Ziebart T, Pabst A, Klein MO, Kämmerer P, Gauss L, Brüllmann D, Al-Nawas B, Walter C. Bisphosphonates: restrictions for vasculogenesis and angiogenesis: inhibition of cell function of endothelial progenitor cells and mature endothelial cells in vitro. Clin Oral Invest. DOI: 10.1007/s00784-009-0365-2.

Bisphosphonate treatment is an effective and beneficial treatment for malignant bone cancers and their metastases in addition to severe osteoporosis and Paget's disease. Unfortunately, there are several unspecific side effects produced by bisphosphonates that limit their clinical use, including osteonecrosis of the jaws. Using an in vitro model, Thomas Ziebart et al.'s 2009 publication evaluated the mechanism of action of bisphosphonase-associated osteonecrosis of the jaws (BP-ONJ). Human umbilicord vein epithelial cells (HUVEC) and endothelial progenitor cells were used to evaluate a multi-concentration exposure of four potent bisphosphonates on cellular migration, apoptosis and viability. The least potent nitrogen-containing compound ibandronate produced a minimal degree of toxicity, leading the authors to conclude that higher concentrations should have been evaluated. The remaining nitrogen-containing bisphosphonates (pamidronate, and zoledronate) produced drastic and concentration-dependent effects on cell viability and apoptosis relative to the non-nitrogen containing bisphosphonate (clondronate). Cellular migration, as observed through a Boyden migration assay and 3-dimensional angiogenesis assay, was also severely inhibited by pamidronate and zoledronate treatment. Accordingly, these results demonstrate that the nitrogen-containing bisphosphonates pamidronate and zoledronate caused significant angiogenesis and vasculogenesis inhibition, which may be responsible to the development and maintenance of BP-ONJ. The authors suggest that future studies evaluate these results using in vivo studies. The results are pertinent to implant dentistry community as they demonstrate the cellular mechanisms of toxicity (migration and apoptosis) that contribute to the pathophysiology of BP-ONJ observed clinically. Furthermore, they demonstrate a elevated level of toxicity with the nitrogen-containing bisphosphonates pamidronate and zoledronate, whose clinical use should be carefully monitored given the breath of negative side effects they produce.

Nicholas Radio, PhD, Molecular Pharmacologist

Wall I, Donos N, Carlqvist K, Jones F, Brett P. Modified titanium surfaces promote accelerated osteogenic differentiation of mesenchymal stromal cells in vitro. Bone. 2009;45:17–26.

This study by Wall and colleagues evaluated the optimal titanium surface properties and hydrophilicity to facilitate bone regeneration. Human bone marrow derived mesenchymal stem cells (hMSCs) were used to characterize the osteogenesis of two modified titanium surfaces: a rough, hydrophobic titanium surface that was sand-blasted and acid-etched (SLA) and an acid-etched titanium surface that contained the same roughness but modified to have greater hydrophilicity (SLActive). No statistical differences were observed for hMSC cell attachment to the titanium surface for either the SLA or SLActive material when compared to a smooth polished titanium surface (SMO) used as a negative control. Human mesenchymal stem cell proliferation was reduced in both SLA and SLActive groups relative to SMO. This is significant as cellular proliferation generally decreases during the differentiation process as the cells become post-

mitotic (M-phase). The greater hydrophilicity of the SLActive surface did not appear to contribute to this effect, as there was no proliferation differences observed between either group. Furthermore, hMSCs cultured on either rough titanium surface (SLA or SLActive) caused significantly enhanced and accelerated osteoblast differentiation relative to SMO, indicated by increased levels of the osteogenic markers SPP1, RUNX2 and BSP as well earlier calcium deposition, observed as early as 1 week post-surface contact. The authors speculated that this acceleration was due to WNt5A signaling and beta-catenin-dependent mechanisms. Comparison between SLA and SLActive titanium surfaces indicated a slight but statistically significant increase in the osteogenic promoter WNT5A response in SLActive surfaces. These results are relevant to the implant dentistry community as they specify the optimal surface properties for titanium implants to accelerate osteogenesis and bone-implant contact to facilitate bonding strength.

Nicholas Radio, PhD, Molecular Pharmacologist

Puckett SD, Taylor E, Raimondo T, Webster TJ. The relationship between the nanostructure of titanium surfaces and bacterial attachment. J Biomat. 2010; 31:706–713.

There are a great number of reasons for non-integration of a dental implant, of which one being post implantation bacterial infection. Several steps have being taken to mitigate such an undesirable event including the use of systemic antibiotics and creating implant surfaces that promote accelerated protein adsorption and subsequent osteoblast functions faster than bacterial adhesion to the implant surface. The reduction in the adhesion of a broad range of bacteria could be an attractive means to decrease infection and allow for subsequent appropriate tissue integration with the implant surface. In this in vitro study, nanometer sized (70–240 nm) topographical features of titanium (Ti) surfaces which have been shown to promote accelerated osteoblast formation were examined as a means to also reduce bacterial adhesion. This study examined the adhesion of *Staphylococcus aureus*, *Staphylococcus epidermidis* and *Pseudomonas aeruginosa* on conventional Ti, nanorough Ti produced by electron beam evaporation, nanotubular and nanotextured Ti produced by two different anodization processes.

The study found that compared to conventional (nano-smooth) Ti, the nanorough Ti surface had the least amount of bacterial attachment of all of the studied nanosurfaces. The study also found that by altering the surface roughness from a micrometered textured surface to a nanometered textured surface, certain cellular functions like osteoblastic activities were enhanced while simultaneously decreasing competitive cells like fibroblast function. By decreasing particle size, the study was able to show decreased bacterial adhesion across all of the nano surfaces compared to the micron surface, but some nano surfaces favored better than others. Clearly this study needs to be duplicated in vivo, but if the initial results are any indication, the implant companies are moving in the right direction with nano textured surfaces.

Pankaj P. Singh, DDS, AAID Fellow, ABOI Diplomate

Yao C, Webster TJ. Prolonged antibiotic delivery from anodized nanotubular titanium using a co-precipitation drug loading method. J Biomed Mater Res B Appl Biomater. 2009;91B:587–595.

Orthopedic implant failure can be due to both mechanical and biological processes; moreover, infection, excessive immune responses, and a lack of initial bone growth

can lead to implant loosening and failure. One way to improve survival rates is to improve the material's mechanical and/or biological properties to be more compatible. Advances in nanotechnologies have led to the development of numerous techniques that can improve the cytocompatibility properties of existing titanium based implant systems including mechanical roughening, chemical etching, and coating with bioactive materials to name a few. An electrochemical process called anodization, or anodic oxidation, has been used to modify the surface morphology and chemistry of titanium implants, resulting in a layer containing ordered titania nanotubules which mimic the size and pattern of components of natural bone. Besides modifying implant material properties to promote bone cell functions, anodized titanium possessing nanotubular structures can be an ideal drug storage matrix to locally deliver a wide variety of chemotherapeutics after implant surgery.

This in-vitro study of a currently used titanium surface was anodized to possess nanotubular surface structures (80 nm in inner diameter and 200 nm deep) capable of drug delivery. These nanotubular surfaces were loaded with penicillin based antibiotics using a co-precipitation method in which drug molecules were mixed in simulated body fluid to collectively precipitate with calcium phosphate crystals. Results showed that this surface could release the drug for up to 3 weeks compared to only a 150 minute period thru simple physical adsorption. In addition, contrary to conventional thinking that a penicillin based drug release should decrease cell functions, this study showed similar osteoblast adhesion between non-drug loaded and drug loaded precipitated calcium phosphate coatings on anodized titanium. These initial results are promising for demonstrating improved bone cell adhesion while delivering not only penicillin based chemotherapeutics but other antibiotics and anti-inflammatory drugs, in the hopes of mitigating the more common local biologic responses (nonmechanical, load, stress related) to implantation that cause implant failure; while simultaneously improving implant functionality by simply creating a co-precipitated anodized nanotubular titanium surface. We in dentistry understand the physiologic benefits of local chemotherapeutic delivery systems vs. systemic delivery systems to treat a local pathophysiologic response, so these are promising findings, as the untoward reactions to dental implant surgery are usually local to the implant site at the onset.

Pankaj P. Singh, DDS, AAID Fellow, ABOI Diplomate