

pathology exercises for dysphagia. The long-term goal is to increase patient outcomes through song-based programs that are accessible, enjoyable, and personalizable. **METHODS/STUDY POPULATION:** We will pilot the use of combined electroencephalography (EEG) and electromyography (EMG) technologies to analyze both central and peripheral contributors to laryngeal control in a cohort of healthy individuals. This approach provides detailed insight into the coordination between neural and muscular activity, which will serve as a baseline for future studies in clinical populations. Song-based vocal exercises will be compared with standard dysphagia exercises prescribed by speech-language pathologists to assess their mechanistic differences. **RESULTS/ANTICIPATED RESULTS:** We anticipate identifying specific song-based tasks, such as variations in pitch, rhythm, and intensity, which differentially impact laryngeal musculature. Additionally, we will localize neural activation hotspots using EEG during these tasks, providing a more comprehensive understanding of how song-based therapy influences both peripheral and central mechanisms. **DISCUSSION/SIGNIFICANCE OF IMPACT:** This project will lay the groundwork for developing evidence-based song-based therapies for dysphagia, providing an alternative to traditional SLP exercises. By creating an engaging therapeutic program, we aim to reduce dysphagia's healthcare burden, including aspiration events, healthcare costs, and related mortality.

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#### **Eight weeks of creatine monohydrate supplementation is feasible and associated with increased brain creatine in patients with AD**

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**OBJECTIVES/GOALS:** The creatine (Cr) system is impaired in Alzheimer's disease (AD). Data show that creatine monohydrate (CrM) supplementation may improve AD symptoms in AD mouse models, but no human studies have been reported. Thus, we investigated whether an eight-week CrM supplementation was feasible and associated with increased brain creatine in patients with AD. **METHODS/STUDY POPULATION:** Twenty participants with probable AD were allocated to an open-label, eight-week intervention of 20 g/day CrM. Fasting blood draws were taken at baseline, 4-, and 8-week visits to measure serum creatine (Quest Diagnostics). 1H magnetic resonance spectroscopy was performed at baseline and 8-week visits to measure brain Cr as a ratio to unsuppressed water. Self-reported compliance (with assistance from study partners) was assessed with daily CrM trackers. The mean compliance percentage across all participants was used to describe overall compliance with the intervention. We used paired t-tests to analyze the mean changes in serum Cr levels from baseline to 4- and 8-week visits and the mean change in brain Cr from baseline to 8-week visits. Statistical significance was set at  $p < 0.05$ . **RESULTS/ANTICIPATED RESULTS:** Participants were 65% male with a mean age of  $73.1 \pm 6.3$  years. All participants completed the study, with 19 out of 20 achieving the dose compliance target of  $\geq 80\%$ . The mean self-reported dose

intake was 90%. Serum Cr levels were significantly increased at 4- and 8-week visits compared to baseline ( $0.6 \pm 0.4$  mg/dL vs.  $14.0 \pm 9.9$  mg/dL and  $15.0 \pm 13.6$  mg/dL, respectively;  $p < 0.001$ ). Brain Cr levels also significantly increased ( $330.5 \pm 36.80$  i.u. vs.  $366.9 \pm 57.52$  i.u.,  $p < 0.001$ ). **DISCUSSION/SIGNIFICANCE OF IMPACT:** We are the first to demonstrate that 20 g/day of CrM for eight weeks is feasible and associated with increased brain Cr in patients with AD. Our findings support further investigation of brain target engagement of CrM and its efficacy in AD. With AD cases expected to rise, CrM could serve as an effective, affordable therapeutic to slow AD progression.

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#### **Engineered multifunctional wound healing patch: an antimicrobial living bandage to improve wound healing**

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**OBJECTIVES/GOALS:** The study focuses on developing a wound patch that employs a biocompatible matrix which incorporates mesenchymal stem cells (MSCs) with wound healing and antimicrobial properties, along with antimicrobial metallic nanoparticles covered with keratinocytes derived from induced pluripotent stem cells to replicate the skin's barrier function. **METHODS/STUDY POPULATION:** In vitro experiments will be conducted to combine bacteria with MSCs and metallic nanoparticles to assess whether bacterial killing is improved by this combination. The MSCs will then be evaluated in the presence of the nanoparticles to confirm that their functionality and phenotype are not altered. To verify the cells' functional integrity, they will undergo trilineage differentiation, surface marker phenotypic testing, and evaluation of their capacity to inhibit lymphocyte proliferation in the presence of the nanoparticles. Subsequently, this living bandage will be created using a biomatrix embedded with induced pluripotent stem cell-derived keratinocytes and tested on a canine wound model to study the impact on healing. The model will assess the rate of healing and cellular response at weekly intervals until healed. **RESULTS/ANTICIPATED RESULTS:** The combination of mesenchymal stem cells and antimicrobial nanoparticles works synergistically to enhance bacterial killing in vitro with *S. aureus*. The presence of the nanoparticles in combination with MSC did not affect the ability of the MSC to undergo trilineage differentiation. We anticipate that the surface phenotype will be similarly unaffected. In addition, we expect that the presence of the nanoparticles should not interfere with the ability of MSC to suppress lymphocyte proliferation. Utilization of the wound patch in the in vivo canine wound model is expected to enhance healing and prevent infection. We expect that we will observe a shift in the cellular composition of the wound with less inflammatory cells and more M2 or wound healing anti-inflammatory monocytes. **DISCUSSION/SIGNIFICANCE OF IMPACT:** The incidence of resistant infections with no pharmacologic therapy available are on the rise. The development of an antimicrobial living bandage that increases the body's ability to fight off infection, while providing a barrier to reinfection would provide a new way to treat infections regardless of their acquired antibacterial resistance.