Non-Thermal Plasma Treatment Safely and Rapidly Eradicates MRSA from Infected Wounds

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INTRODUCTION: *Steri-lysis*™ technology is a novel, non-thermal plasma (NTP)/free radical, portable device that delivers a highly active reactive oxygen and nitrogen species (RONS) mixture within a closed loop system. Previously, we demonstrated that *Steri-lysis*™ technology rapidly and reliably sterilized cell phones, which are well known to harbor microorganisms, including pathogenic *Staphylococcus aureus.* Given the device's remarkable disinfection efficacy, we then investigated its potential applicability to living tissue, and established that NTP is safe for use on wounded and unwounded skin. Herein, we investigate the effectiveness of NTP treatment of infected wounds in an *in vivo* murine model.

MATERIALS AND METHODS: Two 6 mm, full-thickness, splinted excisional wounds were created on both sides of the dorsal midline of C57bl/6 mice, and inoculated with 25 microliters of 1.5×10^8 colony forming units (CFU)/mL methicillin-resistant *Staphylococcus aureus* (MRSA) per wound. Mice were treated with NTP for 10 minutes or 20 minutes, respectively, for a total of 3 treatments every other day for 7 days. Wounds were aseptically swabbed and plated onto trypticase soy agar pre- and post-NTP treatment to monitor interim bacterial reduction during the study period. On day 7, wound tissue was harvested and homogenized for quantitative analysis of CFU/gram tissue remaining.

RESULTS: After NTP treatment, there was no gross or histological evidence of residue, aberrant dermal architecture, or edema in any of the wounds or intact skin. Interim agar growth from pre- and post-NTP aseptic swabbing revealed one logarithmic reduction in bacterial load for the 20-minute group, which was subsequently confirmed with serial dilutions of full-thickness, homogenized wound tissue. Wounds inoculated with MRSA and treated with NTP for 20 minutes had a 90% reduction in MRSA CFUs/gram of homogenized tissue ($p = 0.0441$) compared to untreated wounds. Efficacy of 20 minutes NTP treatment was confirmed by lack of anti-staph ab staining on sectioned wounds.

CONCLUSION: Our novel non thermal plasma device effectively disinfects clinically infected wounds in an *in vivo* murine wound healing model, without any evidence of damage to the wound or surrounding tissues. Effective against pathogenic MRSA, *Steri-lysis*™ is an innovative, low cost, portable technology that could potentially revolutionize device sterilization in hospital and health care settings as well as shift the paradigm of the treatment of colonized and/or infected wounds.