

Nucleation and growth of apatite on NaOH-treated PEEK, HDPE and UHMWPE for artificial cornea materials

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Abstract

The skirt of an artificial cornea must integrate the implant to the host sclera, a major failure of present devices. Thus, it is highly desirable to encourage the metabolic activity of the cornea by using more bioactive, flexible skirt materials. Here we describe attempts to increase the bioactivity of polyether ether ketone (PEEK), high-density polyethylene (HDPE) and ultra-high molecular weight polyethylene (UHMWPE) films. The effectiveness of different strength NaOH pre-treatments to initiate apatite deposition on PEEK, HDPE and UHMWPE is investigated. We find that exposure of PEEK, HDPE and UHMWPE films to NaOH solutions induces the formation of potential nuclei for apatite (calcium phosphate), from which the growth of an apatite coating is stimulated when subsequently immersing the polymer films in 1.5 strength Simulated Body Fluid (SBF). As immersion time in SBF increases, further nucleation and growth produces a thicker and more compact apatite coating that can be expected to be highly bioactive. Interestingly, the apatite growth is found to also be dependent on both the concentration of NaOH solution and the structure of the polymer surface.

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1. Introduction

Corneal disease is the second most common cause of blindness in the world [1]. It is estimated that 45 million people worldwide are bilaterally blind and 10 million are affected by corneal blindness [1]. Corneal blindness mostly affects the population in the equatorial zone, due to the high exposure to UV light. Corneal grafting presents complications such as rejection and post-astigmatism plus the lack of donor material and resources [2]. In allograft material, collagen-based implants may fail to sustain a normal corneal epithelial phenotype or stromal tissue clarity in the hostile ocular surface environment [3]. Additionally

with allografts there are problems with the supply of corneas and disease transmission. Conventional corneal grafting (keratoplasty) is not advised for patients with bilateral corneal blindness or who suffer from a range of clinical problems including tear deficiency, chemical burns and uncontrollable intraocular pressure [4–7]. At present, an artificial cornea, i.e. a keratoprosthesis (KPro), is the only alternative to keratoplasty (cornea donor transplantation).

KPros consist of a clear central optic and a sewing skirt used to anchor the implant [4]. KPros have shown improvement of the vision in some cases [3,7–15], but failure rates of up to 40% at one year have been reported, mainly due to poor integration between the artificial implant and the host cornea [4,5]. Failure may be associated with extrusion, inflammation or retroprosthetic membrane formation. The use of biocompatible materials that allow colonization of the optics by host keratocytes and epithelial cells has shown that it is possible to integrate

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alloplastic materials into the recipient cornea [5,16–18]. However, tissue breakdown and aqueous leakage still occurs [19].

A few promising routes for the fabrication of KPros have been proposed. The Osteo–Odonto–KPro (OOKP) uses tooth enamel—an hydroxyapatite of biological origin—for the production of the skirt. A central polymethyl methacrylate (PMMA) optic is glued into the enamel tooth. OOKPs have been implanted since the 1960s. Falcinelli et al. [7] found excellent long-term retention results (85% survival in 18 years) with 75% of patients seeing 6/12 or better. Liu et al. [20] reported an 80% improvement of vision, with 60% reaching 6/24 and 47% 6/12. However, poor tissue integration results in failure of the devices [3]. Furthermore, it should be noted that it would be desirable to restore the metabolic activity of the cornea by use of a more bioactive material.

Besides KPros based on biological hydroxyapatite, which need to be obtained from tooth enamel—a procedure that requires the donation of one of the patient's teeth—synthetic devices have been investigated as they promise easier fabrication. In addition, it is clinically easier to implant a polymeric skirt than rigid skirts, such as OOKP. Furthermore, one can expect the mechanical compatibility to be improved between the sclera and polymers compared to the sclera and tooth enamel.

Currently, there are two synthetic KPros available commercially: the Legeais BioKpro III (FCI Ophthalmics, Marshfield Hills, MA, USA) and the AlphaCor (Argus Biomedical, Perth, Australia). The porous materials used as the skirt surround for tissue integration are, respectively, opaque fluorocarbon polytetrafluoroethylene (PTFE) in the Legeais BioKpro III and porous polyhydroxyethyl methacrylate (PHEMA) in the AlphaCor. Both of these synthetic devices can be produced fairly simply. BioKpro I and II implanted in patients had failure rates of 40% within one year, with retraction of the surface tissues over the haptic element of the device, retroprosthesis membrane formation, infection or aqueous leakage, and removal of the device was necessary [5]. Hollick et al. [19] recently reported that when BioKpro III was implanted in seven patients, only one was successful at 5 years with improved vision of 6/12, though presenting mucus accumulation on the optic; six patients had extrusions; three had retroprosthesis membranes; and one experienced inflammation. The AlphaCor implanted in 40 patients reviewed at 3 years presented complications associated with poor tissue integration in 12 patients.

Thus, it is clear that the rejection rates for polymeric KPro skirts is unacceptably high compared to biologically derived apatite in the OOKP, which seems to work well, but requires the loss of one of the patient's teeth. Thus, the need exists to find solutions for the fabrication of KPros based on synthetic polymeric materials that lead to the desirable clinical results of the OOKP. We therefore set out to increase the bioactivity of polymer skirts by coating them with a thin layer of hydroxyapatite. The use of

either hydroxyapatite polymer composites or plain hydroxyapatite were both considered but plain hydroxyapatite is brittle, which is highly undesirable in the eye, and composites do not show as high bioactivity as tooth mineral in *in vitro* cell culture studies [3].

Calcium phosphates, such as hydroxyapatite, can be readily prepared by precipitation from supersaturated aqueous solutions comprising calcium and phosphate-containing salts [21]. In the last decade, different strengths of simulated body fluid (SBF) have often been used *in vitro* to aid apatite formation or precipitation on the surfaces of different types of biomaterials, such as metals, polymers and ceramics [22–28].

In order to gain better understanding of the apatite nucleation and growth on polymer surfaces, we, therefore, have studied the apatite formed on polyether ether ketone (PEEK), high-density polyethylene (HDPE) and ultra-high molecular weight polyethylene (UHMWPE) films in SBF with different strength NaOH pre-treatments, with the objective of improving ophthalmic biomaterials related to KPro skirts, for which poor tissue integration, membrane formation and extrusion still occurs.

2. Materials and methods

2.1. Preparation of SBF

SBF with ion concentrations 1.5 times those of SBF-K9 developed by Kokubo et al. [26] was prepared by dissolving reagent-grade NaCl, NaHCO₃, KCl, K₂HPO₄ · 3H₂O, MgCl₂ · 6H₂O, CaCl₂ · 2H₂O, Na₂SO₄ and (CH₂OH)₃CNH₂ in ultrapurified water, and the solution was buffered with HCl to pH 7.20 at 37°C. The SBF solution was poured into a glass bottle and stored in a 4 °C fridge for 3 days to ensure that no precipitation occurred, which is an indication that the SBF was in an appropriate condition for use. The ion concentrations in human plasma and 1.0 and 1.5 SBF are listed in Table 1.

2.2. Preparation of the coatings

Three types of polymers were used through this work: PEEK films supplied by Invibio Ltd. Lancashire, UK; HDPE supplied as granules (SABIC Europe Ltd, Belgium); and UHMWPE supplied as powder (Himont, USA). HDPE and UHMWPE films were produced by compression moulding in a hydraulic press, preheated at 180 °C, applying a load of 10–20 kN for 3–10 min. Polyimide sheets were used as release films to prevent contamination of the samples by the mould.

The PEEK, HDPE and UHMWPE films were cut into discs 5 mm in diameter and with thicknesses of 0.09, 0.06 and 0.26 mm, respectively; they were washed with ethanol and ultra-high purified water, and dried overnight at room temperature. The dried samples were immersed in 1, 5 or 10 M NaOH solutions for 48 h. The NaOH-treated samples and untreated films were then washed and immersed

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