Application of Electron Microscopy in Advanced Musculoskeletal Infection Research

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Statement of Purpose: Infections like COVID-19 have changed our lives and have reminded us of the challenges in dealing with all kinds of infections. The invention and use of antibiotics have been credited for the significant reduction of infections and saving millions of lives. Unfortunately, the wide use of antibiotics has also created challenges related to antibiotic resistant bacteria, which have been increasingly seen in recent decades. It is estimated that between 38.7-50.9% of microorganisms causing surgical site infections are resistant to standard prophylactic antibiotics.[1] In the U.S. alone, antibiotic resistant infections increased by 359% between 1997 and 2006,[2] and antibiotic resistant bacteria cause at least 2.8 million infections, 35,000 deaths a year,[3] and $55 - 70 billion per year in economic impact.[4] Besides antibiotic resistance, musculoskeletal infection treatment faces other challenges including biofilm formation, intra-cellular infection, delayed wound healing, *etc.* The *purpose of this study* was to apply electron microscopy to manage the challenges we are facing with musculoskeletal infections and to examine musculoskeletal infections both *in vitro* and *in vivo*.

Methods: Electron microscopy, especially scanning electron microscopy (SEM), was used to study bone musculoskeletal infections and related therapeutic treatments. SEM and transmission electron microscopy (TEM) were used to observe biofilm formation both *in vitro* and *in vivo*, to visualize biofilm disruption, to examine antimicrobial mechanisms, and to guide the development of innovative antimicrobial biomaterials and therapeutic treatments.

Results and Discussion: The use of electron microscopy to manage the challenges we are facing with musculoskeletal infections was investigated. SEM was used to examine biofilm formation and the disruption of the biofilms using a variety of antimicrobial agents. It was found that cationic antimicrobial peptides like LL-37 lysed the bacteria and were relatively more effective in eliminating biofilms (Fig. 1). Biofilm formation in an open fracture rat model was also detected using SEM (Fig. 2). Moreover, SEM was useful in the development of antimicrobial biomaterials for infection prevention and treatment. Electron microscopy was used to characterize antimicrobial nanoparticles and surface structures, coating stability, and drug delivery vehicle structures, and to determine nanoparticle and bacteria internalization, as well as cell and bacteria attachment and morphology.

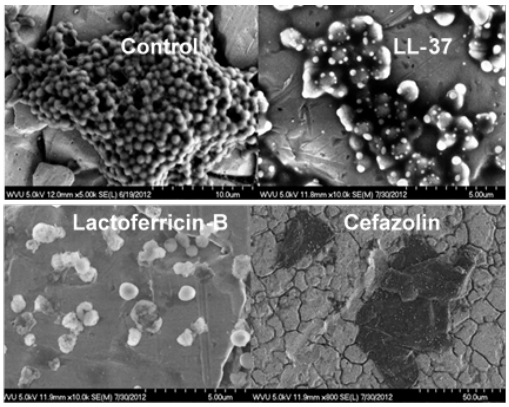

Figure 1. Biofilm formation *in vitro* and effects of antimicrobial treatments.

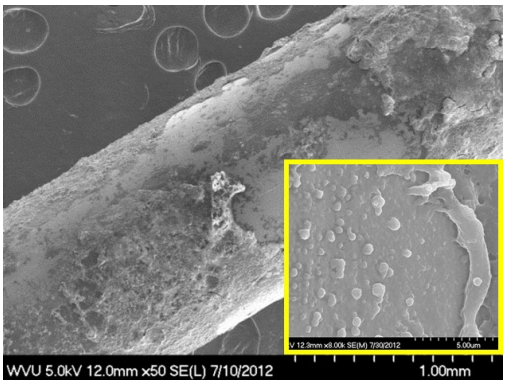

Figure 2. Observation of *Staphylococcus aureus* biofilm formation on an implant used to fix an open fracture in a rat.

Conclusions: Electron microscopy is a useful tool in studying musculoskeletal infections and in develoing innovative antimicrobial biomaterials for preventing and treating musculoskeletal infections.

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