Vancomycin release from point-of-care phosphatidylcholine implant coatings

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Statement of Purpose:
Musculoskeletal trauma often requires implanted biomaterials for fixation or joint replacement, which are susceptible to biofilm formation [1]. Biofilm-forming microorganisms tend to adhere to implanted surfaces and medical devices, which can lead to persistent infections [2]. Biofilm-based infections are resilient to current antibiotic therapy methods, host immune defenses, and have higher likelihood of developing antibiotic resistance. While prophylactic systemic administration of antibiotics is commonly used to prevent biofilm formation, especially in patients with suspected colonization of pathogenic microorganisms such as methicillin-resistant *Staphylococcus aureus* (MRSA), infection still occurs at rate as high as 5% for some implanted orthopedic fixation devices. Orthopedic implant infections can delay recovery time 6 weeks or longer and increase costs for patients and the health care system. Implant failure caused by infection can result in the patient requiring extensive multi-stage revision procedure.
Anti-infective biomaterials have become a primary strategy to prevent implanted biomaterial infections and to inhibit biofilm-forming microorganisms [3]. A novel coating material has been developed that can be loaded with antibiotics and applied directly to an implant surface to release high concentrations of antimicrobial locally. In this study, the elution profile of the antibiotic vancomycin was characterized.
Methods: Antibiotic-loaded coatings were made by combining purified phosphatidylcholine (Phospholipon 90G, Lipoid Gmb, Germany) with 25% vancomycin powder. Stainless steel coupons 15 mm in diameter and Titanium coupons 19 mm in diameter were sanded to a uniform finish using a series of sandpaper grit, and then washed thoroughly and sonicated to remove contaminants. Coupons were then autoclaved at 121°C for 20 minutes for sterilization. Coatings were applied by direct coating until all surfaces of the coupons were visibly coated.
Coupons were placed in 2 ml PBS and samples were taken each day for 7 days, with complete media refreshment each day (n=4). Vancomycin concentration was determined using High Pressure Liquid Chromatography (HPLC). Vancomycin peaks at 1.5 minutes were characterized at 241 nm with a C18 reverse phase column using mobile phase of 35% acetonitrile and 65% buffer (0.08 M disodium phosphate and 0.013 M monosodium phosphate adjusted to pH 3 with phosphoric acid).
Results: In both stainless steel and titanium samples, elution followed a pattern of extended release until day 5, when a burst effect of increased elution was observed (Figure 1). All samples measured were above the reported minimum inhibitory concentration (MIC) for vancomycin against *S. aureus* of 1 μg/ml. There was a large variation between sample concentrations, however. This may have been due to varying amounts of coating applied or variation in coating degradation. Visual observation of samples indicated that coating was still visible on titanium samples throughout the 7 day period, while coating on stainless was less apparent after day 5.

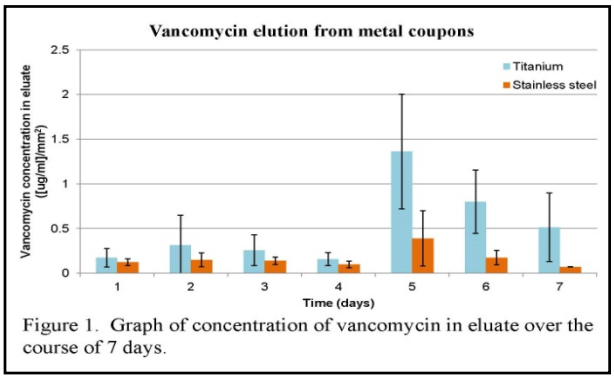

Figure 1. Graph of concentration of vancomycin in eluate over the course of 7 days.

Conclusions: Phosphatidylcholine coatings release vancomycin from metal surfaces at concentrations that are inhibitory to bacteria. The pattern of release can be used to modify the concentration of antibiotic loaded to remain above the MIC without reaching levels toxic to tissue. Differences between elution patterns between titanium and stainless steel may be due to different oxide layers, microstructural differences, or other material properties. The mechanisms for these effects may be examined in future studies, as well as elution from polymeric and ceramic materials. Preliminary studies have confirmed that vancomycin released is active and inhibits biofilm formation *in vitro* and *in vivo* [4]. Studies are under way to characterize the release of other antibiotics and antimicrobials, such as amikacin, from phosphatidylcholine matrices. The combination of antibiotics to achieve broad spectrum coverage against microorganisms is also being evaluated. Future studies are planned to evaluate coatings in the context of contaminated orthopaedic defect models.

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