The Attachment of Human Primary Osteoblast cells to Oxygen Plasma Modified PEEK

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Introduction: Polyetheretherketone (PEEK) has come into the spotlight as a replacement for metals in devices such as spine cages and craniomaxillofacial (CMF) implants, due to its high strength, good wear and radiolucent properties[1]. Evaluation of soft and hard tissue integration to implants by X-ray or MRI can be obscured by the presence of the metal devices. Several implants have therefore been redesigned into this radiolucent material. However, cellular attachment to polymers such as PEEK is restricted due to their intrinsic low surface energy, which can lead to implant loosening, as a result of fibrous encapsulation. Higher energy surfaces have been shown to promote rapid cellular adhesion and spreading, in contrast to surfaces with lower energy[2,3]. To improve cell attachment the surface energy can be increased by plasma surface treatment. The present study aims to investigate the effect of oxygen plasma treatment of PEEK on the attachment and functionality of primary human osteoblast-like cells (HOB).

Materials and Methods: Injection moulded PEEK Optima™ discs (Invibio) with a 13mm diameter were modified by radio frequency (RF) plasma treatment, Thermanox (THX) (Nunc) and Ti ISO 5832/2 (Synthes) were used as the control surfaces. Using an EMITECH RF plasma treater, the samples were exposed to varying treatment times. Surface chemical compositions of treated and untreated surfaces were characterised by XPS, wettability by contact angle; topographic changes by AFM and SEM. HOB cells isolated from femoral heads removed during total joint replacement operations were grown to 70-80% confluence in DMEM (10% FCS in 5% CO_2 at 37°C), and plated at 10000 cells/cm^2. Alpha-MEM (0.1µM dexamethasone and 10mM beta-glycerophosphate) was used as mineralisation media over 21 days. Cell functionality was assessed by alkaline phosphatase expression (ALP), mineralisation by Alizarin red S (ARS) staining of calcium, cell attachment by SEM and cell density through the alamarBlue™ assay.

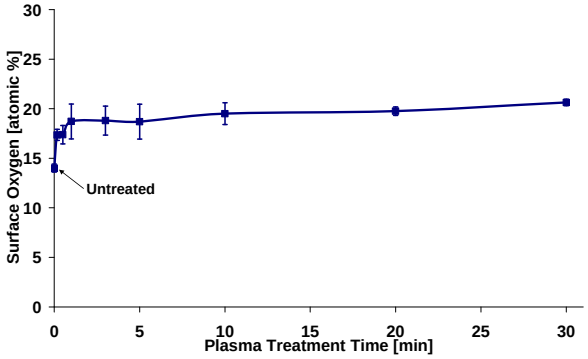

Figure 1: *Surface oxygen concentration of PEEK surfaces with increasing plasma treatment time.*

Results: XPS analysis of the untreated PEEK discs showed 14 atomic% surface oxygen, as expected, indicating that these surfaces are relatively hydrophobic in character[4]. Analysis of the plasma treated PEEK surfaces showed that the surface oxygen concentration increased with increasing treatment time up to ~20 atomic% (Fig 1). High resolution C1s spectra showed a greater increase in C-OR type functional groups than C=O and O-C=O with increasing treatment time. To study the effects of the surface treatment on cell attachment and functionality, the cells were observed after plating on the treated and untreated PEEK, THX and cpTi surfaces. Within 72hrs, the treated surfaces were shown to have higher cell densities than the untreated surfaces (Fig 2). By day 21 the treated surfaces were shown to have similar cell densities to cpTi. Oxygen plasma treatment has been found to etch polymer surfaces, by AFM, this was found to occur after more than 20min plasma treatment.

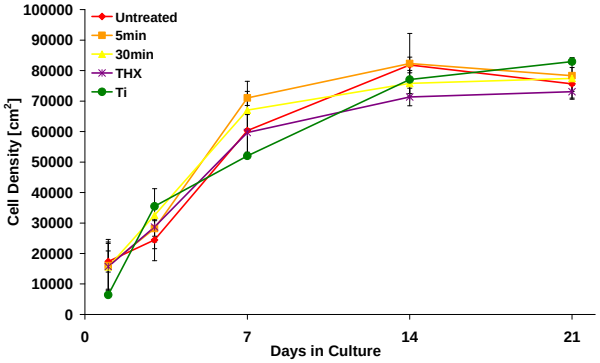

Figure 2: *HOB cell densities over the 21 days of culture, showing the optimised levels of surface treatment in comparison to THX and cpTi.*

Cells were also observed by SEM to attach more readily to the treated surfaces with higher concentrations of C-OR functional than the untreated surfaces. ALP expression was observed to be more characteristic on the PEEK surfaces with higher concentrations of C-OR functional groups. Nodule formation quantified by dissolving the ARS stain was found to be greater on the PEEK surfaces than on the THX surfaces, and similar to the levels on the cpTi surfaces.

Discussion/Conclusions: The incorporation of oxygen through plasma treatment can be used to increase the surface energy and thereby aid the adhesion of HOB cells. Surface treatment of PEEK has led to higher levels of nodule formation than on THX and similar levels to cpTi, indicating that these treated surfaces are likely to improve bony integration to implants.

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