Impact of Nuedexta on Bulbar Physiology and Function in ALS

NCT03883581

Study Protocol

FULL PROTOCOL TITLE
Impact of Nuedexta on Bulbar Physiology and Function in ALS

Protocol Version: 2.0
Protocol Date: 08/01/2019

Protocol Principal Investigators (PPI):
Lauren Tabor-Gray, PhD, CCC-SLP
Department of Neurology
Phil Smith Neuroscience Institute
Holy Cross Hospital
Fort Lauderdale, FL 33308
Email: lauren.tabor@holy-cross.com

Emily Plowman, PhD, CCC-SLP
Department of Speech, Language, and Hearing Sciences
College of Public Health and Health Professions
University of Florida
Gainesville, Florida 32610
Email: eplowman@ufl.edu

Study Intervention Provided by:
ALS Association Clinical Management Grant

Impact of Nuedexta on Bulbar Physiology and Function in ALS.

SIGNIFICANCE.

Progressive degeneration of bulbar musculature results in dysphagia (swallowing impairment) and dysarthria (speech impairment) in individuals with ALS. Regardless of disease onset type, bulbar dysfunction impacts 85% of ALS individuals at some point in the disease course (Carpenter et al., 1977) with patients rating the inability to communicate or eat as the worst symptoms of the disease (Paganoni et al., 2015). Bulbar dysfunction contributes to malnutrition, social isolation, increased caregiver burden, compromised pulmonary function, and increased mortality in ALS (Boitano, 2006; Bouteloup et al., 2009; Tabor et al., 2016). Despite these devastating sequelae, advances in treatment are lacking and primarily consists of a palliative symptom management approach including diet and environmental modifications, feeding tube placement and alternative communication strategies, which are incorporated in a *reactive* manner following identification of impairments (Miller et al., 2009). The lack of targeted treatment options to *proactively* improve or maintain bulbar function represents a crucial clinical management gap in the care of individuals with ALS (Kühnlein et al., 2008; Plowman et al., 2017). **Currently there exist no efficacious treatments to proactively prolong safe and efficient oral intake, or extend functional communication.**

Although advances in the management of bulbar dysfunction in ALS have been disappointing, recent interest has surfaced regarding the therapeutic potential of a pharmaceutical agent, **Nuedexta** (dextromethorphan HBr and quinidine sulfate), for the treatment of bulbar symptomology in individuals with ALS. Although Nuedexta received approval from the Food and Drug Administration (FDA) to target symptoms of pseudobulbar affect (PBA) in ALS; anecdotal reports of improvements in speech, salivation or swallowing were reported from Neurologists treating ALS individuals who were administered Nuedexta (Smith, personal report). Subsequently, a Phase II clinical trial was conducted that reported improvements in speech, swallowing and salivation following 30-days of Nuedexta treatment (Smith et al., 2017). One serious limitation of this study, however, is the fact that the primary outcome employed was a *perceptual patient-report scale* (PRO) (Center for Neurological Study Bulbar Function Scale, CNS-BFS), with *no objective physiologic outcomes* to confirm actual change in bulbar physiology. The absence of any objective clinical physiologic outcomes is particularly important when examining effects of Nuedexta, given that it contains selective serotonin reuptake inhibitors (SSRIs), or serotonergic antidepressants, that can impact the regulation of emotional expression, feelings of wellbeing and modulation of depression (all known to impact the response an individual will provide on a PRO measure). Furthermore, findings based on PRO's must be validated with studies that utilize objective physiologic outcomes of speech and swallowing function. Great excitement exists regarding the potential impact of Nuedexta on bulbar function in ALS with many neurologists prescribing Nuedexta to treat these symptoms in ALS patients. To date, however; **no data exists to examine and determine the physiologic impact of Nuedexta on speech or swallowing physiology**. These data are needed in order to validate the initial patient-reported outcomes of the Phase II clinical trial (Smith et al., 2017) and to provide *evidence-based guidance* to the management of bulbar dysfunction in ALS.

Therefore, the **primary goal** of this proposal is to rigorously evaluate the impact of a pharmacological treatment, Nuedexta, on bulbar function in individuals with ALS. The **central supposition** is that functions of swallowing, airway protection and communication will improve following 30-days of Nuedexta treatment. The following specific aims will test this premise***:***

Aim 1. Determine the effect of Nuedexta on swallowing physiology and airway defense physiologic capacity in ALS.

Hypotheses: Following 30-days of Nuedexta treatment, ALS individuals will demonstrate:

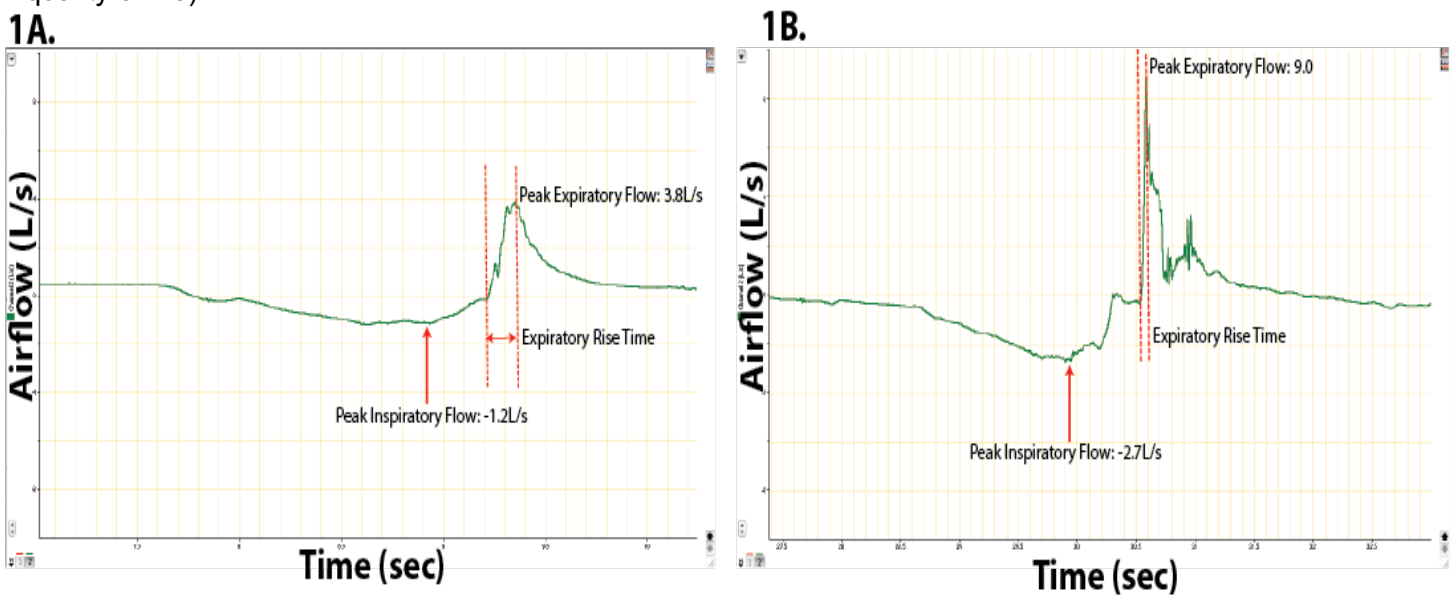
(1) Improved *global* swallowing function as indexed by lower total DIGEST scores
(2) Improved swallowing *safety* as indexed by lower penetration-aspiration scores
(3) Improved swallowing *efficiency* as indexed by lower normalized residue ratio scale scores
(4) Improved *airway defense* physiologic capacity as indexed by increased expiratory cough flow velocity.

Aim 2. Determine the effect of Nuedexta on speech intelligibility, speaking rate and communicative efficiency in ALS.

Hypothesis: ALS individuals will demonstrate increased speech intelligibility, speaking rate and communicative efficiency on the sentence intelligibility test following 30-days of Nuedexta treatment.

Aim 3. Determine the impact of Nuedexta on patient-perceived swallowing function, communication effectiveness, and disease severity and progression.
Hypothesis: Following 30-days of Nuedexta treatment, ALS individuals will report *improvements* on validated, patient-reported measures of swallowing (Eating Assessment Tool-10); communication (Communication Participation Item Bank-10 survey); bulbar function (Center for Neurological Study-Bulbar Function Scale) and overall disease severity (ALS Functional Rating Scale-Revised).

SUPPORTING PRELIMINARY DATA.
We recently completed a randomized sham-controlled trial to investigate the impact of expiratory muscle strength training (EMST) on respiratory and bulbar function in ALS (Plowman et al., 2018). Three individuals enrolled in this trial were also taking Nuedexta. Upon stratification of data following completion of this clinical trial was the finding that patients who were also on Nuedexta treatment (n=3) demonstrated superior gains to those completing EMST alone. An example of one such participant's cough spirometry data is depicted below and demonstrates that their airway defense capabilities to expel secretions or tracheal aspirate significantly improved (Figure 1). Specifically, cough volume acceleration increased by 570%, representing the largest gain of any patient in the entire trial (n=48). Further, peak inspiratory and expiratory flow rates (crucial for high velocity airflow) improved by 125% and 137%, respectively. Of clinical significance is the fact that the *baseline* peak expiratory cough flow of this patient (3.8L/s or 228L/min), did not met the physiological requirements to expel tracheal aspirate or secretions (threshold: 270L/min). However, peak expiratory cough measures take at the end of the clinical trial did (9.0L/s or 540L/min), representing a high-velocity cough proficient for defending the airway. In addition, this patient's forced vital capacity increased from 86% to 97% of predicted, ALSFRS-R Bulbar subscale score from a 10 to an 11, functional oral intake score from a 6 to 7 (higher scores = less dietary restriction), and swallowing-related quality of life score from an 81 to 90 (higher scores= better reported quality of life).



Individual ALS patient who completed eight-weeks of EMST who was also taking Nuedexta. This individual demonstrated superior gains in airway defense capacity on cough spirometry. Peak inspiratory and expiratory flows more then doubled, peak expiratory rise time was significantly reduced and cough volume acceleration improved by 570% and represented the greatest gains of any individual in the clinical trial.

EXPERIMENTAL DESIGN AND METHODS.
Design.
This study represents a **prospective multicenter trial** in 36 individuals with ALS. All eligible and enrolled study participants will be administered the study drug, Nuedexta, as recommended by their treating neurologists, Drs. James Wymer and Eduardo Locatelli. Once enrolled, participants will be assigned a study number and complete a baseline evaluation prior to commencing treatment with Nuedexta. During the baseline assessment, participants will undergo a comprehensive bulbar evaluation of swallowing, airway protection and speech functions, and complete validated patient-reported surveys. Following 30-days of

Nuedexta treatment, participants will be re-evaluated using the same battery of assessments to determine the impact of treatment (Figure 2).

Participants.

This open-label study will include 36 individuals with ALS. We will include patients demonstrating mild to moderate bulbar involvement to avoid both floor and ceiling effects on the outcomes administered.

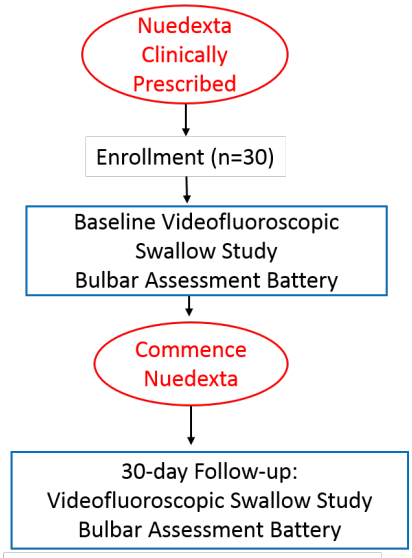


Figure 2. Flowchart of Study Design.

Inclusion Criterion:

1) Diagnosis of probable-definite ALS (El-Escorial Criterion); 2) ALSFRS-R Bulbar subscale score <10; 3) Bamboo oral reading speaking rate ≤140 words per minute; 4) No allergies to barium sulfate.

Exclusion Criterion:

1) Treatment for sialorrhea within the past 3 months that includes either Botox or radiation treatment; 2) Participation in another disease modifying study targeting bulbar or cough function; 3) Use of invasive mechanical ventilation/presence of tracheostomy; 4) Advanced frontotemporal dementia or significant cognitive dysfunction; 5) Nil per oral status for feeding (i.e., NPO, nothing by mouth); and 6) Previously prescribed Nuedexta. Additionally, if participants are taking Riluzole or other medications to control sialorrhea, they must be on a stable dose for at least 30 days prior to enrollment in the current study.

Subject Withdrawal Criteria

Inclusion in the study is entirely voluntary and subjects may withdraw at any time for any reason. Subjects wishing to stop medication will immediately discontinue test article and the testing procedures. Withdrawal due to adverse event will be followed until clearance of sequelae of the adverse event.

Recruitment.

A total of 36 individuals with ALS between the ages of 18-90 will be recruited for the study. ALS patients will be identified and recruited from two large multidisciplinary ALS Centers: 1) the University of Florida Movement Disorders Center in Gainesville, Florida (Site #1), and 2) the Phil Smith Neuroscience Institute at Holy Cross Hospital in Fort Lauderdale, Florida (Site #2). The PI's work alongside the treating neurologists at each site to identify eligible participants throughout the study (Plowman and Wymer at site 1; Tabor and Locatelli at Site 2). Targeted recruitment is 36 participants across sites, with 20 being recruited from the Neuroscience Institute and 16 from the University of Florida clinic. Census data across both sites indicates feasibility, with an average of 45 patients seen each month across the study sites.

Informed Consent

Written informed consent will be obtained from each study participant before any study procedures. The participant's willingness to participate in the study will be documented in writing in a consent form approved by an Institutional Review Board. This document will be signed and a copy provided to the participant.

Overview of Experimental Procedures and Outcome Metrics.

Participants will complete two evaluations (baseline and post-Nuedexta treatment) at each site. All testing procedures and the corresponding outcome measures for Aims 1 – 3 are summarized in Table 1. Measures will be administered at two time points to determine if changes in speech and swallowing physiology (Aims 1 and 2) or patient perceptions of speech and swallowing function (Aim 3) occur following 30-days of Nuedexta treatment. Thirty-days represents a valid time period previously utilized in the original clinical trial investigating the efficacy of Nuedexta in improving pseudobulbar affect (Pioro, 2014) and again utilized in the recent PRO clinical trial (Smith et al., 2017). This timeframe also represents a pragmatic and feasible timeline to minimize attrition and missing data points while ensuring adequate time for potential change in bulbar function.

Table 1. Summary of testing procedures and corresponding outcome measures for each aim.

Bulbar Domain:	Testing Procedure:	Validated Outcome Measure:
Swallowing *(Aim 1)*	Videofluoroscopic Swallowing Evaluation	*Global:* Dynamic Imaging Grade Swallowing Toxicity, DIGEST (Hutcheson, 2017) *Airway Safety*: Penetration Aspiration Scale, PAS (Rosenbek, 1996) *Swallow Efficiency:* Normalized Residue Ratio Scale, NRRS (Pearson et al., 2013)
Cough *(Aim 1)*	Peak Cough Flow Meter	Peak Cough Flow (L/min)
	Cough Spirometry	Inspiratory & Expiratory Flow Rates & Durations, Cough Volume Acceleration
Speech *(Aim 2)*	Sustained /a/	Maximum Phonation Duration (sec) and Amplitude (dB).
	Diadochokinetics (DDK)	Number of CV repetitions for /ba/ and for /ta/
	Sentence Intelligibility Test	Intelligibility (%), Rate (WPM), Communication Efficiency Ratio
	Bamboo Passage	Intelligibility (%), Rate (WPM), Pause Duration (sec)
	Iowa Oral Perf' Instrument	Maximum anterior isometric pressure and Lingual physiologic reserve (KPa)
Patient Perception *(Aim 3)*	Patient Report Outcomes	*Bulbar:* Center Neurological Study Bulbar Function Scale, CNS-BFS (Smith, 2011) *Swallowing:* Eating Assessment Tool-10, EAT-10 (Belafsky, 2008) *Speech:* Communication Participation Item Bank-10, CPIB-10 (Baylor et al., 2013) *Global:* ALS Functional Rating Scale-Revised, ALSFRS-R (Cedarbaum, 1999)

Experimental Procedures:

1. ***Videofluoroscopic Evaluation of Swallowing:*** Videofluoroscopy will be utilized to visualize bolus flow events and swallowing kinematics during the swallow. Videofluoroscopy will be performed using a Phillips BV Endura fluoroscopic C-arm unit (GE OEC 8800 Digital Mobile C-Arm system type 718074) housed within the Swallowing Systems Core laboratory (Site #1) and a Shimadzu videofluoroscopy (Flexavision R3 R/F) remote room housed within the Neuroscience Institute (Site #2). Videofluoroscopic data will be captured at 30 frames per second in the lateral field of view using a TIMS Dicom recording system at both sites (Version 3.2, TIMS Medical, TM, Chelmsford, MA). ALS individuals will complete a videofluoroscopic evaluation of swallowing using the following barium bolus presentations: 5cc thin x2, 20 cc thin x2, 5cc pudding x2, barium tablet with thin barium. All barium bolus presentations will be administered by the speech pathologist and all swallows will be cued to synchronize with videofluoroscopy. Participants will be seated in the lateral position with the nasal spine, oral cavity, pharynx, cervical spine, and upper esophageal sphincter in the viewing plane. A lapel microphone will be utilized to record throat clearing and cough during the videofluoroscopy. To ensure participant safety, a modified bolus viscosity, volume or swallowing compensation will be implemented following a bail out criterion of ≥3 episodes of frank aspiration with ineffective clearance of aspirate material. Airway safety during swallowing, bolus efficiency profiles and global swallowing function will be compared within individuals between baseline and final evaluations using validated outcome measures. Both PIs have performed hundreds of these exams and an example of a lateral still shot image from Dr. Plowman's laboratory is shown in Figure 3 with a C2-4 scalar shown.

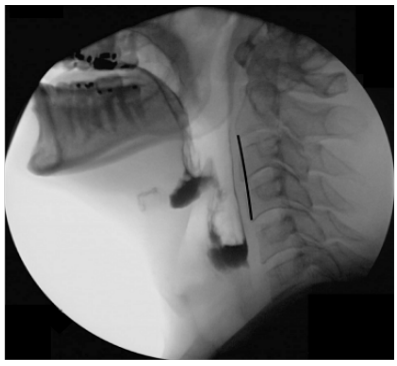


Figure 3. Videofluoroscopic image with a C2-C4 scalar.

2. ***Cough Spirometry Testing:***

Voluntary cough motor output will be assessed using an oral pneumotachograph (MLT 1000, ADInstruments, Inc; Colorado Springs, CO), connected to a disposable spirometry filter (MQ 304 Spirometer Filter, Vacumed; Ventura, CA). Prior to assessment, a three-liter syringe will be used to calibrate airflow and volume. Voluntary

cough testing will be completed in a seated position, with nose clips in place to occlude nasal airflow. Participants will be instructed to hold the spirometry filter comfortably and place their mouth around the mouthpiece prior to starting the task. The clinician will provide assistance for participants who are not able to hold the device due to limb immobility. The clinician will provide a model prior to the task using the following instructions: "Cough hard, like you have something stuck in your throat." This procedure will be repeated three times, with the identical instruction given prior to each trial. The airflow signal will be recorded online to an iMac desktop computer (Apple, Inc. USA) using Lab Chart (AD Instruments, Inc. Version 8), and low pass filtered at 60 Hz for subsequent analysis. The first cough of each epoch (i.e., series of coughs) will be analyzed, yielding a total of three coughs per participant. The maximum cough airflow parameters of the three coughs will be utilized for analysis for each participant. Examples of previously collected voluntary cough spirometry airflow from individuals with ALS are provided in both Figures 1 and 9.

Figure 4. An individual completing peak expiratory cough flow testing using a handheld cough flow meter.

3. Peak Cough Flow Testing:

Peak cough flow will be assessed using a commercially available handheld digital peak cough flow meter (MicroLife PF100). With the participant comfortably seated, and wearing nose clips, they will be instructed to take a deep breath in and then to "cough hard like you have something stuck in your throat" (into the peak flow meter). An example of peak cough flow testing is provided in Figure 4. Three trials will be performed with the best result used for subsequent statistical analyses.

4. Speech Testing:

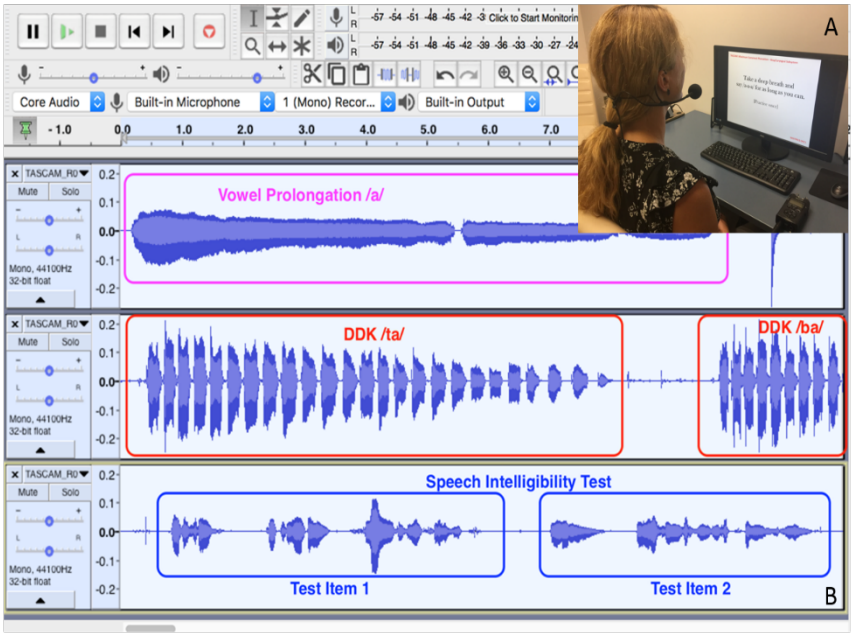


Figure 5. (A) Experimental setup for speech testing and (B) an example of collected speech data output.

Speech testing will occur with the participant comfortably seated at a standardized distance from a computer screen. A digital audio recorder (TASCAM, DR40) connected to a condenser headset microphone (AKG, Inc., HSC271) with a lapel windscreen (eBoot, Inc.) will be placed on the participants head with the headset microphone at a standardized distance of 10cm from the right lip corner. Audio recordings will be digitized at 44.1 kHz with a 16-bit quantization. Calibration of the microphone will be completed prior to each test. Speech testing items will be recorded and saved for subsequent offline analysis. An example of our experimental speech testing setup and collected speech data are provided in Figure 5.

Each participant will complete four speech tasks at the baseline and final evaluation time points that include:

a. *Sustained /a/:* The participant will be instructed to take a deep breath in and then sustain the vowel 'ah' (as in f**a**ther) for as long and steady as possible at a comfortable pitch and loudness.
b. *Diadochokinetics (DDK):* The participant will be asked to perform two DDK tasks for the CV combinations of /ba/ and /ta/. They will be asked to take a deep breath in, and then on one breath repeat the CV combination "ba" as many times as they can until they run out of breath. They will be instructed "Take a deep breath. Say "ba" as clearly and as fast as you can, for as long as you can. /Ba-ba-ba-ba-ba-ba-ba…./". The same instructions will be provided for the /ta/ DDK task.
c. *Speech Intelligibility Test (SIT):* The abbreviated 10-item SIT assessment (SIT for Windows, Madonna Rehabilitation) will be performed. This is a widely used assessment to determine speaking intelligibility of persons with motor speech impairments during oral reading tasks. Participants will be

given ten randomly generated sentences, which get progressively longer in word and sentence length, to read aloud at a comfortable speaking rate and loudness. Sentence intelligibility, speaking rate and communicative efficiency will be generated for analysis.

d. *Bamboo Passage Reading:* The 'Bamboo Passage' is a validated speech testing item developed specifically for individuals with ALS to determine speaking rate, and number and duration of pauses during speech (Green et al., 2004). The clinician will instruct the participant to read the passage in their normal speaking rate and loudness.

5. Lingual Strength Testing:

Lingual strength testing will be performed using the Iowa Oral Performance Instrument (IOPI, IOPI Medical LLC, Woodinville,WA). This is a commercially available device used to index tongue strength using a handheld, portable unit that connects to an air-filled pressure bulb using a connector cable. With the participant comfortably seated, they will be introduced to the device and bulb, and instructed to place the lingual bulb directly behind the central incisors to rest on the alveolar ridge (Figure 6). The participant will be provided a diagram of bulb placement in the oral cavity to facilitate correct placement and the examiner will verify placement of the bulb. Once in place, the participant will complete two tasks, each three times, and the highest recording will be used for subsequent analysis. The participant will be instructed to raise the front portion of their tongue and press as hard as they can onto the bulb. The bulb will remain in the same placement and the participant will be instructed to swallow their saliva normally with the bulb in place. Water will be provided between tasks as needed to facilitate swallow initiation.

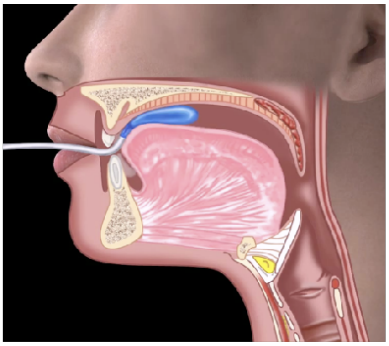


Figure 6. The Iowa Oral Performance Instrument (IOPI) lingual bulb for assessing lingual strength and physiologic lingual reserve.

6. Patient Reported Outcomes:

Four patient report outcomes will be administered to the participant at baseline and final evaluations. Using REDCap, an electronic database capturing system, participants will be electronically sent surveys prior to their appointment to complete in the comfort of their own home to minimize appointment times. Should they require special assistance, we will ask a caregiver to help with reading or entering the data (they will be instructed not to influence the participant or help with scoring). The participant will also be afforded the option to fill out surveys during their clinic appointment if they prefer. The following items will be administered:

a) *Center Neurological Study Bulbar Function Scale, CNS-BFS (Smith, 2011):* The CNS-BFS is a 21-item validated, patient-reported scale indexing degree of bulbar dysfunction in the domains of speech, salivation and swallowing. A copy of the CNS-BFS appears in Appendix A.

b) *Eating Assessment Tool-10 (Belafsky, 2008):*The EAT-10 is a 10-item validated self report scale indexing an individuals perceived level of swallowing impairment. Patients rate each of the 10 items on a 5 point ordinal scale (0=no impairment, 4=severe impairment) with a total EAT-10 score ranging between 1 and 40. A copy of the EAT-10 appears in Appendix B.

c) Communication Participation Item Bank-10, CPIB-10 (Baylor et al., 2013): The CPIB-10 is a 10-item validated, patient-reported survey assessing communicative effectiveness across contexts ranging from 0 (ineffective) to 30 (effective*).* A copy of the CPIB-10 appears in Appendix C.

d) *ALS Functional Rating Scale-Revised, ALSFRS-R (Cedarbaum, 1999):* The ALSFRS-R is the validated, gold standard survey assessing ALS symptom and disease progression to determine disease severity ranging from 0 (severe disease symptomology) to 48 (no overt symptoms). A copy of the ALSFRS-R appears in Appendix D.

Outcome Measures:

All swallow, cough or speech physiologic outcomes will be *double-rated* by two experienced and independent raters. They will be *blinded* to participant ID and also to the test number. If any discrepancies in ratings occur, a consensus meeting will held and if needed, a third rater will be consulted until agreement can be reached.

***Global Swallowing Function*:** The validated Dynamic Imaging Grade of Swallowing Toxicity (DIGEST) will be performed on all collected videofluoroscopic swallowing studies to assess global swallowing function (Hutcheson

and Fuller, 2015). The DIGEST total score is determined using the composite of individual airway safety and bolus efficiency subscores (range: 0-4). The DIGEST total is rated on a 5-point ordinal score ranging from 0 (no dysphagia) to 4 (life-threatening dysphagia). For statistical analysis, DIGEST total scores will be compared within individuals between the baseline and final evaluation time points. DIGEST total scores of <2 indicate functional swallowing and scores ≥ 2 indicate dysphagia. The DIGEST scoring schema is provided in Appendix E.

Swallowing Safety:

The Penetration-Aspiration scale (PAS, (Rosenbek et al., 1996) is a validated eight-point ordinal scale indexing the degree of airway invasion during swallowing, the participant's response, and whether the invasive material is successfully ejected from the airway (Figure 7). PAS scores of 1 and 2 are considered safe, while scores between 3 and 5 indicate penetration of material in the upper airway at or above the level of the true vocal folds. PAS scores greater than 6 indicate aspiration of material below the level of the true vocal folds into the airway. For statistical comparisons across airway safety status levels the convention is as follow: safe swallowing = PAS ≤2 and unsafe swallowing= PAS>3. Figure 7 depicts the PAS and denotes airway safety groups with representative lateral fluoroscopy images representative of each swallowing safety group collected from our labs.

Airway Safety Group	PAS Score	Definition
SAFE	1	Material does not enter airway.
	2	Material enters the airway, remains above VF, and is ejected from airway.
UNSAFE	3	Material enters the airway, remains above VF and is not ejected from airway.
	4	Material enters the airway, contacts VF and is ejected from airway.
	5	Material enters the airway, contacts VF and is not ejected from airway.
	6	Material enters the airway, passes below VF and is ejected into the larynx or out of airway.
	7	Material enters the airway, passes below VF and is not ejected from airway despite effort.
	8	Material enters the airway, passes below VF and no effort is made to eject.

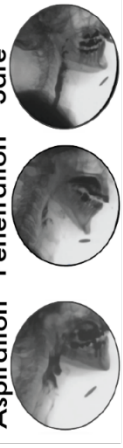


Figure 7. The validated, 8-point penetration-aspiration scale used to rate severity of and reaction to airway invasion during swallowing (Rosenbek et al., 1996).

Swallowing Efficiency:

The validated Normalized Residue Ratio Scale (NRRS, (Pearson et al., 2013) provides a quantitative ratio-based measure of post-swallow pharyngeal residue in the valleculae and piriform sinus (Figure 8). A single frame from the each videofluoroscopy clip will be selected by rater consensus, and ImageJ (ImageJ, free software) will be utilized to derive pixel-based measurements within the pharynx. Specific measurements include: vallecular housing area, vallecular residue, piriform sinus area, piriform sinus residue piriform sinus residue and a C2-C4 anatomical scalar. These will be utilized to calculate severity of pharyngeal residue and are calculated using the *area ratio for the valleculae* multiplied by the *area of the valleculae residue* divided by an *internal scalar* (C2-C4) to account for individual anatomical differences. The same calculation using the piriform area and residue is utilized to determine NRRS of the piriforms. All NRRS ratings will be double rated and inter and intra-rater reliability completed prior to further statistical analysis. NRRS vallecular and piriform sinus values will be compared within-subjects between baseline and final evaluations. In addition, NRRSv values greater than 0.07 and NRRSp values greater than 0.20 indicate greater than normal residue and thus inefficient swallowing.

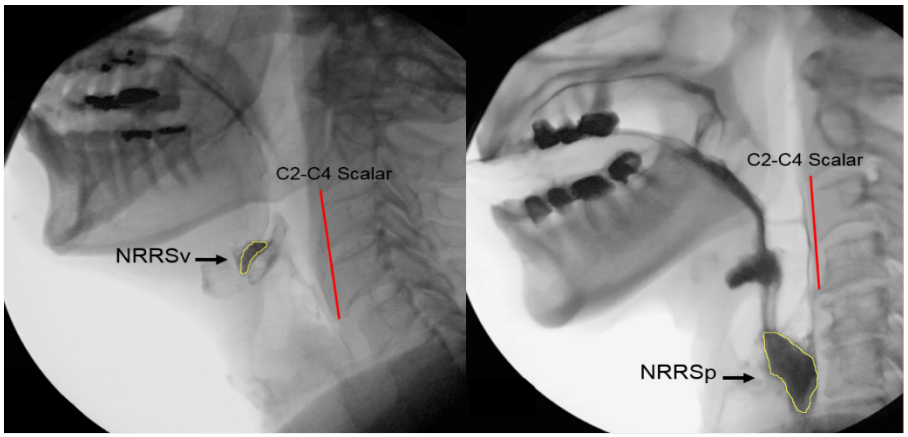


Figure 8. ImageJ imaging software is utilized for analysis of each videofluoroscopic swallowing study. Each rater performs tracing of: (A) Vallecular residue tracing and (B) piriform sinus tracing each with a reference vertebral anatomical scalar (C2-C4) to normalize values to each individual (Pearson et al., 2013)

Cough Function:

Peak cough flow (L/min) and forced expiratory volume within the first second (FEV1) will be derived from peak cough flow testing. The highest flow obtained from three trials will be utilized. Cough spirometry airflow output will be analyzed and six cough parameters derived. The highest values obtained during a single cough epoch will be utilized for analysis. Each parameter is defined and illustrated in Figure 9 below.

Inspiratory phase duration: time from start of inspiration following tidal breathing to the end of inspiration
Peak inspiratory flow rate: time to deepest inspiratory flow during the inspiratory phase of cough
Compression phase duration: apneic period between the end of the inspiratory phase and the start of the expiratory phase
Peak expiratory phase rise: time from beginning of expiration to the greatest peak of the expiratory phase
Peak expiratory phase flow rate: time to reach the peak of the expiratory phase
Cough volume acceleration: peak expiratory flow rate / peak expiratory phase rise time

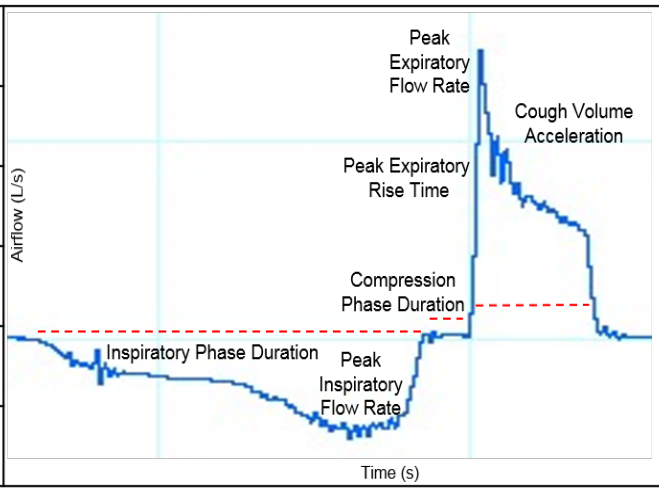


Figure 9. Objective airflow outcome parameters obtained from analysis of voluntary cough waveforms.

Lingual Strength Outcomes:

The following outcomes will be derived from the IOPI testing. The highest maximum anterior isometric pressure and saliva swallowing pressure will be utilized and these metrics will be derived at time of testing. Lingual physiological reserve will be calculated from obtained data.

a) Maximum Anterior Isometric Pressure (MAIP) (KPa).
b) Saliva Swallowing Pressure (KPa)
c) Lingual Physiologic Reserve (LPR) = Maximum anterior isometric pressure – saliva swallow pressure

Speech Outcomes:

Audio recordings obtained during speech testing will be blinded and rated in Audacity (Audacity 2.2.2, WordPress, 2018) for the below speech outcomes.

Table 2. Speech outcomes delineated by speech task.

Speech Task	Derived Outcomes:
Sustained /ah/	Vowel duration (/ah/,sec), maximum amplitude (dB), amplitude fluctuation, pitch fluctuation
DDK	Maximum number of consonant-vowel repetitions produced for /ba/ task and /ta/ task
Speech Intelligibility Test	Intelligibility (%): (number of spoken words correctly transcribed / 110 words spoken) Rate: words per minute = (number of words / 60 * duration of oral reading in seconds) Communication efficiency ratio= (rate of intelligible speech / rate of speech of healthy adults)
Bamboo Passage	Intelligibility (%): (number of spoken words correctly transcribed / 98 words spoken) Rate: Words per minute= (number of words / 60 * duration of oral reading in seconds) Number of Pauses Duration of Pauses

Patient-Reported Outcomes:

a) CNS-BFS: The following scores will be calculated. Total CNS-BFS score (range: 21-111); CNS-BFS Speech subscale score (range: 7-41); CNS-BFS Salivation subscale score (range: 7-35) and CNS-BFS Swallowing subscale score (range: 7-35).
b) EAT-10: A total EAT-10 score will be derived and will range between 0 (no impairment) and 40 (severe impairment).
c) CPIB-10: A total CPIB-10 score will be derived and will range between 0 (severe impairment) and 30 (no impairment).
d) ALSFRS-R: A total ALSFRS-R score will be calculated (range: 0-48) as well as a Bulbar ALSFRS-R Subscale score (range: 0-12).

Statistical Considerations.

Sample Size: Robust improvements in pseudobulbar affect and patient-reported bulbar function were reported following Nuedexta treatment in individuals with ALS, therefore, clinically significant improvements in physiologic parameters of speech and swallow functions are hypothesized (Pioro, 2014). A sample of 30 participants is needed to detect clinically relevant changes in bulbar physiology and function. For repeated measure analyses, this sample will be able to detect minimum differences (between baseline and final evaluations) of 1 L/min peak expiratory flow rate and 25% differences in ordinal outcomes (i.e., PAS, DIGEST). Given that this study represents the first to investigate the impact of Nuedexta on speech function, we utilized a minimal detectable difference for N=30 of 10 words per minute during the oral reading task for the sample size determination. We will recruit an additional 6 participants, assuming a 20% dropout rate to ensure we meet our target data collection number.

Data Analyses.

Dr. Terrie Vasilopoulos, a consultant on this project, will perform all statistical analyses. Analyses will be completed using IBM SPSS Software (Version 24) and SAS Software (SAS Inst., Cary, NC). $P<0.05$ will be considered statistically significant. Intra- and Inter- rater reliability will be performed on all non-automated ratings using the intraclass correlation coefficient. To assess the impact of Nuedexta on swallow, cough, lingual, speech and PRO metrics a repeated measure analysis of variance (ANOVA) model will be used (parametric variables) and a Wilcoxon signed rank test (non-parametric data) with alpha set at 0.05. Corrections for multiple comparisons will be implemented, as needed.

Data Management and Safety Monitoring and Adverse Event Reporting.

Data will be collected across two time points (baseline and post-Nuedexta treatment) at both sites using a centralized data management program (REDCap) for surveys and physiologic data that do not require post-testing analyses. REDCap is an electronic data capture system used for research with a software toolset and workflow methodology for electronic collection and management of research and clinical trial data. REDCap is a scalable, secure, enterprise-level application. The software is delivered via 256 bit SSL-encryption, and features: 1) an intuitive interface for the creation of case report forms (CRFs) and validated data entry; 2) audit trails for tracking data manipulation and export procedures; 3) procedures for importing data from external sources; the ability to relate CRFs to study events and schedule them via a calendar function; and 4) automated export procedures for seamless data downloads to common statistical packages (SPSS, SAS, STATA, R). Data from both sites will be uploaded and accessible through the secured REDCap database.

The UF-IRB will review the human subject protocol and will monitor the quality of research occurring under the approved protocol. The IRB must approve the protocol annually, as the study progresses. Data safety monitoring will occur during weekly meetings to review Adverse Events and Protocol Deviation Logs with the PI and study team. In the event of an unanticipated adverse response, the participant's physician will be immediately notified and the patient will be seen for a complete evaluation. Data, adverse events, and individual subject safety are monitored throughout each subject's evaluation as well as during weekly laboratory meetings with the PI and study team that adhere to the UF IRB regulations for reporting of AE's and protocol deviations. Addtionally, monthly conference calls between sites will occur to share or discuss any adverse events or protocol deviations.

Study Drug Handling and Dispensing.

To offset costs in this small scale grant, the study drug will be clinically recommended and obtained through each participant's standard insurance. Per Drs. Wymer and Locatelli recommendation, Nuedexta will be prescribed in clinic as medically necessary and participants will be subsequently enrolled in the study. All participants will complete their baseline evaluation prior to starting the study drug.

Study Drug Administration and Dosing

The drug will be administered per the efficacy and safety protocol, with no changes in administration method or recommended dose for individuals with ALS. Specifically, Nuedexta (20 mg dextromethorphan HBr and 10mg quinidine sulfate) will be administered orally with 1 capsule every day for the initial 7 days followed by 1 capsule every 12 hours for the remaining 23 days of the study. Medication will be taken whole and should not be divided, crushed, chewed or dissolved.

Study Timeline.

Participants will be enrolled March 2019- March 2020, with all participants' final evaluations completed by October 2020 to permit time for final data analysis and manuscript preparation. An overview of the proposed experimental timeline is detailed in Figure 10.

<table>
<tr><th colspan="4">Study Timeline</th></tr>
<tr><th>March 2019</th><th>March 2019-March 2020</th><th>March-December 2020</th><th>December-March 2021</th></tr>
<tr><td rowspan="2">Obtain IRB approval (Sites 1 and 2)</td><td>Study commencement
Enroll and complete 20 participants</td><td>Enroll and complete 10 participants</td><td rowspan="2">Final data analysis and manuscript preparation</td></tr>
<tr><td colspan="2">Ongoing Recruitment and Data Management</td></tr>
</table>

Figure 10. Proposed study timeline.

Potential Pitfalls and Limitations.

Drug Tolerability and Attrition.

Nuedexta is approved by the United States Food and Drug Administration and has been proven safe and tolerable in several neurological populations including ALS (Pioro, 2014). In the event a participant cannot tolerate the medication, or chooses to discontinue use, they will be considered a drop out of the study. Subject compliance will be assessed at both visits with questioning about compliance and adverse events.

Competing Trials.

Participation in the current study requires the subject to be on a steady dose of additional medications that impact overall disease progression (i.e., Riluzole). Participation in other disease modifying studies targeting bulbar or cough function represents an exclusion criteria for this study. Given that this protocol is only 30 days, we anticipate that interested ALS individuals will be immediately enrolled in the current study and then, once completed, be offered participation in other potentially disease modifying treatments. The PI's will communicate and closely monitor potential participation in novel experimental treatments that may impact bulbar function during the course of the study. Additionally, information regarding standard of care ALS treatments and interventions (i.e., cough assist, BiPap, medications) will be monitored and recorded throughout study participation.

Patient Recruitment.

ALS is considered a rare disease, with a prevalence of 4 in every 100,000 people (Mehta et al., 2014) and patient recruitment and attrition often inhibits successful completion of trials in ALS. To mitigate potential limitations in recruitment, we are working with and recruiting from two large ALS Centers in Florida including: University of Florida- Gainesville (Dr. Plowman) and the Neuroscience Institute (Dr. Tabor). Nuedexta is commonly prescribed in their respective multidisciplinary ALS clinics and both Centers are actively involved in research. Further, both PI's work closely with the clinic neurologists (Drs. Wymer and Locatelli) and census data indicates feasibility for the proposed study sample size. Additionally, Florida has the second highest prevalence of ALS patients in the United States. Therefore, we do not anticipate patient recruitment being a limitation.

Multisite Design.

Standardization of procedures and selected outcomes can represent a potential pitfall in multi-center clinical trials. The PI at study site 2 (Dr. Tabor) was trained for 8 years by Dr. Plowman and has previously run large clinical trials for Dr. Plowman during her training in the Swallowing Systems Core. The two PI's are therefore extremely calibrated in terms of methods, approaches and analytical techniques. Further, they have identical set ups of equipment making such a multisite seamless. Another potential limitation of multi-site trials is communication across sites. Given the long history of the two PI's and a well established track record including approximately 15 publications and successful completion of previous collaborative studies, we view this as a potential strength. Nonetheless, we set in place a monthly conference calls and a quarterly site visit to mitigate potential issues with communications across study sites. Further, the use of REDCap will facilitate collaboration, data management and ease of data sharing throughout the duration of the study.

The University of Florida is the lead site and will be responsible for ensuring that both sites have the most current version of the protocol and consent/HIPAA documents. All required approvals (initial, continuing review, and modifications) will be obtained at each site independently (including by the site's IRB of record). All modifications will be communicated to sites, and approved (including approval by the site's IRB of record) before the modification is implemented. All engaged participating sites will safeguard data, including secure transmission of data, as required by local information security policies. All local site investigators will conduct the study in accordance with applicable federal regulations and local laws. All non-compliance with the study protocol or applicable requirements will be reported in accordance with University or local policy.

Routine practices for ensuring the confidentiality and privacy of all participants will be followed in this study. All research personnel at the University of Florida are required to sign a confidentiality agreement at the time of hire. Research personnel at the Phil Smith Neuroscience Institute at Holy Cross Hospital are similarly bound by a confidentiality agreement outlined by the ethics board at their institution. Participants will be assigned a non-identifying alphanumeric study code, and the master key for this code will be retained separately by the PI and study coordinator in a password-protected file on a secure, password-protected, encrypted research server. Daily back-up of this research server is performed to protect against data loss.

Additionally, any data transfer will occur utilizing REDCap. The transfer of anonymized data containing the information needed for analysis will be covered by a data transfer and sharing agreement. A data transfer agreement is currently being written up between the two sites and will be completely finalized prior to study commencement.

Possible Risks and Discomforts

Although the videofluoroscopic swallowing study represents the gold standard clinical swallowing exam and utilizes an amount of radiation that is thought to be minor, repeated exposures may increase a participant's risk of injury or disease. We will utilize a five minute time cap on actual exposure time to minimize raditation exposure to participants. Additionally, the swallowing study also incorporates the use of barium into the protocol. Some participants might ingest small amounts of barium into their lungs during the swallowing study causing them to cough or feel discomfort.

Possible Benefits

There is no guarantee that participants will benefit from participating in this study; their condition may improve, worsen, or remain the same. Nuedexta has the potential to provide complementary benefits to improve swallowing and speech in people with ALS. Improvement in these parameters will effectively prolong survival and quality of life for people living with ALS.

Conflict of Interest

None of the study members have a conflict interest in regard to this study.

Overall Significance and Future Research.

The proposed study addresses a *critical management gap* in individuals with ALS. Currently, there exist no efficacious treatments to maintain and prolong bulbar function and no group has examined the physiologic impact of this promising pharmacological intervention, Nuedexta, on speech and swallowing function and physiology in this challenging patient population. This proposal will provide the first dataset to determine the potential therapeutic role of Nuedexta for the management of bulbar dysfunction in ALS. Better treatments will lead to improvements in quality of life, functional oral intake, nutrition, communicative effectiveness, pulmonary health and ultimately survival in this patient population. Results of this work will provide valuable insight into a potentially beneficial therapeutic, disease modifying treatment for bulbar dysfunction, highlighting a departure from status quo treatment options.

References:

Baylor C, Yorkston K, Eadie T, Kim J, Chung H, Amtmann D (2013) The Communicative Participation Item Bank (CPIB): item bank calibration and development of a disorder-generic short form. Journal of speech, language, and hearing research : JSLHR 56:1190-1208.

Boitano LJ (2006) Management of airway clearance in neuromuscular disease. Respiratory care 51:913-922; discussion 922-914.

Bouteloup C, Desport JC, Clavelou P, Guy N, Derumeaux-Burel H, Ferrier A, Couratier P (2009) Hypermetabolism in ALS patients: an early and persistent phenomenon. Journal of neurology 256:1236-1242.

Carpenter R, McDonald T, Howard Jr F (1977) The otolaryngologic presentation of amyotrophic lateral sclerosis. Otolaryngology 86:ORL479-484.

Green JR, Beukelman DR, Ball LJ (2004) Algorithmic Estimation of Pauses in Extended Speech Samples of Dysarthric and Typical Speech. Journal of medical speech-language pathology 12:149-154.

Hillel AD, Miller R (1989) Bulbar amyotrophic lateral sclerosis: patterns of progression and clinical management. Head & neck 11:51-59.

Hutcheson K, Barringer, D., Knott, JK., Lin, H. , Weber, RS.,, Fuller C, Lazarus, C., May, AH., Patterson, JM., Roe, J., Starmer, HM., Lewin, JS. (2015) DYNAMIC IMAGING GRADE OF SWALLOWING TOXICITY (DIGEST): SCALE DEVELOPMENT AND VALIDATION. In: Dysphagia Research Society.

Kühnlein P, Gdynia H-J, Sperfeld A-D, Lindner-Pfleghar B, Ludolph AC, Prosiegel M, Riecker A (2008) Diagnosis and treatment of bulbar symptoms in amyotrophic lateral sclerosis. Nature Clinical Practice Neurology 4:366-375.

Mehta P, Antao V, Kaye W, Sanchez M, Williamson D, Bryan L, Muravov O, Horton K (2014) Prevalence of Amyotrophic Lateral Sclerosis — United States, 2010-2011. MMWR Surveillance Summaries 63:1-13.

Miller RG, Jackson CE, Kasarskis EJ, England JD, Forshew D, Johnston W, Kalra S, Katz JS, Mitsumoto H, Rosenfeld J, Shoesmith C, Strong MJ, Woolley SC (2009) Practice Parameter update: The care of the patient with amyotrophic lateral sclerosis: Multidisciplinary care, symptom management, and cognitive/behavioral impairment (an evidence-based review): Report of the Quality Standards Subcommittee of the American Academy of Neurology. Neurology 73:1227-1233.

Paganoni S, Karam C, Joyce N, Bedlack R, Carter GT (2015) Comprehensive Rehabilitative Care Across the Spectrum of Amyotrophic Lateral Sclerosis. NeuroRehabilitation 37:53-68.

Pearson WG, Jr., Molfenter SM, Smith ZM, Steele CM (2013) Image-based measurement of post-swallow residue: the normalized residue ratio scale. Dysphagia 28:167-177.

Pioro EP (2014) Review of Dextromethorphan 20 mg/Quinidine 10 mg (NUEDEXTA((R))) for Pseudobulbar Affect. Neurology and therapy 3:15-28.

Plowman EK, Tabor LC, Wymer J, Pattee G (2017) The evaluation of bulbar dysfunction in amyotrophic lateral sclerosis: survey of clinical practice patterns in the United States. Amyotrophic Lateral Sclerosis and Frontotemporal Degeneration:1-7.

Plowman EK, Tabor-Gray L, Rosado KM, Vasilopoulos T, Robison R, Chapin JL, Gaziano J, Vu T, Gooch C (2018) Impact of expiratory strength training in amyotrophic lateral sclerosis: Results of a randomized sham controlled trial. Muscle & nerve.

Rosenbek JC, Robbins JA, Roecker EB, Coyle JL, Wood JL (1996) A penetration-aspiration scale. Dysphagia 11:93-98.

Smith R, Pioro E, Myers K, Sirdofsky M, Goslin K, Meekins G, Yu H, Wymer J, Cudkowicz M, Macklin EA, Schoenfeld D, Pattee G (2017) Enhanced Bulbar Function in Amyotrophic Lateral Sclerosis: The Nuedexta Treatment Trial. Neurotherapeutics : the journal of the American Society for Experimental NeuroTherapeutics 14:762-772.

Tabor L, Gaziano J, Watts S, Robison R, Plowman EK (2016) Defining Swallowing-Related Quality of Life Profiles in Individuals with Amyotrophic Lateral Sclerosis. Dysphagia.