Antibiotic Loaded Tissue Sealant for the Prevention of Postoperative Infection in Orthopedic Procedures

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Introduction: Bacterial infection resulting from orthopedic implant procedures is a significant clinical problem affecting between 0.5 to 20% of patients [1]. Often these infections are managed by prophylactic and post-surgical treatment with systemic antibiotics, albeit with limited success. Localized delivery of antibiotics shows promise in reducing the rate of post surgical infections by providing high drug concentrations over short periods of time directly at the site of action. In recent years there has been active research in the local delivery of antibiotics for the treatment of osteomyelitis using naturally occurring polymeric implants such as fibrin [2], collagen [3], gelatin [4] and chitosan [5], to achieve prolonged drug exposure over 4 to 6 weeks. However, in the case of post-surgical prophylaxis, rapid delivery over 3 to 10 days is desirable, therefore, new delivery systems are required. Fibrin tissue sealants may be an ideal delivery system for this application, as these materials are capable of maintaining intimate contact with the surgical site for several days. The purpose of this study was to investigate the use of the tissue sealant, CoStasis™, as an antibiotic delivery system for the treatment of orthopedic post-operative infection. We characterized the in vitro release of three antibiotics from CoStasis™: cefazolin, 5-fluorouracil and fusidic acid, and determined the in vivo efficacy of these systems in an orthopedic surgical infection model.

Materials and Methods: The incorporation of either Cefazolin (Cef), 5-fluorouracil (5FU) or fusidic acid (FA) into the tissue sealant was achieved by dissolving appropriate amounts of the drugs in rat plasma. The plasma was mixed with CoStasis™ using the dual syringe delivery system provided. In vitro drug release from 100mg CoStasis™ matrices containing a 5% w/w drug loading was determined in 2mL of pH 7.4 phosphate buffered saline (PBS) at 37°C. At 4, 8, 24, 48 and 98 hrs, the release media was removed for analysis by HPLC and replaced with fresh buffer. Analysis of all drugs was achieved using validated HPLC methods.

The minimum inhibitory concentration (MIC) of the antibiotics was determined using the micro-broth dilution technique. Briefly, serial dilutions (1024, 512, 256, 128, 64, 32, 16, 8, 4, 2, 1, 0.5 and 0.25 μg/mL) of antibiotics were prepared in Mueller-Hinton broth in 96-well microtitre plates. A bacterial suspension of S. aureus Newman was then added to each well in triplicate to achieve a final inoculum concentration of 5×10^8 bacteria/mL. Controls consisted of wells containing growth media alone or bacterial culture without antibiotic. The 96-well microtitre plates were inspected for bacterial growth after incubation at 37 °C for 24 h. The minimum bactericidal concentration (MBC) was defined as the lowest concentration of drug that resulted in more than 99.9% reduction of the initial inoculum.

The in vivo efficacy of the drug loaded tissues sealant was assessed in 300g male rats according to UBC Animal Care Committee ethics. Briefly, a surgical dissection down to an upper lumbar spinous process was performed and a titanium ligating clip was inserted. Into the wound a 100μL suspension of S. aureus Newman in PBS at a concentration of 10^5 CFU was introduced along with 100mg of CoStasis™ containing a 5% w/w loading of Cef, 5FU or FA. Control animals received either no treatment or drug dissolved in plasma. All groups were repeated in duplicate.

After 14 days the animals were euthanised by CO₂ and the bacterial burden evaluated by tissue and wound swabs plated on tryptic soy agar (TSA) plates and incubated overnight at 37°C. Post mortem dissection of control animals without drug treatment revealed a well-encapsulated abscess surrounding the titanium clip with erosion of the boney process. Tissue and wound swabs produced cultures with high bacterial growth and a clip culture count of >10,000 CFU. Animals treated with drugs in plasma generally had tissue and wound cultures with high bacterial growth and clip cultures above 10,000 CFU. Delivery of antibiotics in CoStasis™ dramatically reduced the amount of bacteria cultured from the tissue and wound swabs with culture plates having little or no growth. For all animals tested with drugs in CoStasis™, the cultures from the clips were significantly reduced compared to control animals with maximum cultures of 500 CFU, 400 CFU and 300 CFU for 5FU, Cef and FA treated animals, respectively.

Discussion: For the prevention or treatment of postoperative infections using localized delivery systems of antibiotics, it is desirable to have rapid release of drug for up to 10 days. The in vitro drug release studies showed that release from the tissue sealant, CoStasis™, provided rapid, yet controlled delivery of Cef, 5FU or FA lasting 2 days suitable for the intended purpose. The spinal orthopedic procedure on rats provided an excellent animal model for investigating orthopedic postoperative infection, showing 100% infection levels with 0% mortality. Treatment with drug dissolved in plasma injected at the site showed little to no efficacy, presumably due to rapid clearance of the drug from the site of action. The dramatic decrease in the bacterial load at the infection site and on the clip observed in rats treated with the antibiotic containing tissue sealant system provided evidence of the ability of the tissue sealant to retain the drug at the infection site, prolong the drug release, and provide high local drug concentrations for several days. Although infections were not completely eradicated, these studies support the use of antibiotic loaded tissue sealants as drug delivery systems for prophylaxis of postoperative infection in orthopedic procedures.