

# Antimicrobial and Bone Growth Responses to Nanostructured Porous Tantalum Coatings on Ti and PEEK: An *in vivo* Study

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## ABSTRACT

The effects of nanostructured tantalum (Ta) coatings on antimicrobial responses and bone attachment were studied. Small Ta coated discs of titanium and PEEK were inoculated with *Staph. epidermis* both with and without presoaking in the antibiotic gentamycin. The discs were then implanted in rat calvarial defects and the push out forces after 1 week and 4 weeks were used as measures of bone growth. Uncoated discs and discs coated with hydroxyapatite were used as controls. In all cases the coated discs had greater push out forces than uncoated discs and the antibiotic soaked samples had greater push out forces than the samples without antibiotics. In the case of titanium substrates, the Ta and hydroxyapatite coatings had comparable push out forces and both were measurably better than uncoated titanium. In the case of PEEK substrates, the push out forces of the Ta coated samples were two to three times greater than the push out forces of the hydroxyapatite coated samples and both coated samples had significantly higher push out forces than bare PEEK.

## INTRODUCTION

Devices such as artificial hip, knee and spinal implants now provide a vastly improved quality of life for millions of people worldwide. Two key factors in the success of joint replacement operations are the ability of the body's natural bone to grow into or onto the prosthesis (known as osseointegration) and avoiding infection.

Presently available orthopedic implants have bone contacting elements with characteristic surface features that are on the order of tens of microns. In contrast, surfaces with features having dimensions of tens to hundreds of nanometers have greatly increased surface area, altered surface energy, and unique biological responses.

Osseointegration begins with the adsorption of fibronectin on the surface of the implant, which is part of the natural extracellular matrix (ECM) of bone that ultimately bonds the implant to the bone. Integrins (present on cellular mem-

branes) then mediate the growth and attachment of bone cells to the fibronectin. Previous work has shown that this process can be stimulated by creating implant surfaces with the correct nanostructured features independent of implant surface chemistry [1-5].

Similarly, nanostructures on implant surfaces have also been shown to affect the ability of bacteria to grow [6-8]. One of the most difficult types of infection to deal with is the formation of a bacterial biofilm, which is a colony of bacteria protected from the environment by a hydrated polymeric matrix [9]. This matrix makes the treatment of a bacterial biofilm much more difficult than treating planktonic bacteria responsible for acute infections. One such biofilm producer is *Staphylococcus aureus*, which is responsible for infections associated with catheters, orthopedic implants and even contact lenses for over eleven million patients annually [10]. Current methods to treat medical infections are not completely successful, as is evident by the increasing number of medical device infections and the growing number of antibiotic resistant bacteria (rendering such bacteria unaffected by today's antibiotic therapies).

To help combat the increasing number of medical device infections, the purpose of the work reported here was to study the effects of nanostructured tantalum (Ta) coatings on the growth of bacteria and bone *in vivo*. Ta is a biocompatible material in widespread use in today's orthopedic devices. We have previously reported the ability to sputter deposit Ta coatings having surface features of tens to hundreds of microns in size, which are extremely adherent to metal surfaces [11]. However, the ability of such nanostructured Ta surface features to increase bone formation and decrease bacteria growth remains untested.

## EXPERIMENTAL PROCEDURES

### Tantalum Deposition

Ta was sputter deposited onto small discs of either titanium (Ti) or polyether ether ketone (PEEK) that were approximately 4 mm in diameter by 1 mm thick. The deposition system has been described previously [11, 12]. It consists of two inverted

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cylindrical magnetron cathodes each having an inside diameter of 33 cm and a height of 10 cm, which were spaced 10 cm apart in the axial direction. Commercially pure (99.5%) Ta sputter targets were used and the power delivered to each cathode was 1.0 kW DC. In order to produce an extreme Zone 1 structure [13], the substrates were placed between the sputter targets to accentuate the oblique incidence arrival of the sputtered material. In addition, Kr sputter gas was used at a pressure of 8 mTorr to reduce the incident energy of the arriving species. The substrates were rotated in a dual planetary motion to assure uniform coating coverage. The resulting deposition rate was 33 nm per minute and the total thickness accumulated was 10 microns.

In order to estimate the pore size distribution of the coatings,  $N_2$  adsorption isotherms were measured. Samples were held at a temperature of 77K and the quantity of  $N_2$  adsorbed as a function of pressure was measured. The Barrett-Joyner-Halenda (BJH) model was then used to estimate the pore size distribution [14].

The crystallographic structure of Ta deposited under the conditions of these coatings was determined by performing a  $\theta$ - $2\theta$  scan with a Bragg-Brentano diffractometer using  $Cu K\alpha$  radiation with a diffracted beam monochromator and fixed slits.

#### Implant Preparation and Removal

Samples of both titanium and PEEK were prepared for animal studies using three different treatments: uncoated, coated with Ta and coated with hydroxyapatite. Hydroxyapatite is used as a coating on commercially available implants to stimulate bone growth and it served as a control in this experiment [9]. Samples prepared with each of these three treatments were then left as-is or soaked in gentamycin. Finally, all six variations were inoculated with  $10^5$  colony forming units of *Staph. epidermis* (obtained from ATCC) prior to implantation to invoke an infection. Three samples of each variation were then implanted into rat calvarial defects. The extent of bone in growth was determined by measuring the push-out force of the implants after 1 week and 4 weeks of growth [9, 15]. Experiments were repeated in triplicate.

## EXPERIMENTAL RESULTS

### Tantalum Coating Characterization

Scanning electron micrographs of a cross-section and the surface of coatings made under these deposition conditions are shown in Figure 1 and Figure 2, respectively. The extreme Zone 1 structure can be seen with individual columns having aspect ratios of approximately 50:1 or greater, separated from one another by voids.

The X-ray diffraction measurements showed greater than 95 % body centered cubic (or alpha) phase Ta with a strong [110] texture and less than 5 % tetragonal (or beta) phase Ta with a predominant [001] texture. These results are both

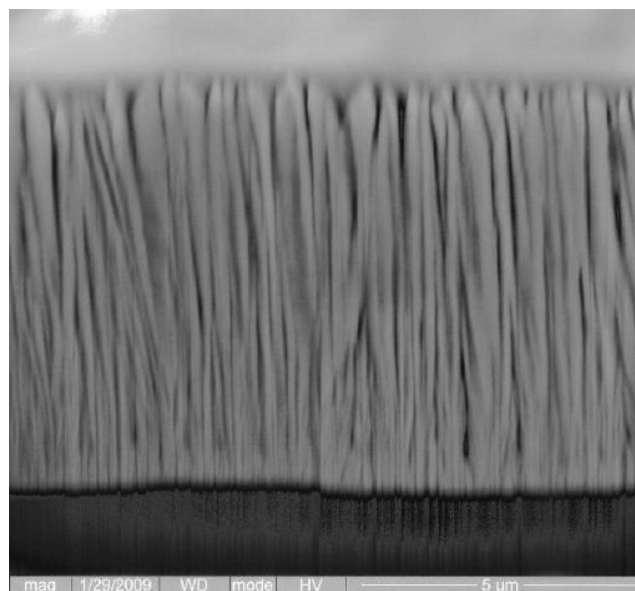


Figure 1: Scanning electron micrograph of the nanostructured tantalum coating, cross section. The coating thickness is 10 microns.

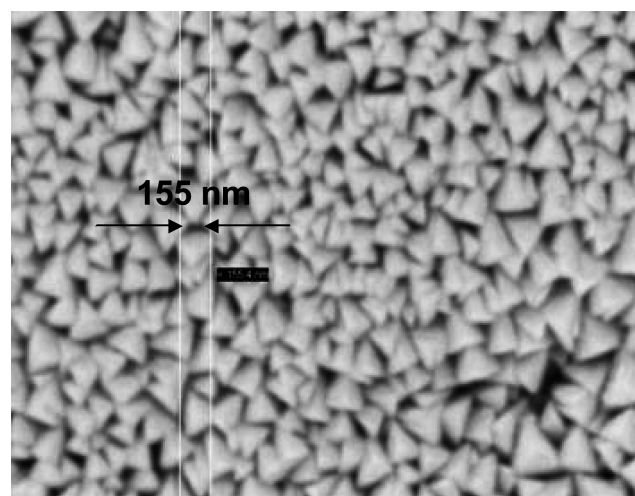


Figure 2: Scanning electron micrograph of nanostructured tantalum coating, top view.

surprising and advantageous. The beta phase of Ta is most often found in sputtered coatings [16], but it is significantly more brittle than the ductile alpha phase. We have found that when the coatings are rubbed or scratched, the columns fold over rather than break and the mechanical properties of the alpha phase may be responsible for this property.

The results of the pore size distribution measurements are shown in Figure 3. We see that there is a broad distribution of sizes in the 5 to 10 nm range along with many smaller pores. These smaller pores could be the ones seen close to the substrate in Figure 1, which are associated with crystallographic orientations having slower growth rates than the dominant bcc oriented columns. Figure 3 is an estimate based on the

assumptions that the pores are cylindrical and 10 microns high, so it should be taken only as an order of magnitude indication of the actual size distribution. Nevertheless, it appears reasonable based on the structures seen in Figures 1 and 2.

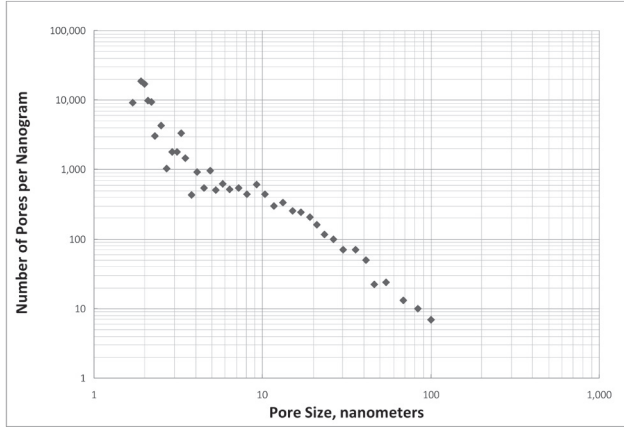


Figure 3: Pore size distribution of the nanostructured tantalum coating.

### In vivo Test Results and Discussion

#### One Week Results

The results of the push-out force measurements after one week of bone growth are shown in Figure 4 for the titanium substrates and in Figure 5 for the PEEK substrates. In the case of titanium, there was a significant increase in push-out force for the drug soaked samples compared with the non-drug soaked samples for all three substrates. We also see that there was an increase in push-out force for the hydroxyapatite coated substrates compared to the uncoated titanium and a corresponding increase for the Ta coated substrates for samples both with and without drug soaking. An increase in push-out force provides strong evidence of significant new bone growth next to the Ta coated substrates [9].

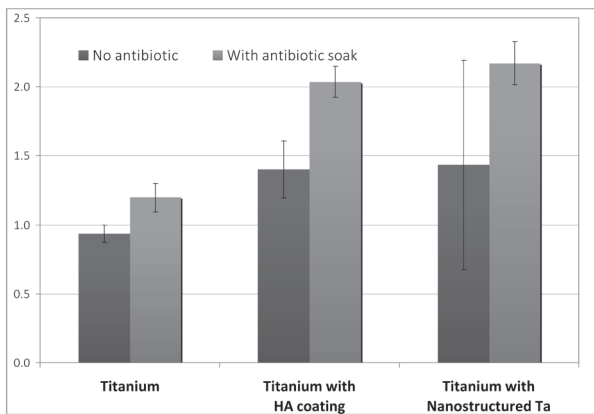


Figure 4: Push out force measurements (MPa), titanium substrate, one week.

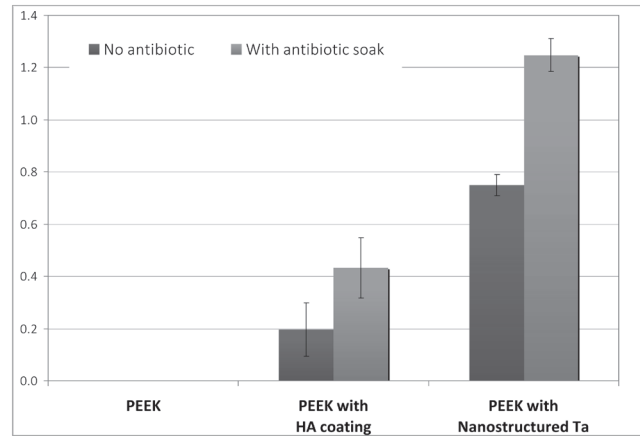


Figure 5: Push out force measurements (MPa), PEEK substrate, one week.

These effects are seen even more dramatically in the case of the PEEK substrates. The push-out force of bare PEEK is negligible. Adding a hydroxyapatite coating produces a push-out force of 0.20 MPa in the case of non-drug soaked substrates and 0.42 MPa in the case of drug soaked substrates. However, the use of a porous Ta coating increases the push-out force significantly to 0.75 MPa for the non-drug soaked substrates and to 1.24 MPa for the drug-soaked substrates.

#### Four Week Results

The four week results are similar to the one week results in relative terms, but the push-out forces increased in magnitude for all of the samples. Figure 6 shows the results for the titanium substrates. The hydroxyapatite coated titanium had approximately twice the push-out force of the uncoated titanium for both the drug soaked and non-drug soaked samples. And after four weeks, there is evidence that the Ta coated titanium showed a slight improvement over the hydroxyapatite coated titanium for both the drug soaked and non-drug soaked samples.

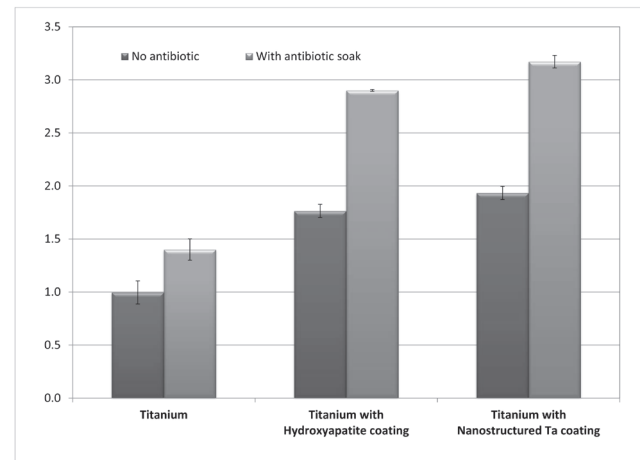


Figure 6: Push out force measurements (MPa), titanium substrate, four weeks.

In the case of the PEEK substrates, the significant improvements continued for all four coating combinations compared to the bare PEEK. Figure 7 shows the four week results for the PEEK substrates. The Ta coated samples continued to show almost a factor of three improvement in push-out force compared to the hydroxyapatite coated samples for both the drug soaked and non-drug soaked substrates. This is significant both statistically and practically. The use of PEEK is growing in orthopedic applications, particularly in spine reconstruction [17], and the ability to improve the extent of osseointegration by a factor of three compared with what is presently available is a critical indicator for the potential clinical success of coating PEEK with nanostructured Ta.

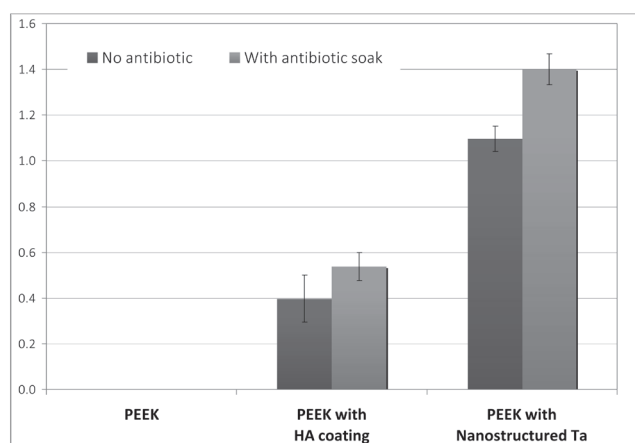


Figure 7: Push out force measurements (MPa), PEEK substrate, four weeks.

## CONCLUSIONS

We have found that nanoporous Ta coatings applied to both titanium and PEEK improves osseointegration in a rat infection model compared to uncoated substrates. The improvement was seen for both drug soaked and non-drug soaked implants that were inoculated with bacteria, indicating that the surface structure may play a critical role in the prevention of biofilm formation (even without using an antibiotic). The improvements in the case of PEEK substrates were particularly significant compared to both bare PEEK and hydroxyapatite coated PEEK substrates. The factor of three improvement in push-out force compared to hydroxyapatite coated PEEK is important given the growing use of PEEK in implanted medical devices.

A remaining question is the locus of failure for the hydroxyapatite and Ta coated materials. It is important to know whether the failure separation during push-out tests occurred at the coating substrate interface or at the coating bone interface and future work will be focused on that question.

## REFERENCES

1. T. J. Webster, R. W. Siegel, and R. Bizios, *Biomaterials* 20:1221-1227 (1999).  
[http://dx.doi.org/10.1016/S0142-9612\(99\)00020-4](http://dx.doi.org/10.1016/S0142-9612(99)00020-4)
2. T. J. Webster, C. Ergun, R. H. Doremus, R.W. Siegel, and R. Bizios, *Journal of Biomedical Materials Research* 51:475-483 (2000).  
[http://dx.doi.org/10.1002/1097-4636\(20000905\)51:3<475::AID-JBM23>3.0.CO;2-9](http://dx.doi.org/10.1002/1097-4636(20000905)51:3<475::AID-JBM23>3.0.CO;2-9)
3. T. J. Webster, R. W. Siegel, and R. Bizios, *Biomaterials* 21:1803-1810 (2000).  
[http://dx.doi.org/10.1016/S0142-9612\(00\)00075-2](http://dx.doi.org/10.1016/S0142-9612(00)00075-2)
4. C. Ergun, R. H. Doremus, R. W. Siegel, and R. Bizios, *Biomaterials* 22: 1327-1333 (2001).  
[http://dx.doi.org/10.1016/S0142-9612\(00\)00285-4](http://dx.doi.org/10.1016/S0142-9612(00)00285-4)
5. H. Liu\* and T.J. Webster, *Biomaterials* 28(2): 354-369 (2007).  
PMid:21898921
6. S. Puckett, E. Taylor, T. Raimondo, and T.J. Webster, *Biomaterials* 31(4): 706-713 (2010).  
<http://dx.doi.org/10.1016/j.biomaterials.2009.09.081>  
PMid:19879645
7. B. Ercan, E. Taylor, E. Alpaslan, and T.J. Webster, *Nanotechnology*, 22(29):957-971 (2011).  
<http://dx.doi.org/10.1088/0957-4484/22/29/295102>  
PMid:21673387
8. E. Taylor and T.J. Webster, *International Journal of Nanomedicine*, 6: 1011-1022 (2011).  
<http://dx.doi.org/10.2217/nnm.11.25> PMid:21707298
9. J. W. Costerton et. al., *Science* (284), 1318-1322, (1999).
10. L. R. Martinez et. al., *J. Invest. Dermatol.* (129), 2463-2469, (2009).
11. D.A. Glocker and M.M. Romach, "Tantalum Radiopaque Coatings for Stents," *51<sup>st</sup> Annual Technical Conference Proceedings of the Society of Vacuum Coaters*, pp. 199, 2008.
12. US Patent No. 8,002,822.
13. J. A. Thornton, *J. Vac. Sci. Technol.* (11), 666, (1974).

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14. S. Lowell, J. Shields, M. Thomas and M. Thommes, Characterization of Porous Solids and Powders, Kluwer Academic Publishers, Netherlands, (2004).  
<http://dx.doi.org/10.1007/978-1-4020-2303-3>
  15. W.J. Dhert, C.P. Klein, I.C. Wolke, E.A. van der Velde, K. de Croot, and P.M. Rozing, *J. Biomedical Materials Research*, 25(10), 1183, (1991).  
<http://dx.doi.org/10.1002/jbm.820251002> PMID:1667400
  16. A. A. Navit and A. M. Hodge, *Scripta Materiala* 63, 867-870, (2010).  
<http://dx.doi.org/10.1016/j.scriptamat.2010.06.037>
  17. "Use of plastic implants is soaring," *Plastics Today*, May 20, (2013).