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Basic Science

Optimizing surface characteristics for cell adhesion and proliferation on titanium plasma spray coatings on polyetheretherketone

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Abstract

BACKGROUND: Titanium plasma spray coating on polyetheretherketone (PEEK) is a recent innovation to interbody spacer technology. The inherent hydrophobic properties of standard, uncoated PEEK implants can hamper cell attachment and bone healing during fusion. The addition of titanium coating not only offers initial stability due to increased surface roughness but also long-term stability due to bony ongrowth created from osteoconductive microenvironment on the device surface. The previously established hydrophilic and osteophilic properties of commercially pure titanium (CPTi) can potentially provide an ideal environment promoting cell attachment and bony ongrowth when applied at the end plate level of the fusion site. Because the surface material composition and topography is what seems to directly affect cell adhesion, it is important to determine the ideal titanium coating for the highest effectiveness.

PURPOSE: The purpose of the study is to determine whether there is an optimal surface roughness for the titanium coatings and whether different polishing methods have a greater effect than roughness or topography in mediating cell adhesion to the surface.

STUDY DESIGN/SETTING: The study was divided into two phases. In Phase 1, the effects of varying surface roughnesses on identical polishing method were compared. In Phase 2, the effect of varying polishing methods was compared on identical surface roughnesses.

METHODS: Coating thickness, porosity, and surface roughness were characterized using an optical microscope as per ASTM F 1854 standards. For both phases, PEEK coupons with plasma-sprayed CPTi were used, and human mesenchymal stem cells (hMSCs) at an initial density of 25,000 cells/ cm² were seeded and cultured for 24 hours before fixation in 10% formalin. The cultured hMSCs were visualized by 4',6-diamidino-2-phenylindole (DAPI) staining, a fluorescent stain that binds to the DNA of living cells. Samples were imaged using an environmental scanning electron microscope (eSEM) (Carl Zeiss Microscopy, Thornwood, NY, USA) using a backscattered detector.

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RESULTS: Image analysis of the CPTi coatings showed uniform and rough surfaces. For Phase 1, roughness was evaluated as fine, medium, and coarse. The eSEM image analysis and cell counting by DAPI demonstrated that hMSCs have a tendency to form stronger adhesion and greater pseudo-podia extensions on fine roughness surfaces. Individual hMSCs were seen forming cytoplasmic processes extending across the width of a pore. There was a 4- and 20-fold reduction in adhered hMSCs with an increase to medium and coarse roughnesses, respectively. For Phase 2, studied groups are (1) medium CPTi coating with zirconia polishing, (2) medium CPTi coating with CPTi polishing, and (3) fine CPTi coating with CPTi polishing. The eSEM image analysis and cell counting by DAPI demonstrated that hMSCs have a tendency to form stronger adhesion and greater pseudopodia extensions on Group 3 over the other two groups. There was a twofold reduction in adhered hMSCs on medium roughness relative to fine. No difference in cell adhesion was found between Groups 1 and 2. Individual hMSCs were seen forming cytoplasmic processes extending across the width of a pore. **CONCLUSIONS:** Previously, it was accepted without much scrutiny that surface coatings were beneficial. This study begins to discover that surface topography directly affects the potential for cells to adhere and proliferate and lead to greater surgical efficacy. © 2016 Elsevier Inc. All rights reserved.

Keywords:

Cell adhesion; Instrumentation; PEEK; Polyetheretherketone; Surface coating; Titanium

Introduction

Spinal fusion with interbody cages is a surgical procedure designed to stabilize the spinal column for disorders such as degenerative disc disease, abnormal curvature, traumatic instability, degenerative instability, and damage from infections or tumors. The interbody device is often used with embedded graft material to bridge the levels and provide anterior column support. Metallic interbody devices are manufactured from materials such as commercially pure titanium (CPTi), titanium alloys, and cobalt chromium (CoCr) alloys. These materials provide mechanical strength and can be finished to provide friction [1]. However, subsidence in metal interbody devices remains a challenge and can increase the risk of loosening or failure of the device mostly related to stress shielding effect [2].

Implant subsidence can be caused by excessive implant stiffness, inadequate implant surface area of bearing surfaces, and a large difference in Young's moduli between the device material's composition and human bone tissues. Most CPTi have a Young's modulus of 116 GPa and cobalt chromium alloys have 210 GPa, which are much higher than the 17–21 GPa of human cortical bone and 10–14 GPa of human trabecular bone [3]. The biomaterials used and the design of interbody devices have undergone a rapid evolution to address this concern, and polyetheretherketone (PEEK) is a viable alternative to metals because of its elastic modulus ranging between 4 and 24 GPa, depending on manufacturing specifications [4]. Polyetheretherketone has also garnered popularity because of its toughness, radiolucent nature, biocompatibility, and relative ease in manufacturing [5].

However, unlike conventional materials like titanium, PEEK does not possess osteoconductive or osteoinductive properties [6]. It has a hydrophobic surface that prevents bone apposition, which increases the risk of long-term sequelae such as micromotion [7]. Micromotion is believed to trigger the formation of fibrous biofilm between the bone and the implant, which lowers the implant's long-term stability [8–10]. As a remedy, coating technologies have been developed to provide a thin layer (80–100 μ m) of more bioactive materials like CPTi, titanium alloy, or hydroxyapatite on traditional metallic substrates. Previous investigations have shown that such coating indeed increases the mechanical retention and bone-to-implant contact [11,12].

Recently, titanium coating on PEEK substrates has become commercially available. It creates an osteoconductive surface that provides short-term stability due to friction, and longterm stability due to cell adhesion leading to bony ongrowth. Although higher surface coating roughness maximizes the initial friction and short-term stability, it may be less optimal for cell viability and long-term stability. In addition, previous investigations in dentistry have shown that zirconia (ZiO₂) polished implants demonstrated increased osteogenic response [13,14]. Currently, there are limited data available on the interaction between surface roughness, polishing, and osseointegration. The objective of the study was to determine the optimal range for surface roughness to promote cellcell interaction and adhesion, and whether polishing media changed the cellular response. The hypothesis was that longterm implant stability is directly related to cellular adhesion that is modulated by surface roughness by a greater extent than polishing media. The hypothesis was tested by using coupons of titanium plasma spray coating on PEEK with (1) various roughnesses of the coating and (2) differing polishing methods.

Materials and methods

Polyetheretherketone coupons at 25.5 mm diameter and 6.25 mm thickness were used in this study (Surface Dynamics, Cincinnati, OH, USA). Each coupon was grit blasted to increase the substrate surface roughness. The samples were then coated in an atmospheric vacuum chamber with CPTi powder via plasma spray processes. Post-coating, the samples

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