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Aerodynamic and nonlinear dynamic acoustic analysis of tension asymmetry in excised canine larynges

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Abstract

Objective/hypothesis—To model tension asymmetry caused by superior laryngeal nerve paralysis (SLNP) in excised larynges and apply perturbation, nonlinear dynamic, and aerodynamic analyses.

Methods—SLNP was modeled in 8 excised larynges using sutures and weights to mimic cricothyroid (CT) muscle function. Weights were removed from one side to create tension asymmetry, mimicking unilateral SLNP. Two sets of weights were used, one light and one heavy. Experimental measurements were made for five conditions: no tension; symmetrical light tension; asymmetrical light tension; symmetrical heavy tension; and asymmetrical heavy tension.

Results—Perturbation parameters were not significantly different across conditions (percent jitter: $p=0.451$; percent shimmer: $p=0.321$). Additionally, many measurements were invalid (error values > 10). Second order entropy (K2) was significantly different across conditions ($p=0.002$), while correlation dimension (D2) was not ($p=0.428$). Validity of these nonlinear dynamic parameters was demonstrated by low standard deviations. Phonation threshold pressure ($p<0.001$) and power ($p=0.05$) differed significantly across conditions, while phonation threshold flow did not ($p=0.396$).

Conclusions—Nonlinear dynamic analysis differentiated between symmetrical and asymmetrical tension conditions while traditional perturbation analysis was less useful characterizing type 2 or 3 vocal signals. Supplementing acoustic with aerodynamic parameters may help distinguish among laryngeal disorders of neuromuscular origin.

Keywords

superior laryngeal nerve paralysis; tension asymmetry; nonlinear dynamic analysis; acoustic analysis; aerodynamic analysis

INTRODUCTION

Paralysis of the superior laryngeal nerve (SLN) impairs voice production by decreasing stimulation to the cricothyroid muscle, thereby preventing normal vocal fold elongation. Superior laryngeal nerve paralysis (SLNP) often results from SLN damage due to trauma or injury during surgery (Sulica, 2004). Unilateral SLNP introduces asymmetries to the vocal folds that result in difficulty speaking. Patients with unilateral SLNP generally present with a weak, breathy, raspy, monotonous and low pitched voice accompanied by a loss of vocal

range (Abelson & Tucker, 1981; Dursun et al., 1996; Roy et al., 2009). The SLN innervates the cricothyroid (CT) muscle, which controls vocal fold elongation. The CT is composed of three bellies, the vertical belly, the oblique belly, and the recently described horizontal belly. The identification of the horizontal belly in human larynges is quite recent (Mu & Sanders, 2009) and therefore the actions of vertical and oblique bellies have been more widely studied. The entire cricothyroid muscle originates at the front and lateral part of the cricoid cartilage before diverging into the vertical and oblique bellies. The vertical belly then inserts at the lower border of the thyroid lamina while the oblique belly inserts at the inferior cornu of the thyroid cartilage. When these two bellies contract, they cause anteroinferior rocking of the thyroid cartilage, shortening the cricothyroid space and lengthening the vocal folds (Sulica, 2004). It is believed that the horizontal belly acts primarily on the thyroid horn and CT joint with the oblique belly (Mu & Sanders, 2009). Collectively, studies have shown that CT muscle contraction is most positively correlated with changes in fundamental frequency (F_0) and is therefore considered to be the primary pitch control muscle (Hong et al., 1998). With unilateral paralysis, the patient is unable to generate tension in one vocal fold, making pitch regulation difficult and introducing asymmetries into vocal fold vibration.

The presentation of SLNP among patients varies greatly since the extent of SLN damage, mechanisms for compensation, and normal anatomy differ from patient to patient. Generally, SLNP is first observed during routine laryngoscopy or videostroboscopy. SLNP is then verified using EMG and clinical acoustic measurements (Sulica, 2004). Although laryngeal EMG is the definitive tool (Bevan et al., 1989; Xu et al., 2007) to diagnose SLNP, visual, acoustic, and aerodynamic diagnosis is less time consuming, less invasive, less expensive, and happens earlier in the course of patient contact. Refinement of visual, acoustic, and aerodynamic diagnostic techniques could supplant the use of EMG in the future. Commonly reported visual indications of SLNP are an oblique glottal configuration, shifting of the glottic space at the onset and offset of phonation and excessive vertical motion of the lax vocal fold. In addition, a recent study by Roy et al. identified ipsilateral epiglottic petiole deviation during high pitched voice production as a potentially valuable sign of unilateral SLNP. Larger clinical studies of the reliability of this finding are still required in order to establish diagnostic precision (Roy et al., 2011). There still exists discrepancy in research findings as to the occurrence of these observations since the reported signs of SLNP are heterogeneous and subtle (Abelson and Tucker, 1981; Dursun et al., 1996; Sulica, 2004; Roy et al., 2009). Additionally, the degree of impairment due to SLNP is not well characterized using linear acoustic, aerodynamic, or auditory-perceptual measures. This is largely due to the fact that SLNP often presents along with concurrent pathologies and it is difficult to determine the origin of symptoms and direct effects of SLNP (Roy et al., 2009). Compensatory mechanisms resulting from prolonged paralysis can also hinder the study of SLNP effects and are particularly difficult to manage since they vary among patients, making assessment difficult. The inability to reliably diagnose SLNP is disadvantageous since early detection and treatment can prevent the vocal abuse and damage that often result from SLNP compensation (Dursun et al., 1996; Heman-Ackah & Barr, 2006). A quantitative measure of the differences between normal larynges and those affected by SLNP would have clinical diagnostic benefits. Further development of acoustic analysis techniques could provide a method of characterizing the vocal signals of both normal larynges and those affected by SLNP to provide more accurate diagnosis and treatment evaluation.

Acoustic voice analysis offers methods of quantitatively assessing vocal quality. Methods of acoustic analysis include both perturbation and nonlinear dynamic measurements. Jitter and shimmer are traditional perturbation analysis parameters and have been widely applied as a noninvasive quantitative method of acoustic analysis. These parameters describe the cycle-to-cycle variation in fundamental frequency and amplitude, respectively. The definition of

these parameters indicates that they require some level of periodicity, and recent studies have shown that that in order to calculate these perturbation parameters, the vocal signal must be nearly periodic or the ability of these parameters to describe the vocal signal greatly degrades (Titze, 1995; Zhang & Jiang, 2005; Jiang et al., 2006; Zhang et al., 2008). Vocal signal type has been divided into four categories. Type 1 vocal signals are nearly periodic, type 2 vocal signals have qualitative changes, and type 3 vocal signals have no apparent periodic structure. Type 4 vocal signals have been identified and defined as noise. Perturbation has been shown to be a reliable analysis method for type 1 vocal signals only (Titze, 1995). When fundamental frequencies and peak amplitudes vary for consecutive phonatory cycles for irregular phonations, jitter and shimmer estimations become unstable and unreliable (Jiang et al., 2003). The use of nonlinear dynamic analysis for vocal signals of type 2 and type 3 has therefore undergone evaluation and development as a method to characterize disordered and aperiodic voices. Comparison of these two methods has shown that there is less variability in the nonlinear dynamics parameters than the perturbation parameters when analyzing disordered and aperiodic voices (Zhang & Jiang, 2005; Jiang, 2006). While perturbation analysis is still a valuable tool for analyzing type 1 signals, nonlinear dynamic analysis has been shown to be more effective at characterizing aperiodic phonation and has the potential to serve as a better diagnostic tool when SLNP or other asymmetric pathologies are suspected (Titze, 1995; Zhang & Jiang 2005; Jiang, 2006; Zhang, 2008). Nonlinear dynamic analysis also offers a means of treatment evaluation for pathologies resulting in aperiodic phonation.

Aerodynamic parameters offer a useful supplement to the assessment of SLNP. Asymmetric tension with preserved vocal fold adduction may cause an increase in phonation threshold pressure (PTP) while maintaining a relatively constant phonation threshold flow (PTF). The increase in PTP is correlated with an increase in vocal effort and has been observed in two studies by Roy et al. (Roy et al., 2009; Roy, 2011), where both objective increases in PTP and subjective increases in vocal effort occurred. PTF and phonation threshold power (PTW), the product of PTP and PTF, are relatively new parameters (Jiang & Tao, 2007; Regner & Jiang, 2010) used primarily in experimental investigations. PTF is sensitive to changes in glottal abduction (Hottinger et al., 2007) and may remain rather stable unless vocal fold bowing and consequent increased glottal gap occur. Using aerodynamic parameters may help clinicians distinguish among disorders characterized by near normal vocal fold morphology but abnormal physiology.

In addition to ambiguity in diagnosis, current understanding of SLNP is limited by the fact that the immediate effects of SLNP are difficult to isolate. Excised larynx experiments have been performed to model SLNP while eliminating confounding variables such as concurrent pathologies and compensation. Compensation is of particular concern in cases of chronic paralysis. The acute model of SLNP differs greatly from chronic paralysis and does not show evidence of consistent phase offsets or longitudinal tension asymmetries as noted in clinical studies. If vibratory changes due to CT denervation cannot be noted, the ability to measure progress or gauge prognosis is reduced. Previous excised studies have modeled CT contraction by longitudinally increasing tension asymmetrically using sutures (Berry et al., 1996; Maunsell et al., 2006). Each of these studies isolated the effects of the CT muscle to evaluate the effects of CT muscle paralysis and paresis. These previous studies indicate that the use of sutures to simulate CT contraction is a valid method for studying the effects of the CT paralysis and paresis. The goal of this study is to investigate the effectiveness of nonlinear dynamics to differentiate between normal and pathological conditions. With weighted sutures used to simulate CT contractile forces, both traditional perturbation and newer nonlinear dynamic techniques were applied to excised larynx vocal fold vibration in five simulated conditions: no tension; symmetrical light tension; asymmetrical light tension; symmetrical heavy tension; and asymmetrical heavy tension. Nonlinear analysis may detect

subtle differences in vocal fold vibration which cannot be characterized by perturbation analysis due to the aperiodic character of the vocal signal generated by the asymmetrical tension in vocal folds affected by unilateral SLNP.

METHODS

Obtaining Larynges

Ten larynges without evidence of trauma or vocal disease were excised postmortem from canines sacrificed for non-research purposes according to the protocol described by Jiang and Titze (Jiang & Titze 1993). Canine larynges have long been considered an appropriate model for studying human laryngeal pathologies on the excised bench (Baer, 1975; Hirano et al., 1981; Kurita et al., 1983). Larynges were frozen in 0.9% saline solution within 24 hours of harvesting the tissue. After thawing at room temperature, the epiglottis, corniculate cartilages, cuneiform cartilages, and ventricular folds of the larynx were dissected away to expose the true vocal folds. The larynx was mounted on the apparatus (Figure 1) as specified by Jiang and Titze (Jiang & Titze, 1993). A metal pull clamp was used to stabilize the trachea on a tube connected to a pseudolung, which served as a constant pressure source. Lateral prongs were used to adduct the arytenoids until vocal fold approximation was achieved. Methodological consistency was maintained by always adducting folds to the midline so that the glottal gap was not noticeable. Vocal fold adduction remained constant for all trials.

Data Acquisition—The pseudolung used to initiate and sustain phonation in these trials was designed to simulate the human respiratory system. Pressurized airflow was passed through two Concha Therm III humidifiers (Fisher & Paykel Healthcare Inc., Irvine, CA) in series to humidify and warm the air. The potential for dehydration was further decreased by frequent application of 0.9% saline solution between trials. Airflow was controlled manually and was measured using an Omega airflow meter (model FMA-1601A; Omega Engineering Inc., Stamford, CT). Pressure measurements were taken immediately before the air passed into the larynx using a Heise digital pressure meter (901 series; Ashcroft Inc., Stratford, CT). Acoustic data were collected using a dBX microphone (model dBX; Harman, Sandy, Utah) positioned at a 45° angle to the vertical axis of the vocal tract and placed approximately 10 cm from the glottis to minimize acoustic noise produced by turbulent airflow. Acoustic signals were subsequently amplified by a Symetrix preamplifier (model 302; Symetrix Inc., Mountlake Terrace, WA). A National Instruments data acquisition board (model AT-MIO-16; National Instruments Corp., Austin, TX) and customized LabVIEW 8.5 software (National Instruments Corp.) were used to record airflow, pressure, and acoustic signals on a personal computer. Aerodynamic data were recorded at a sampling rate of 200 Hz and acoustic data at 40,000 Hz. Acoustic data was recorded at 40,000 Hz. Experiments were conducted in a triple-walled, sound-proof room to reduce background noise and stabilize humidity levels and temperature.

Suture Placement

The CT muscle bellies external to the thyroid and cricoid cartilages were dissected away to facilitate suture placement. Insertion points for the muscles were noted as fibers were removed. Placement of the sutures and resulting force vectors can be seen in Figures 2 and 3.

A suture fixing the cricoid cartilage to the trachea was first placed along the midline, defined as the line drawn from the peak of the thyroid prominence to the midpoint of the cricoid arch; this suture prevented displacement of the cricoid relative to the trachea due to the weights simulating CT contraction. Insertion points for the oblique and vertical bellies were

determined from the visible insertion points of the dissected CT muscle. Sutures were then placed at the center of the insertion points of the oblique and vertical bellies on the thyroid cartilage as visually judged from where the CT muscle was dissected away. The sutures were extended along the line of action of the muscle. After unilateral suture placement, the distance and angle between the suture line and the midline of the larynx were measured. The angle of the vertical belly was zero since these sutures were set parallel to the midline. These measurements were then translated to the other side of the larynx to maintain symmetry. The superior margin of the thyroid insertion point was kept standardized on both sides, but suture length and distance of the cricoid cartilage insertion point from the midline varied slightly. Suture angle and the thyroid insertion point were equivalent between sides since the motion of the thyroid cartilage (relative to the cricoid cartilage) was the targeted output of suture forces. Suture placement is shown in Figure 2.

Simulation of CT contraction

An acknowledged weakness of excised larynx research is the lack of innervation. Though we cannot cause SLNP in the excised larynx, we can simulate it by modeling its effect. As SLN firing causes contraction of the CT muscle and consequent increased vocal fold tension, we can simulate SLNP by eliminating the effect of CT contraction. To simulate CT function, a suture was placed at the insertion points of the oblique and vertical bellies of the CT muscle on the thyroid cartilage and extended along the line of action of the muscle. To simulate contraction of the oblique belly, the suture line was draped over a small metal rod to translate the weight of the mass from the vertical direction (Figure 3) to an equal force in line with the sutures. Resting vocal fold length was measured with a caliper. A weight was then fixed to the end of each cord. When weights were selected that would mimic the physiologic CT contraction forces reported in the literature, the larynx was unstable and could not support the weight. Therefore, weights were kept under 150 g as described in Isshiki, 1977 (Isshiki et al., 1977) for all sutures, and weights on the vertical sutures were half that of the oblique weights (assuming the angle of the oblique suture from vertical was 45 degrees). Additionally, the selected weights produced degrees of vocal fold elongation that are commonly used in excised larynx experiments (Jiang et al., 2008). Symmetrical light tension produced an average elongation of 8.7% and symmetrical heavy tension produced an average elongation of 14.8%. In the heavy tension condition, 40 g weights were used to simulate oblique belly contraction and 25 g weights were used to simulate vertical belly contraction. In the light tension condition, 25 g weights were used to simulate oblique belly contraction and 12 g weights were used to simulate vertical belly contraction.

Five conditions were evaluated: no tension, in which no CT contraction was simulated; symmetrical heavy tension, in which bilateral strong CT contraction was simulated; symmetrical light tension, in which bilateral weak CT contraction was simulated; asymmetrical heavy tension, in which strong CT contraction was simulated on one side only; and asymmetrical light tension, in which weak CT contraction was simulated on one side only. Phonation was initiated for each trial by slowly raising the airflow through the pseudolung until the onset of stable phonation. After phonation onset, a 5 second acoustic recording was made. This was repeated for a total of five trials for each of the five conditions.

Data Analysis

Perturbation analysis was performed using CSpeech software version 4.0 (Milenkovic, 1987). Percent jitter and percent shimmer were calculated for each trial using the software. Percent jitter measures the percent of cycle-to-cycle frequency variation in the voice samples. Percent shimmer measures the percent of cycle-to-cycle amplitude variation in the

voice samples. The comparison of these parameters across conditions in this study is used to determine if significant differences between conditions can be found.

Nonlinear dynamic analysis was performed using numerical algorithms established in previous studies (Jiang & Zhang 2002; Jiang, 2003; Zhang & Jiang 2003; Jiang, 2006). These studies detail the methodology of nonlinear dynamic analysis as applied to vocal signals. A phase space is reconstructed by plotting the voice signal against a time delay. When a signal is periodic, the phase space depicts this signal as a closed loop trajectory. When aperiodic and irregular signals are analyzed, the phase space can quantitatively analyze the nonlinear dynamic behaviors in the system. The two nonlinear dynamic parameters used in this study are correlation dimension, D2, and second order entropy, K2. Correlation dimension describes how strongly two points on the trajectory are correlated and describes irregular phenomena well. D2 quantifies the complexity or irregularity of a trajectory in phase space. Trajectories can be classified into four categories: a zero-dimensional fixed point (static states); a one-dimensional limit cycle (periodic oscillation); a two-dimensional quasi-periodic torus (super position of two or more oscillations with no rationally dependent frequencies); or a fractal-dimensional chaotic trajectory (aperiodic oscillations). A higher D2 value indicates that more variables are necessary to describe trajectory behavior. However, chaos can be distinguished from white noise since the estimate of D2 of white noise does not converge, despite the increase of the embedding dimension, m , while the D2 value of a chaotic system converges to a finite value. Second order entropy quantifies the rate of loss of information during the evolution of a dynamic system over time. For regular behaviors including static states, periodic oscillations and quasi-periodic oscillations, K2 will equal zero. For chaotic systems with finite degrees of freedom, K2 will be a finite value. For truly random behavior, K2 will approach infinity. Therefore, a finite and positive K2 value provides a sufficient condition for chaos (Jiang, 2006). The amount of disorder in a system is positively correlated with the number of state variables required to describe the system (D2) and the second order entropy (K2). Accordingly, increased D2 and K2 values are expected in disordered systems.

Statistical Analysis

One-way repeated measures analysis of variance (ANOVA) was performed to determine if there were significant differences across conditions. Paired t-tests were performed to determine if there were significant differences between individual condition pairs. If data did not meet assumptions for parametric testing, a Wilcoxon-Mann-Whitney signed rank test was used. Tests were two-tailed and a significance level of $\alpha=0.05$ was used. The level of significance was kept constant because the pairwise comparisons were known a priori to be of greater interest than the comparisons across all five groups. SigmaPlot 11.0 was used for statistical analysis (Systat Software, San Diego, California).

RESULTS

Vocal fold length

Summary vocal fold length measurements are provided in table 1. Vocal fold length was shortest in the no tension condition and increased incrementally as the total mass of the weights was increased, reaching a maximal length in the symmetrical heavy tension condition. The effect of each condition on vocal fold position can be seen in figure 4.

Aerodynamic analysis

Summary aerodynamic data are provided in table 2. One-way repeated measures ANOVA demonstrated significant differences across conditions for PTP ($p<0.001$) and PTW ($p=0.05$), but not PTF ($p=0.396$). Application of symmetrical light tension resulted in

increased PTP ($p < 0.001$) and PTW ($p = 0.025$) compared to no tension. Values increased further when applying symmetrical heavy tension compared to symmetrical light tension, though the differences did not reach significance (PTP: $p = 0.065$; PTW: $p = 0.099$). PTP was the only parameter sensitive to the difference between symmetrical and asymmetrical light tension (PTP: $p = 0.054$; PTF: $p = 0.546$; PTW: $p = 0.592$). No parameter showed a difference between symmetrical and asymmetrical heavy tension. Both PTP and PTW were significantly different when comparing no tension to asymmetrical light tension (PTP: $p = 0.007$; PTW: $p = 0.040$) and asymmetrical heavy tension (PTP: $p = 0.002$; PTW: $p = 0.016$). The change in PTF approached significance ($p = 0.087$) for the asymmetric heavy tension condition, but not the asymmetric light condition ($p = 0.549$). A bar graph displaying PTP data across conditions is provided in figure 5.

Perturbation analysis

Results from statistical testing are provided in table 3. No significant differences were found on one-way repeated measures ANOVA for either jitter ($p = 0.448$) or shimmer ($p = 0.449$). No significant differences were seen for any of the pairwise comparisons (table 5). Error values for the perturbation parameters exceeded acceptable levels, indicating that the measures were invalid. A high error count for the perturbation analysis indicates a large number of voice breaks that interrupt the pitch track. These voice breaks may exaggerate the percent jitter and percent shimmer values, reducing the ability of perturbation parameters to characterize the signal (Milenkovic, 1987).

Nonlinear dynamic analysis

Results from statistical testing are provided in tables 4 and 5. There were no differences in D2 for the one-way repeated measures ANOVA ($p = 0.428$) or any pairwise comparison. Significant differences were seen in K2 for the one-way repeated measures ANOVA ($p = 0.002$) and for three pairwise comparisons: symmetrical heavy v. asymmetrical heavy ($p = 0.047$); asymmetrical light v. no tension ($p = 0.015$); and asymmetrical heavy v. no tension ($p = 0.007$). The difference in means between symmetrical light and asymmetrical light also approached significance ($p = 0.071$). A bar graph displaying K2 data across conditions is provided in figure 6.

DISCUSSION

Perturbation analysis revealed no significant difference between any pair of conditions for either percent jitter or percent shimmer. This indicates that perturbation parameters cannot reliably detect vocal fold tension asymmetry. Many of the percent jitter and percent shimmer values also had high error counts, thus rendering them invalid. This was expected as the ability of perturbation analysis to characterize acoustic signals greatly degrades with type 2 and type 3 signals (Titze, 1995; Zhang & Jiang, 2005; Jiang, 2006; Zhang, 2008), which tension asymmetry due to SLNP has been shown to produce (Titze, 1995). These results provide support for this model of SLNP as the experimentally observed invalid perturbation analysis coincides with previous theoretical observations.

Unlike perturbation parameters, the nonlinear dynamic parameters of K2 and D2 had low error values, demonstrating preserved validity when analyzing asymmetric vocal fold vibration. When calculating K2 and D2, the point of greatest convergence is used. K2 and D2 are calculated over a small range including the point of most convergence and a standard deviation is calculated as a measure of reliability. D2 and K2 measurements with standard deviations less than 0.05 are considered a valid description of the nonlinear behavior of the acoustic signal (Olszewski et al., 2011). These results were expected since SLNP creates asymmetry that introduces chaos into the system and generates type 2 or 3 aperiodic

acoustic signals. Results of nonlinear dynamic analysis showed a significant difference across conditions in K2 ($p=0.002$) but not D2 ($p=0.428$). In addition, significant differences were seen in K2 among some of the pairwise comparisons, but no significant differences in D2 were seen for any pairwise comparison. Results indicate that the nonlinear dynamic parameter of K2 can be used to differentiate between symmetric and asymmetric tension seen in SLNP. It has been previously reported that D2 increases from type 1 to type 2 to type 3 voice signals (Jiang et al., 2006). The fact that no significant difference was seen in D2 in this study suggests that the signal types generated by the various conditions were not consistently different. Though D2 was not different across conditions, significant differences in K2 warrant further investigation of nonlinear dynamic analysis as a method of evaluating tension asymmetry. Theoretically, nonlinear dynamics should have the ability to do so since more severe paralysis often leads to greater asymmetry and more chaotic behavior, which is characterized by nonlinear dynamics. However, the model used in this study lacked consistency in modeling increases in severity. Future study might focus on the ability of nonlinear dynamics to differentiate paralysis or paresis severity over a range of tension asymmetries.

Several findings merit additional consideration and yield insight into the use of nonlinear dynamic analysis to evaluate tension asymmetry. Interesting patterns were seen in the differences, or lack thereof, found between conditions (table 5). No significant difference was seen between the symmetrical tension conditions (no tension, symmetrical heavy tension, and symmetrical light tension), indicating that additional weights did not introduce chaos into the voice signal. Though a difference might be expected between the asymmetrical tension conditions (asymmetrical heavy and asymmetrical light) as additional force may exacerbate the degree of asymmetry, none was seen. This could be attributed to the fact that the weights used to simulate CT contraction were kept constant throughout all larynges when modeling the normal and unilateral paralysis states. Consistent relationships between vocal fold length and total mass provide support that the weights were effective at increasing vocal fold tension; however, due to the varying size and stiffness of larynges used, percent elongation of the vocal folds varied. For smaller larynges, the difference in weights between the heavy and light asymmetrical tension conditions may have been great enough to exacerbate the condition, while differences in elongation for larger larynges may not have been sufficient to generate significant differences in the vocal signal. If tension were varied according to percent elongation, significant differences may occur. Finally, it is interesting to note that there is a larger significant difference between the asymmetrical conditions and no tension than there is between the asymmetrical conditions and their corresponding symmetrical conditions for K2. Though there is not a significant difference between no tension and the symmetrical tension conditions, register transitions can occur with increased vocal fold tension, leading to vocal instability and chaos (Tokuda et al., 2007). The increased significance seen between the asymmetrical tension and no tension conditions is likely due to register transitions. Though preliminarily interesting and potentially valuable when forming a differential diagnosis, this observation requires validation by a more tightly controlled study of chaotic vocal fold vibration.

The SLNP model used in this study was relatively novel. Few studies have used weights to approximate CT contraction (Van den Berg & Tan, 1959; Isshiki et al., 1977). Future experiments could benefit from refinement of this technique. It was expected that the asymmetrical heavy tension condition would exacerbate the defect and increase the degree of asymmetry. As previously discussed, a significant difference was not found between the asymmetrical light tension and asymmetrical heavy tension conditions for K2, and no significant differences in D2 were seen for any pairwise conditions. Analyzing the relationship obtained using a wider range of masses and vocal fold elongations may be interesting. Using the same weight for a condition may have resulted in different signal

types generated across larynges. In some, the simulated pathological conditions may have produced type 3 voice while in others chaos did not occur, resulting in a lack of a significant difference for D2. In order to study the effectiveness of nonlinear dynamic analysis to detect differences in SLNP severity, it would be beneficial to gauge severity by percent elongation and not by the absolute weight applied. Symmetrical heavy tension and symmetrical light tension might be defined as narrow ranges of percent elongation of the vocal folds rather than a constant, predetermined weight. The asymmetrical conditions would then be modeled using the same weight as the symmetrical counterpart on only one side of the larynx. The asymmetrical condition might also be described by the difference in elongation between the two vocal folds to characterize asymmetry. Using percent elongation of the vocal folds as a measure of disorder severity rather than absolute weight may yield a better correlation with nonlinear dynamic parameters.

The excised larynx bench apparatus is a valuable tool for studying laryngeal physiology and pathophysiology. A key limitation of using ex vivo rather than in vivo larynges is the lack of neural input. However, while nerve paralyses cannot be recreated, the effects of nerve paralyses can be simulated. In this experiment, weights were used to mimic CT contraction and cause vocal fold elongation. Removal of these weights on one side produced tension asymmetry, as may be observed in unilateral SLNP. Excised larynx experiments provide a foundation for exploring new diagnostic and treatment techniques which can be developed in a repeatable and risk-free environment before being applied to patients in the clinical setting. Though acoustic analysis represents a valuable and frequently used clinical tool, it remains rather nonspecific for evaluation of vocal pathology. While we observed significant changes in the nonlinear dynamic analysis parameter of K2 in this study, such changes can also be observed with vocal fold masses, recurrent laryngeal nerve paralysis, or vocal fold scarring. This study represents a starting point towards improved evaluation of SLNP, but more specific methods of analysis are required.

Supplementing acoustic analysis with aerodynamic analysis can improve diagnostic specificity. When evaluating dysphonia, laryngoscopy can easily identify mass lesions such as polyps, nodules, or cancer. Distinguishing among voice disorders presenting with normal vocal fold morphology presents a greater challenge to the diagnostician. Differential effects on aerodynamic parameters may be useful in differentiating SLNP from other types of dysphonia with a neurological etiology. For example, while both superior laryngeal nerve paralysis and recurrent laryngeal nerve (RLN) paralysis may result in chaotic voice production, their effects on aerodynamic parameters are different. RLN paralysis results in an increased glottal gap which decreases resistance across the glottis, resulting in a greater increase in flow than pressure (Hottinger et al., 2007). It is important to note that pressure can also be increased, particularly as patients develop compensatory strategies to overcome glottic insufficiency and produce voice; however, the increase in flow is greater than the increase in pressure (Hottinger et al., 2007). SLN paralysis, however, as observed in this study, causes a greater increase in pressure than airflow. Our analysis in this paper was limited to threshold aerodynamic parameters; future studies may include additional parameters such as the airflow and subglottal pressure required to produce a given sound pressure level. Considering the difference in glottal gap present between RLNP and SLNP, one could expect that the aerodynamic power required to produce a given sound pressure level would be higher in RLNP than SLNP.

Perturbation analysis has long been a valuable method of characterizing vocal signals; however, perturbation analysis is invalid for aperiodic signals. The results of this study indicate nonlinear dynamic analysis is more appropriate for the evaluation and characterization of the type 2 or type 3 aperiodic vocal signals that are generated by the tension asymmetries typically seen in SLNP. Nonlinear dynamic analysis has demonstrated

the potential to be a valuable diagnostic tool, particularly when detecting the subtle signs of SLNP. When used in combination with visualization techniques, acoustic nonlinear dynamic analysis could provide a quantitative tool for the confirmation of an SLNP diagnosis. In addition, nonlinear dynamic analysis could provide a quantitative means of evaluating treatment effectiveness for the disorder.

CONCLUSION

SLNP was simulated in an excised larynx by creating tension asymmetry, allowing for evaluation of experimental acoustic and aerodynamic assessments. Perturbation parameters were not different between symmetric and asymmetric conditions, while the nonlinear dynamic parameter of K2 was significantly different. In addition, the perturbation parameters had large error values, rendering them invalid, while the nonlinear dynamic parameters had low error values, indicating they are more appropriate for characterizing the aperiodic voice signals generated by tension asymmetry. Supplementing nonlinear dynamic acoustic analysis with threshold aerodynamic measurements may improve diagnostic specificity. Additional improvements to the model used in this study will allow for more thorough evaluation of the range of quantitative measurements which may aid clinicians in the assessment of SLNP.

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Figure 1. A diagram of the testing set up used in the study. Depicted are the pseudolung, computer, pressure source, flow meter, micrometer, microphone and mounted larynx.

Figure 2.

A photograph of the cords used to simulate CT muscle contraction. The cords simulating the vertical belly of the CT are directed straight down with weights. The cords simulating oblique belly of the CT are draped over metal rods to redirect oblique force in the vertical direction. There is also a suture placed from the cricoid cartilage to the trachea for support.

Figure 3.

A diagram of the cords draped over metal rods to redirect oblique force in the vertical direction. Dotted lines indicate the cords falling into the page (vertical) and solid lines indicate the cords are in the plane of the page (horizontal). Also depicted are the micrometer prongs used to adduct the vocal folds to the midline.

Figure 4.

Superior view of larynx across conditions. Larynx is slightly rotated with asymmetrical light tension and more severely rotated with asymmetrical heavy tension. NT = no tension; SL = symmetrical light tension; AL = asymmetrical light tension; SH = symmetrical heavy tension; AH = asymmetrical heavy tension. Left cricothyroid muscle paralysis is simulated in AL and AH.

Figure 5.

Bar graphs depicting the means with error bars representing standard error of the mean for phonation threshold pressure (PTP). NT = no tension; SL = symmetrical light tension; AL = asymmetrical light tension; SH = symmetrical heavy tension; AH = asymmetrical heavy tension.

Figure 6.

Bar graphs depicting the means with error bars representing standard error of the mean for the nonlinear dynamic parameter, K2. NT = no tension; SL = symmetrical light tension; AL = asymmetrical light tension; SH = symmetrical heavy tension; AH = asymmetrical heavy tension.

Table 1

Glottal length (mm).

No tension	Symmetrical light	Asymmetrical light	Symmetrical heavy	Asymmetrical heavy
12.08 ± 2.40	13.13 ± 2.49	12.71 ± 2.54	13.87 ± 2.71	13.37 ± 2.55

Values are presented as mean ± standard deviation.

Table 2

Summary aerodynamic data.

Parameter	No tension	Symmetrical Light	Asymmetrical Light	Symmetrical Heavy	Asymmetrical Heavy	p-value
PTP	12.82±7.15	16.77 ± 6.80	15.01 ± 8.22	20.98 ± 9.49	16.74±8.63	<0.001
PTF	23±16	25 ± 16	24 ± 17	29 ± 18	25±16	0.396
PTW	388±452	486 ± 454	464 ± 530	748 ± 698	531±604	0.050

Values are presented as mean ± standard deviation. PTP = phonation threshold pressure (cmH₂O); PTF = phonation threshold flow (L/min); PTW = phonation threshold power (PTP*PTF; cmH₂O*L/min). P-values represent results of one-way repeated measures ANOVA.

Table 3

Average percent jitter and percent shimmer values for each condition and the p-value from the one-way repeated measures ANOVA.

Parameter	No tension	Symmetrical Light	Asymmetrical Light	Symmetrical Heavy	Asymmetrical Heavy	p-value
% jitter	0.788±0.495	0.551±0.405	0.976±1.090	0.979±1.305	1.303±1.716	0.448
% shimmer	53206±3.631	3.826±2.163	5.236±3.576	5.524±5.124	8.060±6.787	0.449

Values are presented as mean ± standard deviation.

Table 4

Average D2 and K2 values for each condition and the p-value from the one-way repeated measures ANOVA.

Parameter	No tension	Symmetrical Light	Asymmetrical Light	Symmetrical Heavy	Asymmetrical Heavy	p-value
D2	1.472±0.399	1.480±0.353	1.713±0.691	1.510±0.526	1.869±0.664	0.428
K2	0.103±0.0422	0.116±0.0364	0.159±0.0598	0.118±0.0292	0.171±0.0692	0.002

Values are presented as mean ± standard deviation.

Table 5

Pairwise comparisons.

Pair	% jitter	% shimmer	K2	D2	PTP	PTF	PTW
Sym. light/asym. light	0.195	0.390	0.071	1.000	0.054	0.546	0.592
Sym. light/no tension	0.328	0.392	0.227	0.955	<0.001	0.477	0.025
Sym. heavy/asym. heavy	0.612	0.489	0.047	0.264	0.969	0.885	1.000
Sym. heavy/no tension	0.677	0.900	0.313	0.879	0.005	0.167	0.038
Sym. heavy/sym. light	0.313	1.000	0.857	0.813	0.065	0.228	0.099
Asym. light/no tension	0.641	0.987	0.015	0.404	0.007	0.549	0.040
Asym. heavy/no tension	0.123	0.220	0.007	0.070	0.002	0.087	0.016
Asym. heavy/asym. light	0.528	0.193	0.659	0.635	0.003	0.487	0.175

P-values represent results of paired t-tests. Sym. = symmetrical tension; asym. = asymmetrical tension; K2 = second order entropy; D2 = correlation dimension; PTP = phonation threshold pressure; PTF = phonation threshold flow; PTW = phonation threshold power.