



The impact of heat treatment on interactions of contact-poled biphasic calcium phosphates with proteins and cells

C. Wolf-Brandstetter^{a,*}, U. Hempel^b, S. Clyens^c, A.A. Gandhi^d, O. Korostynska^d, S. Oswald^e, S.A.M. Tofail^d, N. Theilgaard^c, H.-P. Wiesmann^a, D. Scharnweber^a

^a Max Bergmann Center of Biomaterials, TU Dresden, Dresden 01069, Germany

^b Institute of Physiological Chemistry, Medical Department, TU Dresden, Dresden 01309, Germany

^c Danish Technological Institute, Gregersensvej, Taastrup 2630, Denmark

^d Materials and Surface Science Institute, University of Limerick, Limerick, Ireland

^e Leibniz Institute for Solid State and Materials Research Dresden, Dresden 01171, Germany

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ABSTRACT

A number of studies have reported improved bone integration for calcium phosphate based materials electrically “poled” by an external electric field prior to implantation. In our study we investigated the effects of electrical polarization of a biphasic ceramic composed of 80% hydroxyapatite and 20% β -tricalcium phosphate. As contact poling involves elevated temperatures as a prerequisite for inducing charge, we used two reference types: samples without any heat treatment and poling, and samples with no poling but heat treatment identical to that of the poled samples. All heat-treated samples (poled or unpoled) showed an improved wettability, which was attributed to a reduced hydrocarbon contamination. Heat treatment alone provoked an accelerated spreading of osteoblast-like cells, whereas on poled samples a retarded cell spreading was observed. While proliferation and several differentiation markers were not influenced by either heat treatment or poling, the release of proinflammatory cytokines interleukin-6 and -8 was significantly reduced for all heat-treated samples, irrespective of additional electrical poling. The study demonstrated that the behaviour of cells in contact with poled biphasic ceramics was influenced by two parameters: heating and charge. Our data revealed that heating of the calcium phosphate ceramics had a much more pronounced effect on cell behaviour than charge.

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1. Introduction

Calcium phosphate-based biomaterials are widely used bio-ceramics in bone substitute applications due to their bioactivity, osteoconductivity and compositional similarities to bone [1–3]. The most prominent calcium phosphates used in this field are hydroxyapatite (HAp, $\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$) and β -tricalcium phosphate (β -TCP, $\text{Ca}_3(\text{PO}_4)_2$). Mixtures of these two phases, also known as biphasic calcium phosphates (BCPs), are usually favoured for clinical applications because their resorption rate can be “tuned” to match the bone-healing rate, resulting in a suitable balance between implant degradation and bone regeneration [4].

A number of studies have reported a further putative improvement of bone integration for calcium phosphate-based materials that have been electrically “poled” by a DC electric field prior to implantation. Several reports, mainly from the group of Yamashita

[5–9] but also from others [10,11], have established that HAp can store electrical charge originating from this electrical “poling”. The poling of mixtures of β -TCP and HAp [12–14] and pure β -TCP [15] have also been reported. The improvement in bioactivity has been attributed to a “vectorial” effect due to induced polarization in calcium phosphates obtained by the application of the DC field [16].

The reported biological effects of electric poling are diverse: poling has been reported to result in increased deposition of bone-like apatite on negatively poled (N-poled) surfaces [7] as well as improved osteoblast adhesion, spreading, proliferation or extracellular matrix deposition on N-poled surfaces [9,14,17]. In contrast, Kizuki et al. found an increased number of MC3T3 cells on positively poled (P-poled) surfaces [18], while other authors observed no differences between N- or P-poled surfaces but an overall improvement in adhesion, attachment and proliferation of osteoblast-like cells for poled samples [5,10]. One of the fundamental aspects of the above reports is that they all have found biological effects on poled calcium phosphates samples compared to unpoled and not heat-treated samples. However, because the poling of calcium phosphates in the reported investigations involved elevated temperatures (typically above 250 °C), a potential contribution

* Corresponding author. Address: Max Bergmann Center of Biomaterials, TU Dresden, Budapester Strasse 27, Dresden 01069, Germany. Fax: +49 351 46339401.

E-mail address: Cornelia.Wolf-Brandstetter@tu-dresden.de (C. Wolf-Brandstetter).