm-diameter from each graft material were randomly divided into two groups: test group (argon plasma treatment) and control group (absence of treatment). Cell morphology (using pre-osteoblastic murine cells) and protein adsorption were analyzed at all samples from both the test and control group. Differences between groups were analyzed using the Mann–Whitney test setting the level of significance at $p < 0.05$. Plasma treatment significantly increased the protein adsorption and cell adhesion in all groups. Cancellous and cortical BM grafts showed higher values in total protein adsorption compared to BCP and Mg-HA samples, both in the control and the test group. In group BM, a higher number of cells were embedded in the pores of the rough surface.

CONCLUSIONS

Outcomes of the present study confirm that bio-activation of graft material (independent of their physical characteristics) can positively influence cell and protein behaviour at substitute surfaces. Actually, bio-functionalization increased protein adsorption by up to 60% compared to untreated graft samples and a similar behaviour was observed analyzing cell adhesion (at least 30% higher cell adhesion for all graft materials). In their conclusions, the Authors affirmed that *"Within the limitations of the present study, the data obtained confirm that non-atmospheric plasma-of-argon treatment is capable of increasing protein adsorption and cell adhesion to different classes of graft materials. Further research is required to assess whether these results might be beneficial in clinical settings"*.