Antibacterial microfilm dressing with silver-nanoparticles promotes healing of contaminated excisional wounds

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Statement of Purpose: Topical antimicrobial ointments and solutions have only few hours of residence time in wounds, require multiple applications with frequent painful dressing changes, and result in toxic build up of antimicrobials such as silver, iodine, bisguanides or chlorhexidine[1]. While expensive advanced dressings loaded with antimicrobials reduce dressing changes, they also result in deposition of toxic levels of antimicrobials in wound-bed[2]. Moreover, when used with biologic dressings, these current standards of care still result in up to 20% infection rate[3]. We have developed a transparent and dissolvable antibacterial *microfilm dressing* for use with primary wound dressings. This *microfilm dressing* is physically robust for handling by clinicians, but dissolves quickly in moist wound to immobilize on wound-bed a polymer nanofilm impregnated with silver-nanoparticles that can subsequently be covered with a primary or secondary wound dressing. Nanofilms provide long-term release of ~100 times less silver/day than currently available silver dressings, thus reducing silver-toxicity concerns and painful dressing changes.

Methods: The microfilm dressing has two components: a water-soluble layer (~100 µm) of polyvinylalcohol (PVA, Mw- 22kda), and a polyelectrolyte multilayer (PEM) nanofilm (~200 nm) composed of oppositively charged polyelectrolytes- poly(allylamine hydrochloride) (PAH) and poly(acrylic acid) (PAA). Nanofilms assembled on elastomeric-sheets via layer-by-layer spray coating were impregnated with silver ions that were subsequently reduced to silver nanoparticles in-situ[3]. The nanofilms were peeled from the substrate along with a PVA cast deposited over them to form the freestanding *microfilm dressing*. Silver loading was characterized by inductively coupled plasma emission spectroscopy[3], elastic modulus by Dynamic Mechanical Analysis (DMA), and thickness by a Profilometer. *In vivo*, two splinted excisional wounds (6 mm) were created surgically in Balb/c mice and inoculated with ~10^5 CFU *Staphylococcus aureus*. One test group (n=10 mice/group) was treated with the silver-*microfilm dressing*, and the other did not receive microfilms. All wounds were ultimately covered with a collagen dressing (Biobrane®, UDL Laboratories).

Results: A *microfilm dressing* with storage modulus (E') of 1x10^9 Pa containing 11±2.3 µg/cm^2 silver released <1 µg/cm^2 of silver/day in water for 10 days. This was sufficient to kill 4 $\log_{10}$ CFU of *S. aureus*, *P. aeruginosa* and *MRSA* in solution within 24 h. Murine wounds treated only with Biobrane had an average of 3.0x10^5 CFU of *S. aureus* and 47.1%±7.0% (mean ± SEM) of original wound size, at 9 days post-surgery (**Fig. 1**). In contrast, wounds treated with silver-*microfilm dressing* and Biobrane had significantly fewer bacteria (2.8x10^3 CFU) and a smaller wound size (13.3%±3.1%) (p<0.05). Histopathology of wounds treated with silver-*microfilm dressing* showed significantly smaller epithelial gap, more collagen, and less inflammation compared to the controls (p<0.05). Nanofilms ultimately disintegrated into microfragments removed during reepithelialization.

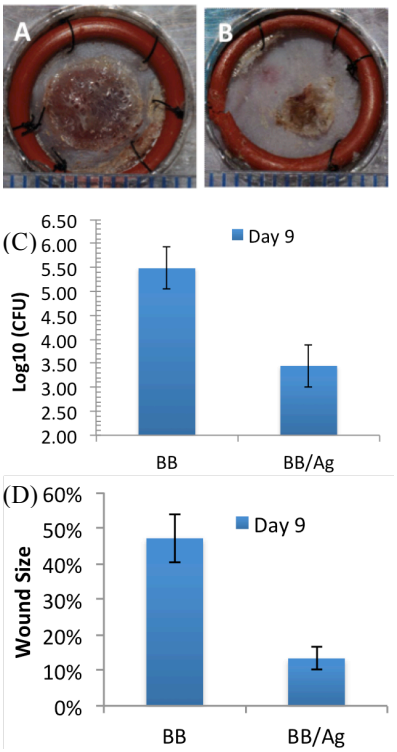

Fig.1. *Microfilm dressing* with 11±2.3 µg/cm^2 silver, immobilized on wounds under Biobrane, reduced colonization of *S. aureus* and expedited wound closure at day 9 post-surgery: (A,B) Gross images of splinted wounds with Biobrane (BB), or silver-microfilm/Biobrane (BB/Ag), respectively. (C) Bacterial burden in wounds. (D) Mean wound size. (Average ± SEM, n=10, p<0.05).

Conclusion: *Microfilm dressing* with silver-nanoparticles present a unique substitute to topical antimicrobial formulations or silver-loaded wound-dressings, as it effectively reduces wound bioburden under primary dressings by providing long-term release of less than 1 µg/cm^2 of silver/day and expedites wound closure. Tests in porcine dermal wounds are underway. **Funding** provided by NIH SBIR grant# 1R43AR061913-01A1.

References: (1) Sheridan, et al, J Burn Care Rehab. 1997;18:139. (2) Trop et al, J Trauma. 2006;60:648. (3) Shores et al, Adv Skin Wound Care. 2007;20:493. (4) Agarwal et al, Biomaterials, 2010;31:680.