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Synthesis and characterization of poly (vinylidene fluoride)–calcium phosphate composite for potential tissue engineering applications

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Abstract

Tricalcium phosphate is the most commonly used biomaterial because of its good biocompatibility. However, its poor mechanical strength under complex stress and powder-like appearance hinder its application. The use of a composite biomaterial which maintains a fine balance between excellent mechanical properties and biocompatibility can be a solution to this problem. In the present study, we aimed to fabricate porous composite scaffolds via phase-inversion technique without using any additional toxic chemicals which can interfere with their biocompatibility. The composite materials fabricated of poly (vinylidene fluoride) and tricalcium phosphates were prepared, using polyvinyl pyrrolidone as a dispersant. The resulting scaffolds were characterized by using attenuated total reflection infrared spectroscopy (FTIR-ATR), scanning electron microscopy (SEM), thermo gravimetric analysis (TGA), differential scanning calorimetry (DSC), X-ray diffraction (XRD) and universal tensile strength (UTM) analysis. The composites showed well blend of materials and internal porous structures. The XRD results indicated a mixture of α and β-phases due to successful incorporation of tricalcium phosphate in polymer blends, thereby, exhibiting a crystalline structure. The fabricated composites showed an efficient thermal stability at around 400 °C. The tensile strength of scaffolds increased from 140 ± 1.6 to 148 ± 2.2 g/mm^2, which makes the composite scaffold potential candidate for hard tissue applications.
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Keywords: Poly (vinylidene fluoride); Calcium phosphate; Scaffolds; Bio-composite; Tensile strength

1. Introduction

Recently, regenerative medicine has perfectly aided biomedical sciences by introducing new avenues for efficient tissue reconstruction. Moreover, this has influenced lot of attention towards biomaterial science thereby influencing tissue engineering offering new and promising alternatives for existing traditional implants [1], prostheses in orthopedics [2], dentistry [3] and organ and tissue replacement [4]. Further, among various existing biomaterials such as bovine bone, recent trend shows that calcium phosphate is often used as bone substitute mainly due to its chemical similarities with that of mineral component of bones and hard tissues, therefore, proved to improve bone healing in rabbit calvaria [5]. In this regard, lots of approaches are considered for extraction of calcium phosphate from bovine bones [6,7]. Besides having the aforementioned importance, calcium phosphate has low density, high chemical stability, high wear resistance and excellent biocompatibility [8]. However, there still exist some limitations for using calcium phosphate directly such as lack of elastic properties which reduces its mechanical strength needed for its final application.

Composite technology finds solution to these problems with synthesized materials having good mechanical and biological compatibilities which can meet the requirements for tissue engineering applications. Therefore, composites including polymeric materials have been successfully and vigorously promoted as possible alternative for orthopedic replacements. Some successful cases of these materials include poly (L-lactic acid) (PLLA) [9], poly (glycolic acid) (PGA) [10], poly (D, L lactic acid-co-glycolic

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acid) (PLGA) [11], polyethylene [12], polyamide [13], cellulose [14], collagen [15] polyetheretherketone (PEEK) [16], polyaryletherketone (PAEK) [17] and polysulfone [18]. All these aforementioned polymers can be used as suitable blends for preparation of composite materials used for tissue engineering applications.

Poly (vinylidene fluoride) (PVDF) is a semi-crystalline ferroelectric polymer [19,20], which can be easily processed, has high chemical resistance, excellent mechanical properties and good thermal stability [21]. Hence, it has been used for a wide range of scientific and technological applications; it had been used for from simple protective coatings for pipes, transducer devices, smart materials, and ferroelectric memory devices [22]. Furthermore, PVDF is also found to have potential applications in biomedicine by serving as a nerve guidance channels in neural tissue regeneration [23] and, in the case of cell culture studies, as a cell culture substrate to maintain cellular functions and promote adhesive functionality [24,25]. The ease at which PVDF can be handled as a monofilament and its resistance towards biodegradation makes it an attractive alternative for a suture material in performing cardiovascular surgery [26]. It has been proven to have good performance as compared to polypropylene (PP) in tendon repair surgery [27]. It is worth to mention, that when PVDF was used as monofilament sutures, for tendon repair, it gave a minimal tissue response, long-term stability and excellent pliability [27].

The idea of making PVDF–calcium phosphate composites is to combine biological affinity of calcium phosphate and the good tensile properties of PVDF to obtain a desirable material which can be applied in tissue engineering. The advantage of using composite material for tissue implant is that by varying the type and distribution of the reinforcing phase in the composite can be varied. So, it is highly possible to obtain a wide range of mechanical and biological properties, and hence to optimize the structure of the implant and its interaction with the surrounding tissues [28]. Finally, the composite scaffold containing tricalcium phosphate does not only promote osteoblast adhesion and proliferation but also can help to retain high resistance and durability, due to the presence of PVDF.

In this study, we aimed to introduce porous PVDF scaffolds containing TCP for future hard tissue engineering application. This study involves the use of a phase-inversion method to prepare PVDF/calcium phosphate composites as a potential candidate for bone tissue application. The composites were characterized by attenuated total reflection infrared spectroscopy (FTIR-ATR), X-ray diffraction (XRD), thermo gravimetric analysis (TGA), differential scanning calorimetry (DSC), scanning electron microscopy (SEM) and universal tensile strength (UTM).

2. Experimental

2.1. Materials

Polyvinylidene fluoride (PVDF, M_w ca. 2.2×10^5 g/mol) (T1100) was purchased in a powder form from Kureha Chemical Industry, Japan. N-methyl-2-pyrrolidone (NMP), 99.0% purity, was purchased from Kanto Chemical, Japan. Polyvinylpyrrolidone (PVP, M_w ca. 2.5×10^5 g/mol) was obtained from Sigma-Aldrich, USA. Calcium phosphate tribasic ($Ca_3(PO_4)_2$) was from Samchun Chemicals, Republic of Korea. All these chemicals were used without further purification.

2.2. Scaffold preparation

Pre-dried PVDF pellets (in vacuum oven) were dissolved in N-methyl-2-pyrrolidone (NMP) solution under vigorous stirring at 80 °C while calcium phosphate tribasic powder was carefully dispersed separately in NMP solution. In order to get a good dispersion of the inorganic particles, PVP (1 wt% of total solution) which served as a dispersant together with ultrasonic vibrations was used to obtain emulsion mixture. Both solutions were mixed together and further stirred to obtain a homogeneous mixture. The casting solution was degassed and composite scaffolds were prepared via phase-inversion method in water bath. The calcium phosphate tribasic (TCP) content was varied at 0, 10, 20 and 30 wt % with respect to PVDF. It is worthy to mention that TCP when used more than 30 wt% with respect to PVDF resulted in high precipitation. Therefore, maximum 30 wt% of TCP was used to modify the PVDF in this study. Furthermore, the scaffolds were soaked in distilled water to remove the associated residual solvents and finally dried in an oven.

2.3. Characterization of composite scaffolds

The chemical structures of composite scaffolds were observed using attenuated total reflection (FTIR-ATR) spectra using Varian 2000 spectrometer from 600 to 4000 cm^{-1} with 2 cm^{-1} resolution and 32 scans in transmittance mode. The crystal structures of the scaffolds were analyzed using an X-ray diffraction (PANalytical diffractometer, X'pert-Pro) with CuKα radiation with Cu, Cr ($\lambda = 1.540$ A) radiation over Bragg angle ranging from 10° to 60°. Thermal properties of the scaffolds were studied by TGA (SETARAM 92) and DSC (TA instrument Q10). TGA was conducted under nitrogen atmosphere from 50 °C to 1000 °C at a heating rate of 10 °C/min. The samples for DSC were sealed in aluminum pan, heated from 50 °C to 220 °C at a rate of 10 °C/min held for 10 min, and then cooled to 50 °C at a rate of 10 °C/min under a nitrogen atmosphere. The surface and internal morphologies of scaffolds were investigated by scanning electron microscopy (Hitachi S-3500N). Samples were fractured in liquid nitrogen to reveal the cross-section morphology and sputter coated with thin layer of platinum for 180 s at two consecutive cycles at 45 mA prior to imaging. The micrographs of each sample were taken at an accelerating voltage of 25 kV, at 4 K and 0.3 K magnifications, for surface and cross-section viewing, respectively. Finally, the tensile strength of the samples were measured using Universal Testing Machine (Lloyd Instruments™, Ametex®, Inc.; England). All values from the mechanical test were averaged over five measurements and represented in this paper.

3. Results and discussion

Fig. 1 shows the FTIR/ATR spectra of the pristine PVDF scaffold and TCP. The bands appearing at 642 cm^{-1} are due to CH_2 rocking and asymmetric stretching and at 880, 1072, 1180

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