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Phonation threshold power in ex vivo laryngeal models

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Abstract

This study hypothesized that phonation threshold power is measureable and sensitive to changes in the biomechanical properties of the vocal folds. Phonation threshold power was measured in three sample populations of ten excised canine larynges treated with variable posterior glottal gap, variable bilateral vocal fold elongation, and variable vocal fold lesioning. Posterior glottal gap was varied from 0 mm to 4 mm in 0.5 mm intervals. Bilateral vocal fold elongation was varied from 0% to 20% in 5% intervals. Vocal fold lesion treatments included unilateral and bilateral vocal fold lesion groups. Each treatment was investigated independently in a sample population of ten excised canine larynges. Linear regression analysis indicated that phonation threshold power was sensitive to posterior glottal gap ($R^2=0.298$, $p<0.001$) and weakly to vocal fold elongation ($R^2=0.052$, $p=0.003$). A one-way repeated measures ANOVA indicated that phonation threshold power was sensitive to the presence of lesions ($p<0.001$). Theoretical and experimental evidence presented here suggests that phonation threshold power could be used as a broad screening parameter, sensitive to certain changes in the biomechanical properties of the larynx. It has not yet been measured in humans, but because it has the potential to represent the airflow-tissue energy transfer more completely the phonation threshold pressure or flow alone, it may be a more useful parameter than these and could be used to indicate that laryngeal health is likely abnormal.

Keywords

phonation threshold; power; energy; pressure; flow; excised larynx

INTRODUCTION

Phonation is the transduction of aerodynamic energy from the respiratory system to acoustic energy known as the voice source. Understanding phonation from an energetic standpoint is critical; because energy is a conserved quantity, the phonatory function of the larynx can be evaluated using quantitative measures based on energy measurements. The respiratory system can only output aerodynamic power to the larynx within certain energy bounds. Given this pre-existing range of possible pulmonary outputs, changes in the biomechanical properties of the vocal folds that increase the amount of energy required for a given state of phonation may be viewed as maladaptive. Energetically inefficient phonation in some cases

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is a pathological problem that may restrict vocal abilities, inhibit communication, or even reduce the quality of life.

The aerodynamic power produced during phonation is a useful quantity that is easy to determine given certain assumptions. The vocal tract is assumed to be characterized by one-dimensional flow, whereby the air flow, pressure, density, and temperature are uniform over a given cross-section of the superior-inferior axis ($\hat{z}$). During phonation the air is forced

towards positive $\hat{z}$ with some axial force $\vec{F}(t)$ through a subglottal cross-sectional area A , such that $F(t) = P_s A$, where P_s is the subglottal pressure. The work performed by this force is the line integral of force with respect to distance: $W(t) = F(t)z$. Assuming the subglottal pressure is constant and equal to the intrapulmonic pressure and the intrapulmonic pressure is not affected by glottal movements, the first time-derivative of work is the power:

$$\mathcal{P}(t) = \frac{d}{dt} [P_s A z] = P_s U \quad (1)$$

where U is the glottal flow. Thus, by quantifying both the subglottal pressure and flow, one can quantify the aerodynamic power used during phonation. This parameter has been used to directly quantify the power expenditure during phonation and has also been used as a ratio of the acoustic power to quantify the vocal efficiency (1, 2). If measured at the moment phonation begins, this parameter is the phonation threshold power ($\mathcal{P}_{th}$), the product of phonation threshold pressure and flow. To understand the sensitivities of this parameter, the sensitivities of its arguments, phonation threshold pressure and flow, must be understood.

Phonation threshold pressure (P_{th}) was first proposed by Ingo Titze and since has received much attention for its ability to reflect the biomechanical properties of the larynx (3, 4). Using a one-mass mucosal wave model to simulate the vocal folds, Titze found:

$$P_{th} = \frac{k_t B c x_0}{T} \quad (2)$$

where k_t is a transglottal pressure coefficient, B is the damping constant, c is the mucosal wave velocity, x_0 is the prephonatory glottal half-width, and T is the vocal fold thickness. Pathologies which alter these physical properties of the larynx would be expected to influence P_{th} . Indeed, P_{th} has been found in studies to be sensitive to pathologies (5, 6). Since the introduction of this parameter, many new formulations of the P_{th} equation have been made, for example to include the oscillation frequency or vocal tract inertance explicitly (7, 8). Under additional simplifying assumptions, most of these new formulations can be reduced to the original formulation as presented in Eq. 2. These models allow for relatively simple solutions to be found for P_{th} , but at the cost of using lumped parameters and coefficients. Intricate experiments or models of greater sophistication are needed to establish these model parameters.

Phonation threshold flow (U_{th}) is defined as the glottal flow at the critical condition of phonation. Jiang and Tao (9) proposed the parameter, and used a one-mass model to formulate an equation for U_{th} which was dependent explicitly on the biomechanical properties of the vocal folds:

$$U_{th} = 4L \sqrt{\frac{2Bc x_0^3}{\rho T}} \quad (3)$$

where L is the vocal fold length and ρ is the air density. Like P_{th} , U_{th} is expected to vary predictably with pathological states which elicit changes in the configuration and properties of the vocal folds. Experimentation on excised canine larynx models has shown that U_{th} is sensitive to changes in posterior glottal gap (10), vocal fold elongation (11), and surface dehydration (12). It is of particular importance that U_{th} was found to be more sensitive to changes in the posterior glottal gap than P_{th} , suggesting that in some cases, U_{th} may be a more robust indicator of the state of laryngeal health than P_{th} (10). U_{th} has also been investigated in humans, and statistically significant differences were found between the mean U_{th} in those with no known laryngeal pathology, and those with polyps or nodules (13).

Phonation threshold power ($\mathcal{P}_{th}$) is the minimum aerodynamic power required to initiate phonation. Substituting the equations for P_{th} (Eq. 2) and U_{th} (Eq. 3) into Eq. 1, $\mathcal{P}_{th}$ can be determined:

$$\mathcal{P}_{th} = P_{th} \times U_{th} = k_t L \sqrt{\frac{8B^3 c^3 \lambda_0^5}{T^3 \rho}} \quad (4)$$

This represents the rate of energy transfer whereby a steady state oscillation of the vocal folds ensues: the energy absorbed from the airflow by the vocal fold tissues equals the energy dissipated by the damping force of the tissues. This also provides a theoretical basis for the increased vocal effort required in many pathological voices, because as the critical level of power is increased by abnormal biomechanical properties, it becomes more respiratorily strenuous to phonate. In other words, pathologies which reduce the efficiency of the larynx as an energy transducer will increase the required amount of aerodynamic power provided by the respiratory system per unit of acoustic power output.

The present study investigated the sensitivity of $\mathcal{P}_{th}$ to variable physiological and pathological conditions in a sample population of thirty excised canine larynges. We hypothesized that $\mathcal{P}_{th}$ is sensitive to changes in posterior glottal gap, bilateral vocal fold elongation, and vocal fold lesioning.

MATERIALS AND METHODS

Sample population

Thirty excised canine larynges were included in the sample population. The canines were sacrificed for purposes unrelated to this study. The larynges were harvested immediately postmortem and quick-frozen in 0.9% saline solution. They were stored at -12°C until use, whereupon they were thawed slowly in cold water. Specimens were included in one of three sub-studies, investigating the effect of posterior glottal width changes ($N=10$), bilateral vocal fold elongation ($N=10$), or unilateral and bilateral vocal fold lesions ($N=10$) on $\mathcal{P}_{th}$.

The larynges were dissected in a manner similar to the procedure outlined by Jiang et al (14). Briefly, the larynges were excised by severing the trachea about 4-5 cm below the cricoid cartilage and lacerating the pharyngeal tissues superior to the thyroid cartilage. Tissues superior to the vocal folds were dissected away immediately prior to experimentation for adequate visualization, and a sufficiently small portion of the posterior thyroid cartilage was dissected away to allow micromanipulators to approach the arytenoids.

Apparatus

Each larynx was mounted on an excised larynx phonation system, as illustrated in Figure 1. The larynx was fastened to the pseudolung of the system using a metal pull clamp. Three-pronged bilateral micromanipulators were inserted into the lateral facets of the arytenoids to provide precise control of vocal fold elongation. A lateral micromanipulator was fastened by suture to the laryngeal prominence of the thyroid cartilage to provide precise control of vocal fold elongation.

This system was designed to simulate the human respiratory system. Pressurized airflow from an internal building source was routed through two Concha Therm III heater-humidifiers (Fisher & Paykel Healthcare Inc.; Laguna Hills, California). A pseudolung directly below the larynx simulated the volumetric and capacitance characteristics of the lungs. Subglottal air flow was measured using an Omega mass flowmeter (model FMA-1601A, Omega Engineering Inc.; Stamford, Connecticut) and subglottal air pressure was measured using a Heise digital pressure meter (901 series, Ashcroft Inc.; Stratford, Connecticut). The acoustic signal was recorded using a Sony microphone (model ECM-88, Sony Electronics Inc.; New York, New York) and a Symmetrix pre-amplifier (model 302, Symetrix Inc.; Mountlake Terrace, Washington). All measurements were recorded on a personal computer through a National Instruments Data Acquisition Board (model AT-MIO-16, National Instruments Corp.; Austin, Texas) and using a custom LabVIEW (National Instruments Corp., Austin, Texas) computer program. The entire excised larynx phonation system was housed in a triple-walled sound attenuated room to mitigate the confounding effects of background noise.

Aerodynamic and acoustic measurements were recorded at the threshold of phonation by increasing the airflow until phonation ensued, then decreasing it until it ceased. This procedure was repeated to record replicate phonation onset tokens.

Posterior glottal gap investigation procedure

Ten larynges were included in the variable posterior glottal gap sub-study. Each larynx was mounted on the excised larynx phonation system, and posterior glottal width was controlled as an independent variable by inserting variable-width shims between the arytenoids. These shims ranged in width from 0.0 mm to 4.0 mm, with 0.5 mm interval steps. Figure 2 illustrates the procedure used to control posterior glottal gap. Five phonation thresholds were recorded for each posterior glottal width, and 45 phonation thresholds were measured in total per larynx.

Vocal fold elongation procedure

Ten larynges were included in the variable vocal fold elongation sub-study. Each larynx was mounted on the excised larynx phonation system, and vocal fold elongation was achieved by adjusting the anterior micrometer slowly. Five phonation thresholds were recorded under five vocal fold elongation conditions: 0% (anatomically relaxed), 5%, 10%, 15%, and 20%. A total of 25 phonation thresholds were measured in total per larynx.

Vocal fold lesioning procedure

Ten larynges were included in the vocal fold lesioning sub-study. Each larynx was mounted on the excised larynx phonation system, and ten phonation thresholds were recorded under control conditions. Then, using a soldering iron heated to 850° F, a unilateral lesion was simulated by burning a small area on the medial midpoint of the vocal fold. Burns were always positioned at the vocal fold midpoint, and were approximately 3 mm in length. Ten phonation thresholds were recorded under the unilateral lesion condition. Bilateral lesions were simulated by burning the remaining fold in the same fashion. Figure 3 illustrates the

treatment used to simulate unilateral and bilateral lesions. Ten phonation thresholds were recorded under the bilateral lesions condition. For each larynx, thirty phonation thresholds were recorded in total.

Data analysis

Data were analyzed using a custom MATLAB program (R2006a, The Mathworks, Inc.; Natick, Massachusetts). The phonation threshold was determined by finding the time point in the waveform and spectrogram of the acoustic signal at which a small-amplitude oscillation began. $\mathcal{P}_{th}$ were calculated using Eqn. 4. The conversion factor used to convert from $(\frac{mL}{s} \times cm \text{ H}_2O)$ to watts was 9.807×10^{-5} . The sensitivity of $\mathcal{P}_{th}$ to the different physiological variables controlled in these experiments was investigated by performing linear regression analysis (for continuous independent variables) and one-way repeated measures ANOVA (for categorical independent variables) on the aggregate $\mathcal{P}_{th}$ data with respect to treatment group.

RESULTS

Figure 4 shows the $\mathcal{P}_{th}$ of ten excised larynges with variable posterior glottal width. A linear regression analysis yielded the following linear model:

$$\mathcal{P}_{th} = 0.231 + 0.215x_0, \quad R^2 = 0.298$$

indicating that a positive relationship exists between these two variables ($p < 0.001$). Table I presents the post-hoc pairwise comparisons using Tukey's method. Tukey's method was used because the data were heteroscedastic. Because of a possible floor effect in the posterior glottal width domain below 1.5 mm and a ceiling effect above 3.0 mm, a linear regression analysis was performed on the $\mathcal{P}_{th}$ data in the domain [1.5, 3.0] and yielded the following linear model:

$$\mathcal{P}_{th} = 0.031 + 0.251x_0, \quad R^2 = 0.198$$

indicating that within this subdomain, a positive relationship exists between the two variables ($p = 0.005$).

Figure 5 shows the $\mathcal{P}_{th}$ of ten excised larynges with variable vocal fold elongation. A linear regression analysis yielded the following linear model:

$$\mathcal{P}_{th} = 0.099 + 0.026\epsilon, \quad R^2 = 0.052$$

indicating that a weak positive relationship existed between these two variables ($p = 0.003$).

$\mathcal{P}_{th}$ of ten excised larynges before lesions, after unilateral lesioning, and after bilateral lesioning are illustrated in Figure 6. One-way repeated measures ANOVA indicated that statistically significant differences existed between the means of the treatment groups ($p < 0.001$). Post-hoc comparisons using the Holm-Sidak method indicated each pair-wise comparison was statistically significant ($p < 0.001$ for all pairwise comparisons).

DISCUSSION

The results of these experiments are in partial agreement with our hypothesis that $\mathcal{P}_{th}$ is sensitive to changes in vocal fold geometry and configuration. As expected, $\mathcal{P}_{th}$ was found to be sensitive to changes in posterior glottal width. Posterior glottal width, as seen in Eqn. 4, was expected to affect $\mathcal{P}_{th}$ more than any other variable. According to Eqn. 4, we expected $\mathcal{P}_{th} \propto x_0^{2.5}$; while our $\mathcal{P}_{th}$ data tended to increase with posterior glottal width, they did not satisfy the exponential relationship seen in Eqn. 4 (Figure 4). The incongruence between the theoretical expectations and the experimental results could be due to the methods used. During the excised larynx phonation experiment, only the positions of the arytenoids and the laryngeal prominence of the thyroid cartilage were precisely controlled. While abducting the vocal folds to increase the glottal gap, other configurational variables may have been inadvertently altered. The transglottal pressure coefficient, for example, is one variable that is predicted by Eqn. 4 to affect $\mathcal{P}_{th}$. This coefficient is sensitive to glottal geometry and viscous resistance (3). It is also uncertain how posterior glottal width may have affected the mucosal wave velocity (c). These biomechanical properties may have been altered during abduction or by dehydration: although the larynx was phonated using heated, humidified air and saline solution was applied frequently and liberally, the presence or extent of tissue dehydration in the ex vivo state is a confounding variable that all excised larynx experiments must consider.

It is interesting to note from Figure 4 the presence of floor and ceiling effects. From Table I, a floor effect exists below 1.5 mm and a ceiling effect above 3.0 mm in the posterior glottal width domain. Future research with more advanced modeling may be required to elucidate possible mechanisms for this phenomenon. The present study lacks sufficient data to adequately investigate what is occurring; however, it is reasonable to conservatively conclude from these data that phonation threshold power exhibited the greatest sensitivity to posterior glottal width in the domain 1.5 mm to 3.0 mm, and that the relationship is positive.

While $\mathcal{P}_{th}$ was found to be sensitive to the presence of vocal fold lesions, it was not found to be strongly sensitive to changes in vocal fold elongation. Vocal fold tissue lesions are characterized by morphological changes and disruption in the lamina propria that induce changes in the damping characteristics, vocal fold stiffness, and the mucosal wave velocity. As the vocal folds become more stiff and the tissue characteristics change, more energy is required to initiate phonation. This is illustrated in Figure 6, where successive lesioning treatments increased the $\mathcal{P}_{th}$. Figure 5 indicates that $\mathcal{P}_{th}$ exhibited a positive relationship with elongation, but that the relationship was weak ($R^2=0.052$). Bilateral vocal fold elongation was expected to cause a monotonic increase in $\mathcal{P}_{th}$. From Eqn. 4, $\mathcal{P}_{th}$ is linearly proportional to vocal fold length. However, as the vocal folds are elongated and assuming incompressibility of the vocal fold tissues, the volume remains constant such that

$$V = L(1+\varepsilon) \times \frac{1}{\sqrt{(1+\varepsilon)}} D \times \frac{1}{\sqrt{(1+\varepsilon)}} T \quad (5)$$

where V , L , D , T , and ε represent the vocal fold volume, length, depth, thickness, and elongation. Assuming the volume remains roughly constant, as the vocal folds are elongated, the depth and thickness decrease. From Eqn. 4 and assuming other variables remain unchanged, we see that as the thickness decreases, the $\mathcal{P}_{th}$ is expected to increase:

$\mathcal{P}_{th} \propto (1+\varepsilon)^{1.75} L/T^{1.5}$. Thus, both the changes in length and thickness would be expected to cause an increase in $\mathcal{P}_{th}$. Figure 5 illustrates the observed relationship between bilateral vocal fold elongation and $\mathcal{P}_{th}$, showing a positive relationship between $\mathcal{P}_{th}$ and elongation

that is weak ($R^2=0.052$) but whose p -value (0.003) satisfies the predefined significance level ($\alpha=0.05$). In interpreting these results, it is important to recognize that vocal fold elongation is not an isolated independent variable, and it is likely that elongation induced changes in the transglottal pressure coefficient, damping, and mucosal wave velocity. These properties are confounding variables that are difficult to measure or control, even in an *ex vivo* setup.

The characteristics and sensitivities of $\mathcal{P}_{th}$ have not been previously investigated outside of a theoretical framework. For example, while this study did not include or investigate the effect of vocal tract loading on $\mathcal{P}_{th}$, previous theoretical investigations by Jiang and Tao (9) elaborated upon the power relationships at the phonation threshold and proposed:

$$\mathcal{P}_{th} = \mathcal{P}_{tract} + \mathcal{P}_{glottis} + \mathcal{P}_{air} = R_2 U_{th}^2 + k_t \left(\frac{\rho U_{th}^3}{2 \bar{a}_2^2} \right) \quad (6)$$

where $\mathcal{P}_{tract} = R_2 U_{th}^2$ is the power expenditure by vocal tract resistance, $\mathcal{P}_{air} = \rho U_{th}^3 / 2 \bar{a}_2^2$ is the power expenditure by the glottal exit airflow, and $\mathcal{P}_{glottis} = (k_t - 1) \mathcal{P}_{air}$ is the power expenditure by glottal resistance. In this formulation, subcritical levels of power are completely consumed by airflow and the resistance of the vocal tract and glottis. However, once the critical level of power is reached, any additional power works towards initiating vocal fold oscillation. Our experimental data reflect the $\mathcal{P}_{th}$ with a vocal tract resistance of zero, which may make the presently reported values underestimates of what they would be expected to be *in vivo*.

The present study serves as a preliminary investigation of a novel parameter, phonation threshold power. From a purely scientific perspective, studying the energy relationships at the onset of phonation can lead to a better scientific understanding of phonation. $\mathcal{P}_{th}$ was predicted by previous theoretical works to be reflective of the biomechanical properties of the larynx. Because of this dependence, $\mathcal{P}_{th}$ may be an indicator of general laryngeal health. It has not yet been measured in humans, but because it has the potential to represent the airflow-tissue energy transfer more completely than U_{th} or P_{th} alone, it may be a more useful parameter and could be used to indicate that laryngeal health is likely abnormal. Eqn. 4 suggests that $\mathcal{P}_{th}$ could be used as a broad screening parameter, sensitive to certain changes in the biomechanical properties of the larynx.

Future studies are needed to explore this parameter in humans; the current study suggests that pathologies which alter the physiological state of the larynx, such as laryngitis, vocal fold masses, vocal fold lesions, or paralysis, could result in aberrant $\mathcal{P}_{th}$ values. In the case of neurological pathologies such as unilateral vocal fold paralysis, the abnormally large posterior glottal width may make laryngeal energy transduction sufficiently inefficient to induce clinically conspicuous increases in $\mathcal{P}_{th}$. However, it is important to note that it is unlikely this parameter will replace perceptual voice evaluation, stroboscopy, or a neurological examination. Much more research is needed to elucidate the nature of this new and complex parameter.

CONCLUSION

Phonation threshold power ($\mathcal{P}_{th}$), the minimum aerodynamic power required to initiate phonation, was investigated in a sample population of 30 excised canine larynges. Subpopulations were treated with variable posterior glottal width, bilateral vocal fold elongation, and vocal fold lesion conditions. $\mathcal{P}_{th}$ was measured in these larynges under control and experimental conditions. Statistically significant differences in $\mathcal{P}_{th}$ were found

under the variable posterior glottal width and vocal fold lesion treatments, and the variable vocal fold elongation treatments exhibited a weak positive influence on $\mathcal{P}_{th}$. This weakness may be due to the interaction between vocal fold elongation and other biomechanical variables. $\mathcal{P}_{th}$ is a novel parameter that is sensitive to certain changes in laryngeal configuration and properties, and may be a useful parameter in indicating the general health of the larynx as an energy transducer. More theoretical and empirical insight is required, however.

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Figure 1.

The excised larynx phonation system. Pressurized air from a building source (A) was routed through a pneumotachometer (B), two heater-humidifiers in series (C), and a pseudolung (D) before being routed through the larynx (not shown, G). Bilateral three-pronged micromanipulators (E) were inserted into the arytenoids and an anterior micromanipulator (F) was sutured to the laryngeal prominence of the larynx. The acoustic signal was acquired using a microphone (H) and a pre-amplifier (I). A manometer (K) recorded the subglottal pressure. A high-speed digital camera (J) recorded images of the position and configuration of the larynx.

Figure 2.
An excised larynx mounted on the excised larynx phonation system with an interarytenoid shim to simulate a 2.0 mm posterior glottal width.

Figure 3.

An excised larynx (a) before lesioning treatments, (b) after unilateral lesioning, and (c) after bilateral lesioning.

Figure 4.

The $\mathcal{P}_{th}$ in watts of ten excised canine larynges with variable posterior glottal width. Points indicate the mean and error bars indicate standard deviations. Linear regression analysis indicated a positive relationship exists between the two variables ($R^2=0.298$, $p<0.001$).

Figure 5.

$\mathcal{P}_{th}$ in watts of excised larynges with variable bilateral vocal fold elongation. The $\mathcal{P}_{th}$ data are plotted on a logarithmic scale for ease of visualization. The top and bottom edges of the boxes indicate the third and first interquartiles. The lines in the middle of the boxes indicate the medians. Whiskers above and below the boxes indicate the 90th and 10th percentiles. Points above and below the boxes indicate the 95th and 5th percentiles. Linear regression analysis indicated that a weak positive relation exists between the two variables ($R^2=0.052$, $p=0.003$).

Figure 6.

$\mathcal{P}_{th}$ in watts of excised larynges before vocal fold lesion treatments, after unilateral vocal fold lesioning, and after bilateral vocal fold lesioning. The top and bottom edges of the boxes indicate the third and first interquartiles. The lines in the middle of the boxes indicate the medians. Whiskers above and below the boxes indicate the 90th and 10th percentiles. Points above and below the boxes indicate the 95th and 5th percentiles. The differences between the means of each group were statistically significant ($p < 0.001$, one-way repeated measures ANOVA).

Table 1

Results (q-values) of pairwise comparisons of phonation threshold power data between posterior glottal width groups using Tukey's method. Statistically significant q-values are bolded.

		Posterior Glottal Width (mm)								
		0	0.5	1	1.5	2	2.5	3	3.5	4
Posterior glottal width (mm)	4	6.697	6.697	6.986	6.524	4.503	3.002	1.501	0.462	
	3.5	6.235	6.235	6.524	6.062	4.041	2.54	1.039		
	3	5.196	5.196	5.485	5.023	3.002	1.501			
	2.5	3.695	3.695	3.984	3.522	1.501				
	2	2.194	2.194	2.483	2.021					
Posterior glottal width (mm)	1.5	0.173	0.173	0.462						
	1	0.289	0.289							
	0.5	0								
	0									