cortex, and temporoparietal junctions in the DMN. RESULTS/ ANTICIPATED RESULTS: Boruta ML analysis identifies the DMN as the most salient functional network for differentiating OUD from HC, with 33% of DMN features found significant (p < 0.05), compared to 10% and 0% for the SN and ECN, respectively. The Boruta ML algorithm identifies age and education as the most significant demographic features. Brain activity mapping shows heightened neural activity in the DMN for OUD. The DMN exhibits the greatest discriminative power, with a mean AUC of 69.74%, compared to 47.14% and 54.15% for the SN and ECN, respectively. Fusing DMN BOLD features with the most important demographic features improves the mean AUC to 80.91% and the F1 score to 73.97%. Follow-up Boruta analysis highlights the mPFC as the most important functional hub within the DMN, with 65% significant features. DISCUSSION/SIGNIFICANCE OF IMPACT: Our study enhances the understanding of OUD neurobiology, identifying the DMN as the most significant network using ML rs-fMRI BOLD feature analysis. Ethnicity, education, and age rank are the most important demographic features and the mPFC emerges as a key functional hub for OUD. Future research can build on these findings to inform treatment of OUD.

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## Using machine learning to analyze voice and detect aspiration

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OBJECTIVES/GOALS: Aspiration causes or aggravates lung diseases. While bedside swallow evaluations are not sensitive/specific, gold standard tests for aspiration are invasive, uncomfortable, expose patients to radiation, and are resource intensive. We propose the development and validation of an AI model that analyzes voice to noninvasively predict aspiration. METHODS/STUDY POPULATION: Retrospectively recorded [i] phonations from 163 unique ENT patients were analyzed for acoustic features including jitter, shimmer, harmonic to noise ratio (HNR), etc. Patients were classified into three groups: aspirators (Penetration-Aspiration Scale, PAS 6-8), probable (PAS 3-5), and non-aspirators (PAS 1-2) based on video fluoroscopic swallow (VFSS) findings. Multivariate analysis evaluated patient demographics, history of head and neck surgery, radiation, neurological illness, obstructive sleep apnea, esophageal disease, body mass index, and vocal cord dysfunction. Supervised machine learning using five folds cross-validated neural additive network modelling (NAM) was performed on the phonations of aspirator versus non-aspirators. The model was then validated using an independent, external database. RESULTS/ANTICIPATED RESULTS: Aspirators were found to have quantifiably worse quality of sound with higher jitter and shimmer but lower harmonics noise ratio. NAM modeling classified aspirators and non-aspirators as distinct groups (aspirator NAM risk score 0.528+0.2478 (mean + std) vs. non-aspirator (control) risk

score of 0.252+0.241 (mean + std); p DISCUSSION/SIGNIFICANCE OF IMPACT: We report the use of voice as a novel, noninvasive biomarker to detect aspiration risk using machine learning techniques. This tool has the potential to be used for the safe and early detection of aspiration in a variety of clinical settings including intensive care units, wards, outpatient clinics, and remote monitoring.

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## Automated assessment of facial nerve function using multimodal machine learning

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OBJECTIVES/GOALS: Current popular scoring systems for evaluating facial nerve function are subjective and imprecise. This study aims to quantify speech and facial motor changes in patients suffering from facial palsy after cerebellopontine angle (CPA) tumor resection to lay the foundation for a scoring algorithm that is higher resolution and more objective. METHODS/STUDY POPULATION: We will obtain audio and video recordings from 20 adult patients prior to and after surgical resection of unilateral CPA tumors between October 2024 and February 2025. We will obtain preoperative recordings within two weeks prior to surgery and postoperative recordings following a preset schedule starting from the day after surgery up to one year. Audio recordings entail patient readings of standardized passages and phonations while video recordings entail patient performance of standardized facial expressions. We will analyze video data for key distance measurements, such as eye opening and wrinkle size, using DynaFace. We will process audio data using VoiceLab to extract metrics such as prominence and tonality. We will perform statistical tests such as t-tests and ANOVA to elucidate changes across time. RESULTS/ ANTICIPATED RESULTS: I expect to obtain 9 sets of audio and video recordings from each of the 20 participants. In terms of speech, I expect average speech duration to increase postoperatively. Similarly, I expect to find increases in time spent breathing, number of breaths taken, and mean breathing duration. In terms of facial movement, I expect nasolabial fold length to decrease postoperatively, as well as eye opening size and left-right symmetry at rest. For both audio and video, I expect these changes to revert towards their preoperative baseline as time passes. I also expect average House-Brackmann and Sunnybrook facial grading scores to increase postoperatively and then decrease with time, correlating strongly with the video and audio findings. I will use trajectory analysis and time point matching to handle any missing data. DISCUSSION/SIGNIFICANCE OF IMPACT: This study will validate our analysis platform's ability to automatically quantify measurable changes that occur to speech and facial movement which correlate strongly with existing scoring systems. Future work will synthesize these data streams to move towards establishing biomarkers for facial nerve function that aid clinical decision-making.