Improved Bioactivity and Bacteriostasis of PEEK Polymers by Addition of Silicon Nitride Particles

Giuseppe Pezzotti¹⁻⁵, Wenliang Zhu⁶, Elia Marin¹, Alfredo Rondinella¹, Francesco Boschetto¹, Ryan M. Bock⁷, Bryan J. McEntire⁷, and B. Sonny Bal^{7,8}

¹Ceramic Physics Laboratory, Kyoto Institute of Technology, Kyoto, Japan; ²Department of Orthopedic Surgery, Tokyo Medical University, Tokyo, Japan; ³The Center for Advanced Medical Engineering and Informatics, Osaka University, Osaka, Japan; ⁴Department of Molecular Cell Physiology, Graduate School of Medical Science, Kyoto Prefectural University of Medicine, Kyoto, Japan; ⁵The Center for Advanced Insect Research Promotion (CAIRP), Kyoto Institute of Technology, Kyoto, Japan; ⁶Department of Medical Engineering for Treatment of Bone and Joint Disorders, Osaka University, Osaka, Japan; ⁷Amedica Corporation, Salt Lake City, UT, USA; ⁸Department of Orthopaedic Surgery, University of Missouri, Columbia, MO, USA

Disclosures: Giuseppe Pezzotti (3B-Amedica Corp.), Wenliang Zhu (N), Elia Marin (N), Alfredo Rondinella (N), Francesco Boschetto (N), Ryan M. Bock, Bryan J. McEntire, and B. Sonny Bal (3A and 4-Amedica Corp.)

Introduction: Biomaterials that support appositional bone ingrowth and concurrently resist bacterial adhesion and biofilm formation are advantageous for use in spinal fusion surgery. Although polyetheretherketone (PEEK) is widely used as an interbody spacer, the material has poor osteoconductive and bacteriostatic properties.^{1, 2} In contrast, monolithic silicon nitride (Si₃N₄) has shown enhanced osteogenic and antimicrobial behavior.^{3, 4} Therefore, it was hypothesized that a PEEK-Si₃N₄ composite might improve bone bonding while impeding biofilm formation.

Methods: A PEEK polymer (Otsuka Chemical) was melted and compounded with three different silicon nitride powders at 15 wt.%, including: (i) α-Si₃N₄; (ii) a liquid phase sintered (LPS) β -Si₁N₄; and (iii) a β -SiYAlON mixture. These three ceramic powders had slightly different solubilities, polymorphic struc-

tures, and/or chemical compositions. Osteoconductivity was assessed by seeding specimens with 5×10^5 /ml of SaOS-2 osteosarcoma cells within an osteogenic media for 7 days. Antibacterial behavior was determined by inoculating samples with 1 x 10⁷ CFU/ml of Staphylococcus epidermidis (S. epi.) in a 1 x 108/ml brain heart infusion (BHI) agar culture for 24 h. After staining with PureBlu™ Hoechst 33342 or with DAPI and CFDA for SaOS-2 cell adhesion or bacterial presence, respectively, samples were examined with a confocal fluorescence microscope (BZ-X700; Keyence) using a 488 nm Krypton/Argon laser source. Images were also acquired using a FEG-SEM in secondary and backscattered modes on gold sputter-coated specimens (~20-30Å, 108auto, Cressington). Hydroxyapatite (HAp) deposition was measured using a laser microscope (VK-X200 K Series, Kevence). Raman spectra were collected for samples in backscattering mode using a triple monochromator (T-64000, Jobin-Yvon, Horiba Group) using a 532 nm excitation source (Nd:YVO4 diode-pumped solid-state laser; SOC JUNO, Showa Optronics Co. Ltd.).

Results: PEEK composites with 15 wt.% α -Si₃N₄, LPS β -Si₃N₄, or the β -SiYAlON mixture showed significantly greater SaOS-2 cell proliferation (>600%, p<0.003, cf., Fig. 1(a)) and HAp deposition (>100%, p<0.003, cf., Fig. 1(b)) when compared to monolithic PEEK. The largest increase in cell proliferation was for the β-SiYAION composite, while the greatest amount of HAp was found on the LPS B-Si₃N₄ composite. With respect to the bacterial tests, the composite containing the LPS B-Si₃N₄ powder showed one order of magnitude reduction in adherent live bacteria (p<0.003, cf., Fig. 1(c)) as compared to solid PEEK. It is interesting to note that the composite containing α -Si₃N₄ had the worst bacterial resistance (*i.e.*, ~100% higher than monolithic PEEK), suggesting that the bacteriostatic effectiveness of Si₃N₄ bioceramics is apparently dependent upon the presence of selective sintering additives, including yttria and alumina.

Discussion: Monolithic PEEK has been utilized as an orthopedic biomaterial since the 1980s.⁵ However, because of its petroleum-based nature, it has neither effective bone apposition nor adequate bacteriostasis. Compounding PEEK with HAp particles or coating PEEK with HAp, titanium, or tantalum have been developed as potential solutions. Although improvements in osteoconductivity have been noted,⁶ these composite devices still lack anti-microbial resistance. Compounding PEEK with Si₃N₄ represents a significant advancement due to its ability to provide both improved bone apposition and bacteriostatic behavior.

Significance/Clinical Relevance: The addition of 15 wt.% of specific Si₃N₄ powders to PEEK showed enhanced SaOS-2 cell adhesion, proliferation, and HAp deposition when compared to monolithic PEEK. These same composites also showed resistance to S. epi. adhesion and biofilm formation. PEEK/Si₃N₄ composites may improve spinal fusion outcomes and offer an advanced alternative to other PEEK-based composites used as interbody spacer devices, such as PEEKhydroxyapatite and PEEK-titanium.

References: ¹K. Phan et al., J. Clinical Neurosci., 24 138-140 (2016); ²T. J. Webster, et al., Acta Biomater., 8 [12] 4447-4454 (2012); 3R. M. Bock et al., J. Biomed. Mater. Res. Part B Appl. Figure 1. (a) Fluorescence microscopy evaluation of Biomater., 1-13 (2017); ⁴R. M. Bock, et al., J. Biomed. Mater. Res. Part A, 105 [5] 1521-1534 SaOS-2 cell adhesion; (b) 3D laser microscopy results (2017); ⁵S. M. Kurtz and J. M. Devine, *Biomaterials*, 28 [32] 4845–4869 (2007); ⁶ I.V. Panayotov, et. al., J. Mater. Sci. Mater. Med., 27 [7] (2016).



for HAp deposition; and (c) CFDA/DAPI stain analysis for live S. epi. bacteria; n = 3 for each material and test; p < 0.003; p < 0.05.