oral and intravenous antibiotics; however, this treatment is ineffective in eradicating bacterial biofilms, leads to antibiotic resistance, and can impair physiologic wound repair mechanisms. Our lab has developed a novel human collagen hydrogel (cHG) embedded with antibiotic (cHG-abx) for topical treatment of infected chronic wounds that is able to mitigate the risks of current treatment while providing physiologic ECM proteins needed for wound repair. We hypothesize that topical administration of our novel cHG-abx will effectively inhibit growth of multiple clinically important and common bacteria while maintaining mammalian cell viability.

METHOD: C. perfringens and MRSA were treated with 100X minimum inhibitory concentration (MIC) of clindamycin (100 μg/ml) and gentamycin (500 μg/ml), respectively. Human collagen hydrogel preparation: 2.5% cHG was fabricated from a previously established protocol. Prepared cHG was mixed with antibiotic and incubated to induce gelation. Modified Kirby-Bauer: cHG-abx was gelled onto polycarbonate films and allowed to elute in PBS for various timepoints before being placed onto inoculated agar. After 12 hours of treatment, the zone of inhibition was measured. Crystal violet assay: Various eluted cHG-abx were added to bacterial suspensions, incubated, then stained with crystal violet solution. Absorbance was measured at 595 nm and compared to a non-treated well. Mammalian cell cytotoxicity: Wells seeded with human and mouse fibroblasts and ADSCs were treated with cHG-abx for 24, 48, and 72 hours before quantifying viability.

RESULTS: No significant mammalian cell death was seen at any time point. Both Kirby-Bauer and crystal violet assays demonstrated significant bacterial inhibition for 48 hours compared to no treatment for C. Perfringens and MRSA. Furthermore, significant differences in bacterial elimination over elution time points indicate sustained release of antibiotics.

CONCLUSION: Human collagen hydrogel embedded with antibiotics is capable of sustained low-dose antibiotic release to successfully inhibit growth of various clinically relevant bacterial strains in vitro. Furthermore, the gel shows promise for in vivo application, as no significant mammalian cell death was found.

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**TRACK: CRANIOMAXILLOFACIAL/HEAD AND NECK**

A Single-institution, Retrospective Chart Review of Outcomes and Complications Following Infant Ear Molding with the EarWell System

**Presenter: Jamasb Sayadi**

**Co-Authors: Arhana Chattopadhyay, MD, Jagmeet Arora, Rohit Khosla, MD**

**Affiliation: Division of Plastic and Reconstructive Surgery, Stanford University, Stanford, CA**

**PURPOSE:** Approximately one-third of infants are born with ear anomalies, more than two-thirds of which do not self-correct.1 Ear molding is the mainstay of nonsurgical correction and is commonly initiated as soon as possible after birth while cartilage remains malleable due to high concentrations of circulating maternal estrogens in the infant bloodstream.1 The purpose of this study was to examine the outcomes and complications associated with infant ear molding using the EarWell system at a single institution.

**METHOD:** We conducted a retrospective chart review of all infants who underwent ear molding with pediatric plastic surgery from October 2010 to March 2021. Ear anomalies were classified as deformations, malformations, or multiple anomalies. Age at initiation, duration of treatment, temporal gaps in treatment, comorbidities, and details regarding any complications were also extracted for included patients. The primary outcomes assessed were degree of ear anomaly correction, incidence of skin complications, and unanticipated cessation of treatment. Parents of included patients were also sent a questionnaire regarding their children’s experiences with the ear molding process in which four outcomes regarding ear appearance (overall appearance, ‘natural’ look, symmetry, prominence) were rated on a 4-point Likert scale and cumulatively scored out of 16.

**RESULTS:** A total of 184 ears of 113 patients were treated during the 11-year study period. Mean age at treatment initiation was 21 days, and average duration of treatment was 40 days. Helical rim deformities (N=50 ears) and lop ear (N=40 ears) were the most common anomalies present. Nine ears possessed characteristics consistent with two different anomalies. A total of 181 ears (98.4%) achieved either a complete (N=125 ears, 67.9%) or partial correction...
(N= 56 ears, 30.4%) upon treatment completion. There was no statistically significant association between age at initiation (p = 0.314), duration of application (p = 0.198), or type of anomaly (p = 0.192) and partial vs. complete correction. The most common complications were eczematous flares (N=27 occurrences among 25 ears, 13.6%) and pressure ulcers (N=23 occurrences among 21 ears, 12.5%). Incidence of complications was not significantly associated with age at application (p = 0.269), duration of application (p = 0.238), or type of anomaly (p = 0.106). Infants who experienced a complication were 3.36 times more likely to achieve partial correction (p < 0.001; 95% CI 1.66-6.81) relative to complete correction. Questionnaire responses were received for 24 out of 113 patients (21.2%) and were categorized into ‘Successful’ (N=21, 87.5%) and ‘Unsuccessful’ groups (N=3, 12.5%) depending on whether respondents denoted the ear molding process as successful or unsuccessful, respectively. The average cumulative appearance score for the ‘Unsuccessful’ and ‘Successful’ groups was 11 ± 2.6 and 15.4 ± 1.6, respectively, with a statistically significant difference between groups (p = 0.002).

CONCLUSION: The EarWell system is an effective treatment strategy for infant ear anomalies, with most patients achieving complete correction. Addressing complications early may help providers optimize outcomes.

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TRACK: RESEARCH/TECHNOLOGY PAPER

Nitric Oxide Releasing Gel Increases Expression of Fibronectin, TGF-β1, and Accelerates Wound Healing in Diabetic Mice

Presenter: Dharshan Sivaraj

Co-Authors: Chikage Noishiki, Kellen Chen, PhD, Dominic Henn, MD, Melissa C. Leeolou, Artem Trotskyuk, Hudson Kussie, David Perrault, MD, Jagannath Padmanabhan, MD, Geoffrey C. Gurtner, MD

Affiliation: Stanford University, Stanford, California

PURPOSE: According to the American Diabetes Association (ADA), over 9-12 million patients suffer from chronic ulceration each year which costs the healthcare system over 25 billion annually. There is a significant unmet need for new and efficacious therapies to accelerate closure of non-healing wounds. Nitric Oxide (NO) plays an important role as a messenger molecule during wound repair. NO levels typically increase rapidly after skin injury in the inflammatory phase and gradually diminish as wound healing progresses. The molecular mechanisms of how increased NO concentration affects wound healing and leads to re-epithelialization and wound closure remains incompletely understood. In this study, we sought to investigate the effect of local administration of an NO-releasing gel on excisional wound healing in diabetic mice.

METHOD: We utilized 15-week-old C57BL/6 mice with leptin receptor deficiency in a splinted excisional wound model to mimic human wound healing through deposition of new granulation tissue and re-epithelialization rather than contracture. Excisional wounds of each mouse received either NO-gel or control phosphate buffered saline (PBS)-gel treatment twice daily until full wound closure (N=5 mice per group). The wound areas were quantified and expressed as a percentage of the original wound area. To assess angiogenic signaling and vasculature in the healed wounds, sections were stained for fibronectin and TGF-β1.

RESULTS: Topical administration of NO releasing gel significantly accelerated the rate of wound healing as compared with PBS treated mice. The mean time for complete wound healing was 14.0 ± 0.75 days in the NO gel treated group compared to 16.0 ± 0.75 days in the PBS-treated group (*p < 0.05). At day two and day seven of healing, as well as after wound closure, we found that immunofluorescent staining for fibronectin and TGF-β1 was significantly increased in the NO treatment group compared to the control group.

CONCLUSION: We have shown that application of NO releasing gel rapidly upregulates fibronectin and TGF-β1 to accelerate closure, facilitate improved ECM reconstruction, and promote tissue regeneration in chronic impaired wounds. The results of this work may have important clinical implications for the management of patients with non-healing wounds.