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# Bioactivity of sol–gel-derived $TiO_2$ coating on polyetheretherketone: In vitro and in vivo studies

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ABSTRACT

A polyetheretherketone (PEEK) surface was modified using a sol–gel-derived $TiO_2$ coating in order to confer bone-bonding ability. To enhance the bonding strength of the coating layer, pretreatment with either $O_2$ plasma or sandblasting was performed prior to sol–gel coating. Additionally, post-treatment with acid was carried out to confer apatite (calcium phosphate)-forming ability to the surface. Biomechanical and histological analyses performed using an in vivo rabbit tibia model showed that PEEK surfaces modified with sol–gel-derived $TiO_2$ and acid post-treatment had better bone-bonding properties than uncoated PEEK surfaces. These modified surfaces also performed well in terms of their in vitro cell responses due to their modified surface chemistries and topographies. Although $O_2$ plasma or sandblasting treatment were, for the most part, equivocal in terms of performance, we conclude that sol–gel-derived $TiO_2$ coating followed by acid post-treatment significantly improves the bone bonding ability of PEEK surfaces, thus rendering them optimal for their use in surgical implants.

**Statement of Significance**

The role of polyetheretherketone (PEEK) as an alternative biomaterial to conventional metallic implant materials has become increasingly important. However, its low bone bonding ability is yet to be resolved. This in vivo and in vitro investigation on the functionalization of PEEK surfaces highlights the utility of this material in clinical interventions that require implants, and may extend range of applications of PEEK.



## 1. Introduction

Two decades have passed since polyether-ether-ketone (PEEK) emerged as a leading high-performance thermoplastic material that could be used instead of metal implants, particularly with regard to replacements in spine surgery. While PEEK has some clear advantages, such as radiolucency and low elastic modulus that is close to the corresponding value for human bone, concerns have been raised about its low bioactivity due to its chemical inertness [1]. In fact, some clinical reports indicate that PEEK is inferior to titanium over long-term usage [2,3]. Thus, further improvements to the bioactivity associated with PEEK surfaces are required if the clinical potential of this material is to be fully realized.

Several studies have described surface coating approaches aimed at increasing the bioactivity of PEEK surfaces; these include plasma-sprayed hydroxyapatite (HA) and titanium coating [4–7]. However, HA coatings are susceptible to degradation over long time periods [8] and there are enduring concerns regarding titanium and its low bonding strength; this is since titanium requires a thick coating layer [6]. In contrast, the sol–gel method provides extremely thin uniform oxide layers that are deposited on the material surface and never degrade [9]. Furthermore, the temperature used in the process of sol–gel coating is significantly lower than the above-mentioned coating technique and, therefore, does not exceed the glass transition temperature of PEEK (about

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143 °C). We have previously reported the bioactivity of the sol–gel derived $TiO_2$ layer on polyethylene terephthalate (PET) in vivo [10]. To our knowledge, however, there have been no publications investigating the effect of sol–gel-derived $TiO_2$ layers on PEEK surfaces.

Material bioactivity (which hereafter is used to describe its bone-bonding ability in vivo), is often evaluated using the response of osteoblasts or mesenchymal stem cells (MSC) in surrogate in vitro assays. These cellular responses reflect the surface characteristics of materials including both surface chemistry and topography [11–13] and are, therefore, useful for the preliminary evaluation of the surface characteristics. However, prediction of clinical in vivo bone bonding ability is most robustly estimated using animal models. Since the first development of bioactive glass ceramic material in 1991 [14], however, we have presented the theory that the most effective in vitro predictor of in vivo bone bonding ability is the apatite (calcium phosphate)-forming ability of material surfaces in simulated body fluid (SBF) [15]. Recently, we found that adding a sol–gel-derived $TiO_2$ layer to PEEK confers apatite forming ability in SBF if an acid post-treatment step is performed [16]. In the report, $O_2$ plasma pretreatment was applied before the sol–gel coating to ensure that the sol–gel layer adhered strongly to the PEEK surface. In the current study, we also evaluated whether sandblasting of $TiO_2$ particles onto PEEK could be an alternative pretreatment option that could facilitate the adhesion of sol–gel layers.

The aims of this study were (1) to evaluate the in vivo bone bonding ability of a sol–gel derived $TiO_2$ layer that was pretreated with either $O_2$ plasma or sandblasting and post-treated with acid, and (2) to determine the surface characteristics of $TiO_2$ layers and the cellular responses elicited by these surfaces; particular attention was focused on evaluating the apatite-forming ability of the surfaces.

## 2. Materials and methods

### 2.1. $TiO_2$ layer coating

Two types of form, a disc-shaped type measuring 18 mm × 2 mm or a plate-shaped type with 15 mm × 10 mm × 2 mm were cut from the PEEK substrate (TECAPEEK natural, Ensinger Gmbh, Germany: Poisson's ratio 0.4, specific gravity 1.3, flexural modulus 4.2 GPa, tensile strength 97 MPa) and used for in vitro and in vivo studies, respectively. These PEEK materials were coated with $TiO_2$ using one of 5 distinct processes, as described in Table 1 (Fig. 1A). The BH group, which was not coated with sol–gel, was included in this study because the sandblasted $TiO_2$ layer alone might have bioactivity following acid post-treatment.

#### 2.1.1. Pretreatment before sol–gel $TiO_2$ coating ($O_2$ plasma or sandblast treatment)

PEEK materials were polished with 800 grit Sic paper and then washed sequentially with 2-propanol and ultrapure water in an ultrasonic cleaner for 30 min. After polishing, substrates were subjected to $O_2$ plasma treatment or sandblast treatment before coating via the sol–gel method. In the $O_2$ plasma treatment, substrates were placed in the chamber of a polymerization system (PD-10S, SAMCO Inc., Japan). Microwave plasma at 100 watts was applied in an $O_2$ atmosphere with 50 Pa pressure for 5 min. In the sandblast treatment, $TiO_2$ particles with a median diameter of 7.62 μm (TOHO Titanium, Kanagawa, Japan) were blasted using a blast-gun with a pressure of 0.5 MPa for 30 s.

**Table 1**
PEEK implants and associated $TiO_2$ coating methods.

| PEEK implants | Pretreatment | Sol–gel $TiO_2$ coating | Acid (HCl) post-treatment |
|---|---|---|---|
| Uncoated | – | – | – |
| OS | $O_2$ plasma | + | – |
| OSH | $O_2$ plasma | + | + |
| BH | Sandblast | – | + |
| BSH | Sandblast | + | + |

#### 2.1.2. Sol–gel $TiO_2$ coating and post-treatment with acid (HCl)

After either pretreatment, PEEK materials were dipped into the $TiO_2$ sol solution consisting of titanium tetraisopropoxide (TTIP), $H_2O$, ethanol (EtOH), and nitric acid ($HNO_3$) with a TTIP:$H_2O$:EtOH:$HNO_3$ molar ratio of 1:1:37:0.1. The substrates were removed from the solution after 1 min at a rate of 1 cm/min and then air-dried at 80 °C for 24 h. After drying, materials were soaked in 0.1 M HCl solution at 80 °C for 24 h and then gently washed with ultrapure water.

### 2.2. Surface characterization

#### 2.2.1. Scanning electron microscopy (SEM)

Surface morphology and titanium distribution of coated and uncoated PEEK samples were examined by SEM with Energy Dispersive X-ray Spectroscopy (EDX) (S-4700; Hitachi Ltd, Tokyo, Japan) after coating with Pt/Pd.

#### 2.2.2. Water contact angle

The hydrophobic characteristics of PEEK samples were determined by measuring the water contact angle. A 4-μL droplet of ultrapure water was dropped onto the surface using a microsyringe. The shape of the droplet was observed and the contact angle was measured from a photographic image.

#### 2.2.3. Zeta potential measurements

The zeta potential of the PEEK materials was measured using a zeta potential analyzer (ELS-Z1, Otsuka Electronics Co., Japan). A rectangular PEEK specimen 33 × 15 $mm^2$ in size and 1 mm in thickness, which was coated with the $TiO_2$ gel layer, was used for the zeta potential measurement. Dispersant monitoring particles of polystyrene latex (size = 500 nm) coated with hydroxyl propyl cellulose were used. The zeta potential was measured with an applied voltage of 60 V in 10 mM NaCl solution.

#### 2.2.4. Surface roughness (micrometer scale)

A contact probe profilometer (Mitutoyo Surftest SV-2000) was used to measure the topography of the surface coatings of the micrometer scale. We note that this profilometer does not give the submicron-scale roughness measurements that were observed in SEM images because the probe has a 2-μm edge width, which lowers the resolution. The average surface roughness (Ra) of implants coated with each method was measured. Ra in the direction perpendicular to the polished direction was measured at five areas of each implant. The average Ra was calculated from these five areas, and is represented as the sample mean ± standard deviation.

#### 2.2.5. Apatite-forming ability of surfaces

The apatite-forming ability of the samples was examined by soaking them in SBF [15,16] at pH 7.40 for 3 d at 36.5 °C. Soaking at 36.5 ± 0.5 °C is recommended by ISO 23317, and SBF has been shown to generate reproducible results [17]. The ion concentrations (all in mM) were as follows: $Na^+$, 142.0; $K^+$, 5.0; $Ca^{2+}$, 2.5; $Mg^{2+}$, 1.5; $Cl^-$, 147.8; $HCO^{3-}$, 4.2; $HPO_4^{2-}$, 1.0; $SO_4^{2-}$, 0.5. The samples were removed from the SBF, washed with distilled water, and dried on a clean bench. Their surfaces were examined with SEM and SEM–EDX, and apatite formation was determined by