A novel nanoporous silica calcium phosphate material (nSCP) as a promising osteogenesis-inducing biomaterial for bone regeneration

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Keywords— Bone regeneration, MSC, bone, osteogenesis

INTRODUCTION

In the fields of orthopaedics and odontology, autologous bone transplantation is considered the gold standard for the reconstruction of large bone defects that exceed the reparative capabilities of bone tissue. However, alternatives to autologous transplants are needed due to the limited availability of host material and donor site morbidity [1]. This has prompted the field to search for osteoinductive materials that possess similar structure, composition, and properties to the main inorganic component of bone tissue [2]. In this study, we utilized a 3D bone marrow-derived mesenchymal stem cell (BMSC) and type I collagen model to evaluate the osteogenic potential of a novel nanoporous silica calcium phosphate material.

MATERIALS AND METHODS

Cytotoxic effects of nSCP material were assessed by Live/Dead cell viability assay at various concentrations.
BMSCs from four healthy donors were embedded in type I collagen hydrogels, with an approximately $3x10^5$ cells contained in 200 µl of type I collagen (conc. 3,9-4,4 mg/ml). The gels were manually cast. Experimental groups consisted on undifferentiated and chemically differentiated groups, with or without the addition of nSCP or HA/βTCP (hydroxyapatite/β-tricalcium phosphate, a commonly used biomaterial serving as a positive control [3]). The hydrogels were cultured for 3-5 week. The adipogenic potential of nSCP was also measured by inducing adipogenic differentiation. Osteogenesis-reporting stainings (Alizarin red, Von Kossa, Osteoimage®) were used to detect mineralization levels. Osteogenic-specific gene expression was analysed by qRT-PCR. Furthermore, a single molecule detection system (nCounter®, NanoString) was used to quantify the expression of 748 metabolic genes in response to osteogenic differentiation.

RESULTS AND DISCUSSION

In this study, we introduced a nanoporous silica calcium phosphate with the composition: $CaO\text{-}SiO_2\text{-}P_2O_5$. This nanostructured and mesoporous material displayed no cytotoxic effects at concentrations below 10 mg/ml. Histological analysis showed clear ossification of the hydrogels even in absence of chemical differentiation factors, thus indicating positive osteoinductive properties. Due to its smaller particle and aggregate size, nSCP could be homogeneously distributed in the scaffold compared to the positive control HA/βTCP. qRT-PCR data suggested higher levels of some osteogenesis-related genes in the HA/βTCP compared to the nSCP group. Adipogenesis was inhibited by nSCP as expected. However, the multivariate analysis of the 748 metabolic-related genes showed different genes being expressed between nSCP and HA/βTCP, suggesting alternative differentiation pathways for both materials. The effect of nSCP were not enhanced by the presence of differentiation medium. The most significantly upregulated and downregulated genes by the presence nSCP were: *SLC16A6*, *GMPR*, *GOT1*, *IDH2* and *LEPR*. Taken together, our results indicate that nSCP alone can induce osteogenesis in BMSCs.

CONCLUSIONS

This study shows evidence of the osteogenic potential of the novel nanoporous calcium silica phosphate material (nSCP).

ETHICAL STATEMENTS

The local ethics committee for research at Umeå University (Dnr 2013-276-31M and 03-425) approved collection, processing, culture, storage, and usage of all clinical isolates in this study. All methods were performed by following the relevant guidelines and regulations. Informed consent was obtained from all donors.

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ACKNOWLEDGEMENTS

Special thanks to Anna-Karin Olofsson and other members of the NRCT group.