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Short communication

Enhancement of osteogenesis on micro/nano-topographical carbon fiber-reinforced polyetheretherketone–nanohydroxyapatite biocomposite

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ABSTRACT

As an FDA-approved implantable material, carbon fiber-reinforced polyetheretherketone (CFRPEEK) possesses excellent mechanical properties similar to those of human cortical bone and is a prime candidate to replace conventional metallic implants. The bioinertness and inferior osteogenic properties of CFRPEEK, however, limit its clinical application as orthopedic/dental implants. The present work aimed at developing a novel carbon fiber-reinforced polyetheretherketone–nanohydroxyapatite (PEEK/CF/n-HA) ternary biocomposite with micro/nano-topographical surface for the enhancement of the osteogenesis as a potential bioactive material for bone grafting and bone tissue-engineering applications. The combined modification of oxygen plasma and sand-blasting could improve the hydrophily and generate micro/nano-topographical structures on the surface of the CFRPEEK-based ternary biocomposite. The results clearly showcased that the micro-/nano-topographical PEEK/n-HA/CF ternary biocomposite demonstrated the outstanding ability to promote the proliferation and differentiation of MG-63 cells *in vitro* as well as to boost the osseointegration between implant and bone *in vivo*, thereby boding well application to bone tissue engineering.

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1. Introduction

Carbon fiber-reinforced polyetheretherketone (CFRPEEK) is becoming a primary candidate to replace metallic implants, since CFRPEEK composite has an adjustable mechanical property close to that of human cortical bone, which can mitigate concerns over the risks of bone resorption caused by stress shielding as a result of elasticity mismatch between the implants and human bones [1,2]. Besides, CFRPEEK inherits the non-toxicity, good chemical resistance, natural radiolucency, and even MRI compatibility of polyetheretherketone (PEEK) [3]. However, although the materials have attracted much attention as orthopedic/dental implants since 1980s, the bioinertness of CFRPEEK impedes osteointegration after implantation thereby severely hampering its clinical adoption.

Integration of implants (such as dental implant and total joint replacement prosthesis) with the surrounding bone, a process termed osseointegration, is important for successful bone regeneration and healing in dental and orthopedic applications. The desire to accelerate and improve osseointegration drives many implantology research and development efforts, particularly for patients whose bones have been compromised by disease or age. To improve the bioactivity of PEEK-based materials, many researchers have tried to make PEEK-based hybrid materials using nanohydroxyapatite (n-HA, $Ca_{10}(PO_4)_6(OH)_2$) because n-HA is a constituent of living bone [4,5] and is considered as a superb promoter to osteogenic differentiation of bone cells [6,7]. Simple PEEK/HA binary mixtures show better biocompatibility, but critical fractures often occur due to the debonding between HA and PEEK, a reduced ultimate tensile strength, and a decreased fatigue limit [4], along with the confined osseointegration. Recent effort in the field of bone tissue engineering highlights the importance of ternary composite in compensating shortcomings originated from conventional binary composite and promoting advanced mechanical properties and bioactivity of materials to better mimic the constituents and structure of natural bone [8–10]. Feng et al. fabricated a carbon fiber-reinforced polyetheretherketone–hydroxyapatite (PEEK/n-HA/CF) ternary composite and their results suggested that the strength and modulus of the composite were improved significantly with the inclusion of carbon fiber [11], nevertheless, to the best of our knowledge, no previous studies have been reported on the surface modification of the ternary composite, and the *in vitro* osteoblast responses (such as cell attachment, proliferation, and differentiation) and *in vivo* osseointegration to the PEEK/n-HA/CF biocomposite.

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Surface topographical modifications of implant at micrometer scale, such as those that are induced by acid etching or sand-blasting, have been used effectively to enhance osteoblastic lineage cell differentiation *in vitro* [12] and osseointegration *in vivo* [13], and have been clinically [14] compared to smoother surfaces. Currently, the addition of nanostructures onto implant interfaces such as plasma treatment, to better mimic the hierarchical structure of the bone, has also shown promising results *in vitro* [15–17] and *in vivo* [18], validating the biological relevance of nanotopography for bone formation. Moreover, oxygen plasma modification is known to increase the hydrophily without altering the bulk properties [19], resulting in creating an environment conducive to cell adhesion [20,21] and bone ingrowth [22]. Hence, in this study, we innovatively developed a PEEK/n-HA/CF ternary biocomposite modified *via* the combined treatments of oxygen plasma and sand-blasting to obtain micro-/nano-topographical surface for enhanced biocompatibility and osseointegration, which would hold a promising in bone/dental repair even in the future biomedicine applications.

2. Material and methods

PEEK/n-HA/CF biocomposite was fabricated containing 25 wt.% n-HA (<200 nm particle size, Sigma, USA), 15 wt.% CF (T700SC yarns of 12000 filament count, Toray, Japan) and 60 wt.% PEEK (450 G, the density about 1.30 g/cm^3, Victrex, UK) by a compounding and injection-molding process. In brief, n-HA powder, carbon fiber and PEEK powder were dispersed in alcohol using a ball grinder to obtain a homogeneous mixture. After well dispersed, the mixture was dried in a forced convection oven at 90 °C for 24 h to remove the excess alcohol. The ternary composites were produced at an injection temperature of 380 °C using an injection-molding machine (BA-300/050CD, Battenfeld, Belgium). The resulting composite was injected into two specially designed molds, *i.e.*, disks ($\Phi = 15 \times 2$ mm^3) for physical and chemical characterization and *in vitro* testing, and cylindrical implants (4.0 mm diameter and 7.0 mm length) for *in vivo* measurement. Samples of PEEK/n-HA/CF composite were polished with a series of increasing abrasive papers (400, 1000, 1500, 2000 grit), cleaned ultrasonically for 20 min in baths of acetone, anhydrous ethanol and D.I. water, respectively, and then dried at 50 °C overnight.

In order to superimpose micro-/nano-topography on the surface, samples firstly were processed in an Empire Suction Blasting Cabinet (JG-5832, Jing-Gong Medical Co. Ltd) by the blasting media flow with 110–150 μm size of TiO_2 particles in a distance of approximately 50 mm for duration of 60 s. Then the samples were treated by oxygen plasma treatment for 15 min (gas-flow rate 30 cm^3/min) at a gas pressure of 54.5 Pa and a bias voltage of −250 V, which were named as p-m-PEEK/n-HA/CF (plasma-modified micro-structured PEEK/n-HA/CF). The smooth PEEK/n-HA/CF samples that were only treated by oxygen plasma were denoted as p-PEEK/n-HA/CF (plasma-modified smooth PEEK/n-HA/CF), whereas bare PEEK/n-HA/CF samples in absence of treatments were as control groups.

The alteration of chemical constituents and wettability of the treated biocomposites were analyzed by XPS (Kratos, USA) and contact angle measuring device (SL200B, Kono, USA), respectively. The binding energies were calibrated by the C 1s hydrocarbon peak at 284.8 eV, and the quantitative analysis and the curve fitting were conducted by the CasaXPS software package. The morphology and surface roughness of samples were assessed by FE-SEM (HITACHI S-4800, Japan) and a mechanical profilometer (Dektek8 stylus profiler, Veeco, USA).

In the cell and animal experiments, the commercial pure titanium (Ti, grade 2, purchased from Northwest Institute for Non-ferrous

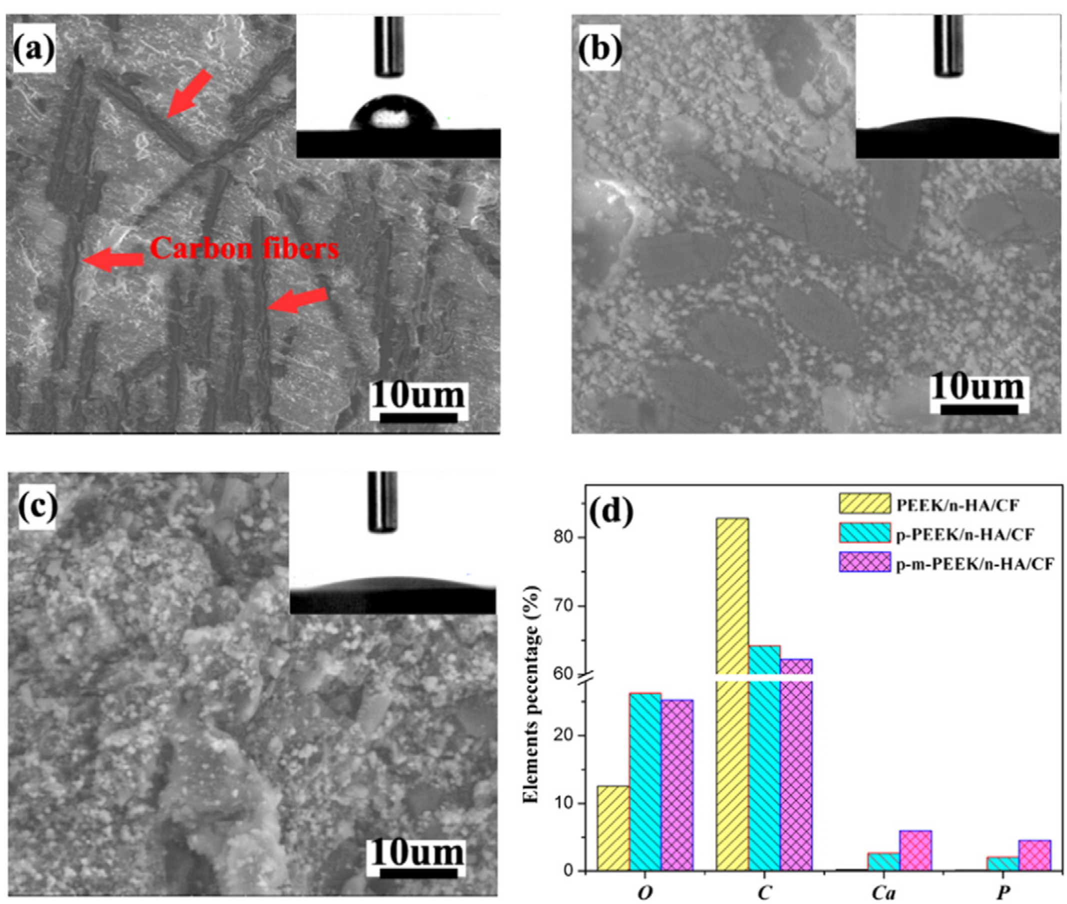

Fig. 1. Surface morphologies and contact angles of (a) PEEK/n-HA/CF, (b) p-PEEK/n-HA/CF, and (c) p-m-PEEK/n-HA/CF; (d) chemical compositions determined by XPS analysis of these biocomposites.

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