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Effects of a simulated system of straw phonation on the complete phonatory range of excised canine larynges

Jing Kang¹, Austin Scholp², Jacob Tangney², and Jack J. Jiang^{1,2}

¹EENT Hospital of Fudan University, Department of Otolaryngology-Head and Neck Surgery, Shanghai, China; 200031

²University of Wisconsin-Madison, School of Medicine and Public Health, Department of Surgery, Division of Otolaryngology-Head and Neck Surgery, Madison, WI, 53792.

Abstract

Purpose—This study investigated the effects of straw phonation therapy on the aerodynamic and acoustic parameters of the vocal folds at different levels of elongation and subglottal pressure.

Methods—20 excised canine larynges were used in both experimental (straw phonation therapy simulation) and control conditions. Aerodynamic parameters, including phonation threshold pressure (PTP), phonation instability pressure (PIP), phonation pressure range (PPR), phonation threshold flow (PTF), phonation instability flow (PIF), phonation flow range (PFR), were obtained at different level of vocal fold elongation (0%, 10%, 20%). Acoustic parameters, including fundamental frequency (F0), jitter, shimmer, signal noise ratio (SNR) were detected at different levels of vocal fold elongation (0%, 10%, 20%) and subglottal pressure (15 cmH₂O, 20 cmH₂O, 25 cmH₂O).

Results—Significant decreases in PTP and PTF and significant increases in PIP, PIF, PPR, PFR were occurred in experimental condition at all levels of elongation when compared with control condition. However, no significant changes of acoustic parameters were obtained between conditions at all levels.

Conclusion—At different levels of vocal fold elongation, straw phonation not only lowered the onset of normal voice, but also elevated the onset of chaotic voice, indicating a better voice economy and voice control. Moreover, the improved phonatory range demonstrated straw phonation had the potential to prevent voice users who have high voice demand from voice fatigue and vocal damage.

Keywords

Excised larynx; Semi-occluded vocal tract; Straw phonation; Aerodynamics; Chaotic voice

>**Corresponding Author:** Jack J. Jiang, M.D., Ph.D., Address: 1300 University Avenue, 2745 Medical Sciences Center, Madison, WI 53706, Phone: (608) 265-9854, Fax: (608) 265-2139, jjjiang@wisc.edu.

Conflicts of Interest:

None

INTRODUCTION

Approximately 25 million people suffer from vocal disorders in the United States. This is especially prevalent for occupational voice users, such as teachers and salesmen [1]. Continuous voice overuse may lead to higher incidences of dysphonia, which could further negatively impact individual's quality of life and even social economy. Considering the daily voice requirements of these occupations, the development of a simple, accessible, and effective vocal therapy is a crucial task for laryngologist and speech pathologist.

One such therapy is the use of semi-occluded vocal tract exercises (SOVTEs). These are characterized by a reduction in the cross-sectional area of the distal end of the vocal tract through methods such as straw phonation, lip trills, tongue trills, humming, hand-over-mouth, etc.[2]. SOVTEs have been widely used in vocal exercises and vocal rehabilitation, which have been thoroughly investigated and validated for their positive effects [3]. The benefits to the voice from SOVTEs are salient for both speaking voice and singing voice. Fatigue resistance and skill acquisition can be developed through targeted vocal therapy. Decreased contact quotient, phonation threshold pressure (PTP) and phonation threshold flow (PTF) were obtained after SOVTEs, indicating reduced vocal collisions and improved fatigue resistance [4,5]. Acoustic metrics, such as jitter, shimmer, singing power ratio, and glottal-to-noise excitation ratio were also observed to improve after SOVTEs [6,7].

Because of the positive clinical experiences with SOVTEs, there has been an increasing interest in scientific explanations for the observed aerodynamic and acoustic effects. Previous studies have investigated the mechanisms of SOVTEs using theoretical modeling. Inertance, a positive type of impedance, increases during SOVTEs based on the equation $I = \rho L / A$ (I is inertance, ρ is the density of the oral and laryngeal air column, L is its length, and A is its cross-sectional area) [8]. Due to the increased inertance, a time-delayed buildup of supraglottal pressure leads to a negative supraglottal pressure during the closing phase, thus creating suction that pulls the vocal folds apart. This push-pull relationship facilitates self-sustained vibration and decreases PTP [9]. Additionally, an occluded vocal tract leads to increased subglottal pressure and decreased transglottal pressure [10]. This aerodynamic alteration, which helps keep the vocal folds slightly abducted, in turn, is expected to produce a lower contact quotient and reduce the impact stress during vibration. Furthermore, semi-occluded vocal tracts are beneficial for nonlinear source-filter coupling due to the better impedance match between the vocal tract and glottis [11]. More output acoustic power can be produced with improved nonlinear coupling because stored energy in the vocal tract is fed back to the source to increase the glottal flow energy [12].

Although all of SOVTEs may share the mechanism mentioned above, straw phonation is surmised to be one of the best ways to achieve optimum therapeutic inertance [13]. In addition, straw phonation enjoys several outstanding features such as easy accessibility, great efficiency, and controllability when compared with other SOVTEs [14].

Recent studies have validated straw phonation results in a reduced PTP and PTF [5,15], which provide valuable insights into the effort required to initiate vocal fold oscillation, however, these parameters do not offer information on the aerodynamic characteristics of

chaotic voice production and normal phonatory range. Phonation instability pressure/flow (PIP/PIF), the subglottal pressure/flow at which vocal fold vibration becomes irregular, are valuable in assessing onset of chaotic voice. Phonation pressure/flow range (PPR/PFR), the difference between PIP/PIF and PTP/PTF, are used to describe the range of subglottal pressures/flow over which normal phonation occurs, further completing a comprehensive picture of aerodynamic activity. Such studies are of significant clinical implication because the changes in aerodynamic phonatory range could be indicative of vocal health, and the observed results might be helpful in designing corrective schemes necessary to optimize voice production. No study to date has investigated the effects of straw phonation on the onset of chaotic voice and the phonatory range. Moreover, to our best knowledge, the articles regarding straw phonation acoustic outcomes all focus on a comfortable loudness and pitch. The relative extreme conditions are found to be more sensitive to evaluate vocal metrics than the conversational condition [16]. Including different levels of vocal length and subglottal pressure could provide us better understanding of the mechanism behind vocal production and voice therapy.

This study aimed to investigate if straw phonation could have a positive effect on aerodynamic parameters, including PTP, PTF, PIP, PIF, PPR, PFR, at different levels of vocal fold elongation. It also aimed to observe the effects of straw phonation on acoustic parameters at different levels of subglottal pressure and vocal length. An excised canine larynx bench apparatus was used to simulate straw phonation therapy. It is hypothesized that the onset of both normal voice and chaotic voice, as well as the range of normal phonation at different levels of vocal fold length could benefit from straw phonation. Additionally, significant differences of acoustics, including fundamental frequency (F0), jitter, shimmer, signal noise ratio (SNR), were obtained between experimental and control groups at the different levels of subglottal pressure and vocal fold length.

METHODS

Larynx Preparation

A total of 20 canine larynges were used in this study. All the larynges were obtained postmortem from canines sacrificed for purposes unrelated to this study. Canine larynges were dissected based on the method described by Jiang and Titze [17]. A thorough and careful inspection was conducted on every larynx to ensure that no vocal fold irregularities or trauma were present. Any larynx found to have damage was discarded. The larynges were then frozen in 0.9% saline solution and thawed individually upon use.

Apparatus

The human respiratory system was simulated by the excised larynx bench apparatus (Fig. 1). Pressurized subglottal airflow was passed through two humidifiers (Fisher and Paykel Healthcare, Inc., Laguna Hills, CA) in order to humidify and warm the air. An airflow meter (model FMA-1601 A; Omega Engineering, Inc., Stamford, CT) was used to measure the airflow, which could be manually controlled during the experiment using a needle valve. A digital pressure meter (PX Series; Omega Engineering, Inc., Stamford, CT) was used to measure the subglottal pressure. Airflow and pressure were recorded simultaneously at a

sampling rate of 10 kHz using a data acquisition board (USB-6229; National Instruments, Corp., Austin, TX) and a customized LabVIEW 2015 program (National Instruments, Corp., Austin, TX). Acoustic signals were collected using a microphone (model RTA-M; dbx Professional Products, USA) positioned 10 cm from the glottis. The apparatus was housed in a triple-walled, sound-attenuated room to reduce background noise and stabilize humidity levels and temperatures.

Excised Larynx Procedure

The larynx was mounted on the bench apparatus as specified by Jiang and Titze [17]. A metal hose clamp was used to secure the trachea to a pipe that was connected to the pseudolung. Bilateral three-pronged micrometer devices were used to stabilize the arytenoid cartilages and precise control the vocal fold position. A 3–0 silk suture passed through the thyroid cartilage, superior to the anterior commissure, was connected to another micrometer, allowing for precise control of vocal fold elongation.

The artificial vocal tract and straw phonation simulation were based on the methods described by Smith et al. [18] and Conroy et al. [5] (Fig. 2a). The artificial vocal tract was made of acrylonitrile butadiene styrene plastic and included three parts: a small tube (referred to as the laryngeal insert) (25 mm length, 6 mm diameter), a conical transition (25 mm length) and a larger straight pharynx tube (95mm length, 25mm diameter). The laryngeal insert acts as an adapter that allows the tract to be placed securely within the epilarynx. This design roughly resembles the configuration of the vocal tract [5]. An extension was attached above the artificial vocal tract to simulate the use of a straw (30 cm length, 6 mm inner diameter) in the experimental condition. The simulated vocal tract was inserted superior to the vocal folds to prevent any excess tissue from obstructing phonatory vibrations. The epiglottis and the rest of the tissue were then adhered around the laryngeal insert using tissue glue to prevent air leakage (Loctite 401 Instant Adhesive, Part No. 40104; Henkel Corporation, Düsseldorf, Germany). (Fig.2b)

Protocol

All the larynges were used in both experimental and control conditions. The condition order was randomized to avoid any potential compounding effects, such as dehydration or fatigue. Prior to the test, the length of vocal fold was measured. The vocal folds were kept hydrated by frequent application of 0.9% saline solution between trials. Vocal fold adduction was constant across all trials conducted with a single larynx.

Aerodynamic parameters, including PTP, PTF, PIP, PIF, PPR, PFR were acquired under different levels of vocal fold elongation (0%, 10%, 20%). Airflow was gradually increased until stable phonation started, at which time pressure and flow were recorded as PTP and PTF, respectively. Airflow was then gradually increased until to the stable harmonic frequency structure was lost and only aperiodic phonation with noise-like broadband spectra were observed on a real-time spectrogram, at which time pressure and flow were recorded as PIP and PIF [19]. Every trial was followed by five second periods of rest. Three trials were performed in either condition for each of three levels of vocal fold elongation.

Acoustic parameters, including F0, jitter, shimmer, SNR, were obtained under three levels of vocal fold elongation (0%, 10%, 20%) when controlling the subglottal pressure 20 cmH₂O, as well as three levels of subglottal pressure (15 cmH₂O, 20 cmH₂O, 25 cmH₂O) when controlling vocal fold elongation 0%. Airflow was kept on a plateau for approximately 5 seconds of phonation at each subglottal pressure. Acoustic samples were measured during this period. Trials were separated by approximately five second periods of rest. Three trials were performed in both conditions for each level of vocal fold elongation and subglottal pressure.

Data collection

PTP, PTF, PIP, PIF were determined manually for each trial using traces of pressure or flow over time. PPR was calculated by subtracting PTP from PIP, and PFR was calculated by subtracting PTF from PIF. The mean values of three trials were used for statistical analysis.

TF32 software (University of Wisconsin, Madison, WI, USA) was used to obtain F0, jitter, shimmer, and SNR from the acoustic recordings. The mean values of three trials were used for statistical analysis.

Statistical analysis

Statistical analysis of the data was carried out using SigmaPlot 12.3 (Systat, San Jose, CA). Aerodynamic and acoustic parameters were compared across conditions using a two-way analysis of variance (ANOVA) with repeated measures. To determine pairwise comparisons between elongation or subglottal pressure levels, a Tukey multiple comparison test was used as post hoc analysis in all measurements with α set at 0.05. Normality was evaluated using the Shapiro-Wilk test, and tests for equal variance were also performed.

RESULTS

Aerodynamic analysis

For PTP and PTF, significant differences were found in the main effects of condition and elongation ($P < 0.001$ for the condition and elongations in both PTP and PTF), as shown in Table 1 & Fig. 3a, b. Results from between-group comparisons demonstrated that PTP and PTF significantly decreased in the experimental conditions when compared to control at all three elongation levels ($P < 0.001$, $P < 0.001$, $P < 0.001$, $P = 0.013$, $P = 0.001$, $P < 0.001$, respectively) (Table 2). Results from within-group comparisons showed that PTP and PTF increased significantly with the vocal fold elongation in both experimental and control conditions (Table 3).

Regarding PIP and PIF, the main effects of condition were significant ($P < 0.001$, $P < 0.001$, respectively) (Table 1 & Fig. 3c, d). Results from between-group comparisons demonstrated that PIP and PIF increased significantly in experimental condition when compared to the control condition at all three levels of vocal fold elongation ($P < 0.001$ for the condition and elongations in both PIP and PIF) (Table 2).

Considering PPR and PFR, there were significant main effects of condition and elongation ($P < 0.001$, $P = 0.018$, $P < 0.001$, $P = 0.019$, respectively) (Table 1 & Fig. 3e, f). Results

from between-group comparisons showed that increased PPR and PFR were acquired in experimental condition when compared with control condition at all three levels of vocal fold elongation ($P < 0.001$ for the condition and elongations in both PPR and PFR) (Table 2). Results from within-group comparisons revealed that a significantly decreased PPR was obtained when comparing 20% vocal fold elongation to 0% elongation in the control condition ($P = 0.046$). Moreover, significantly decreased PFR was found when comparing 20% vocal fold elongation to 0% elongation in both the experimental and control conditions ($P = 0.039$ and $P = 0.029$) (Table 3).

Acoustic analysis

For F0, significant differences were detected in the main effects for elongation and subglottal pressure ($P < 0.001$, $P < 0.001$, respectively), while no significant differences between conditions were obtained. (Table 6 & 7) However, results for jitter, shimmer, SNR showed no significant main effects for the condition nor elongation and no interaction effects between condition and elongation. (Table 6) Additionally, there was no significant main effect of the condition nor subglottal pressure and no interaction effects between condition and subglottal pressure acquired on jitter, shimmer, SNR. (Table 7)

DISCUSSION

This study used an excised larynx straw phonation simulation to model the effects of straw phonation through different levels of vocal fold elongation and subglottal pressure. Aerodynamic and acoustic parameters were expected to improve in the experimental condition when compared with control condition. The aerodynamic results support our hypotheses; decreased PTP and PTF as well as increased PIP, PIF, PPR, and PFR were obtained during the experimental condition compared to control condition at all levels of elongation. However, no significant changes in acoustic parameters were detected between conditions.

Aerodynamic measurements offer objective, quantitative data reflective of general vocal health [20]. One foundational aerodynamic parameter, PTP, was first proposed by Titze and is defined as the minimum air pressure necessary to produce phonation [21]. Jiang and Tao then proposed another aerodynamic parameter, PTF, which is defined as the minimum airflow necessary to initiate vocal fold vibration [22]. PTP is a better descriptor of vocal fold viscoelasticity, whereas PTF is a better descriptor of vocal fold width [23]. Thus, PTP and PTF are irreplaceable landmarks in the aerodynamic assessment of laryngeal function [24–26]. Due to the sensitivity of vocal fold properties, such as tissue viscoelasticity, mucosal wave velocity, and prephonatory glottal area, PTP and PTF have been valuable indicators of voice economy, fatigue resistance and are widely used to distinguish normal from pathologic states in clinical studies [27,28].

Based on the mechanism mentioned before, increased inertance during straw phonation creates a push-pull interaction, which facilitates vocal vibration and further decreases PTP and PTF [29]. Our results for PTP and PTF, as well as other articles that use physical models or human subject testing, have confirmed the effects for improving PTP and PTF of straw phonation. In Conroy et al.'s study, a significant decrease in PTP and PTF occurred with a

straw configuration simulation when compared with control in excised larynges [5]. Reduced PTP was obtained after a long-duration straw phonation task in 20 healthy subjects in Mills et al.'s study [30]. PTP was also found to have decreased significantly in 26 healthy subjects after 10 minutes of straw phonation in Kang et al.'s study [15]. Results for PTP and PTF revealed that less input energy was needed to produce a desired voice when phonation through a straw. Better vocal efficiency is expected to occur during vocal fold vibration, which may further result in lower incidence rate of vocal injury for voice users.

Apart from the between-group differences, within-group differences in PTP and PTF were also obtained. PTP was previously found to increase by reducing the vocal fold thickness and increasing mucosal wave velocity, which associated with changes in vocal fold length [27]. PTP increasing with vocal fold elongation has also been validated by previous research [31,21]. However, no significant difference was observed for PTF when comparing 20% and 10% elongations in the experimental condition while a significant difference was observed when comparing 20% and 10% elongation in the control condition. This indicates that straw phonation might have the potential to slow down the trend of rising PTF with vocal fold elongation.

Nevertheless, PTP and PTF alone are not enough to quantify the voice physiology and pathology. When subglottal pressure or airflow is sufficiently high, vocal fold vibrations became irregular, and phonations are rough or harsh. This sharp transition from periodic motion to chaos was defined as PIP and PIF [32,17]. Bifurcation and chaos above the normal phonatory range are frequently observed in voices from patients with laryngeal pathologies [33]. In addition, chaotic and asymmetric vibrations could in turn lead to greater impact stress and vocal damage during vocal fold vibration [34]. Thus, sufficiently high PIP and PIF are essential factors of normal voice function, especially for occupational voice users, such as performers and salespersons.

Our present study demonstrated that straw phonation therapy could significantly increase PIP and PIF. An elevated PIP and PIF could help people avoid chaotic and irregular vibration when intense voice production is required through their occupation or from socializing. To our best knowledge, this is the first article evaluating the effects of straw phonation on PIP and PIF. The mechanism behind it is still unknown, thus future studies are warranted. A possible explanation for this might be related to the aforementioned increased inertance during straw phonation and pull-push cycle. This source-filter interaction may play a role in avoiding strong turbulence and increasing vibration synchronization of the vocal folds. Another possible explanation for this is that increased supraglottal pressure ensures vocal fold adduction and contact, resulting in more effective vibration.

Although significant differences were obtained in PIP and PIF between conditions, within-group comparison displayed no significant change of PIP and PIF at different levels of vocal fold length. These results matched those observed in earlier studies. PIP was not responsive to changes in either elongation in excised larynges in Zhang et al.'s study [35]. PIP and PIF were also not significantly dependent on elongation in excised canine larynges in Hoffman et al.'s study [32].

Similar to the voice range profile providing boundaries of intensity and frequency ranges, PPR and PFR are also important displays of vocal aerodynamics as a function of vocal properties, revealing the pressure and flow ranges of type 1 and type 2 voice signals (i.e. non-chaotic voice signals) [33]. Significant increases were obtained on PPR and PFR in the experimental condition when compared with control. One who has a greater phonatory range can better control their voice and sustain regular and periodic patterns at different levels of pitch and intensity. Furthermore, PPR and PFR were found the greatest at resting vocal fold length (0% elongation) and decreased as the vocal folds were elongated in both conditions. However, the rising tendency of PPR did not reach significance in the experimental condition, indicating that straw phonation may reduce the deterioration of PPR.

Although our study only measured the aerodynamic improvements when phonation through a straw, several studies provided evidence that the effects of straw phonation could be sustained for a period of time after straw phonation. The computerized tomography results in Guzman et al.'s studies demonstrated that the participant could maintain increased vocal tract inertance by adjusting laryngeal configuration, such as narrowed epilaryngeal tube and wide oropharyngeal cavity, even after completing straw phonation exercises [36,37]. Additionally, the lingering effects of straw phonation on PTP has been demonstrated by Kang et al. [38]. Therefore, the aerodynamic improvements seen in this study are also expected to be sustained for a period of time following straw phonation exercise, resulting in continuous voice comfort and phonation efficiency.

There are two hypotheses concerning straw phonation and its effect on acoustics. The first is that improved aerodynamic parameters led to less vocal fold collision, which was expected to result in better vocal fold vibration and acoustic output [11]. In addition, straw phonation, acting as a vocal warm up, facilitated laryngeal blood circulation, which may give rise to more elastic vocal folds and reduced vocal perturbation [39]. However, contrary to our expectations, this study did not acquire significant differences between conditions in acoustic parameters. This discrepancy may partly be attributed to the excised setup, which failed to reflect the effects of blood circulation variation. More importantly, although vocal fold length and subglottal pressure were increased, the larynges used were healthy and undamaged. This may indicate that acoustic parameters are not adequately sensitive to reflect the subtle conformational changes in a healthy state, while the acoustics may show improvement in pathological voice. No significant changes in acoustic parameters were observed after straw phonation in healthy participants in both of Kang et al.'s studies,[40,38] while reduced roughness, breathiness, and noise measurements as well as increased glottal to noise excitation ratio were observed after straw phonation in 27 dysphonic children [41]. The within-group comparison revealed that F0 was dependent on the vocal fold elongation and subglottal pressure. Increased F0 was found with the vocal fold elongation and greater subglottal pressure. These findings were in accordance with clinical results and animal models [42–44].

A limitation of this study was that vibration patterns of the vocal folds during straw phonation over the phonatory range were still unclear. Possible solutions to this would be to add electroglottography and combine flexible endoscopy with high-speed digital imaging. Electroglottography is an indirect measure of vocal fold movement dependent on the cutoff

threshold used, thus endoscopic visualization would be preferred [45]. The technical hurdles of vocal fold visualization during straw phonation still need to be addressed.

CONCLUSION

The beneficial effects of straw phonation therapy were investigated in a simulation model using an excised bench apparatus which provided the possibility to monitor the complete phonatory range through different levels of vocal fold elongation and subglottal pressures. This study confirmed the benefits to aerodynamic parameters from straw phonation therapy including decreased PTP and PTF as well as increased PIP, PIF, PPR, and PFR. While no significant changes were obtained in acoustic parameters, this work indicated that straw phonation has the potential to improve vocal economy and reduce the risk of vocal fatigue and damage for voice users who have high vocal demands.

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Fig. 1.
Schematic of experimental excised larynx bench apparatus

Fig. 2. Straw phonation simulation setup **a** Schematic of artificial vocal tract configuration with extension including the laryngeal insert (25 mm length, 6 mm diameter), a conical transition (25 mm length), a larger straight pharynx tube (95mm length, 25mm diameter) and the extension (30 cm length, 6 mm inner diameter) **b** An enlarged view of the excised larynx setup

Fig. 3.
Variation of aerodynamic parameters in experimental condition and control condition

Table 1

Results from two-way ANOVA with repeated measures for aerodynamic parameters.

Source of Variation	PTP		PTF		PIP		PIF		PPR		PFR	
	F	p	F	p	F	p	F	p	F	p	F	p
Condition	139.396	<0.001*	38.324	<0.001*	129.408	<0.001*	50.876	<0.001*	169.176	<0.001*	78.371	<0.001*
Elongation	59.063	<0.001*	22.873	<0.001*	2.864	0.069	0.641	0.532	4.451	0.018*	4.414	0.019*
Condition × Elongation	2.987	0.062	3.614	0.037*	0.0919	0.912	1.777	0.183	0.107	0.899	0.133	0.876

Note: comparisons with significant results are denoted with an asterisk

Table 2
Between-group comparisons of aerodynamic parameters between different elongation levels.

Dependent Variables	Elongation Levels (%)	Experimental Condition	Control Condition	P
PTP (cmH ₂ O)	0	7.67 ± 2.22	8.87 ± 2.39	<0.001*
	10	9.24 ± 2.60	10.42 ± 2.72	<0.001*
	20	10.42 ± 2.35	12.12 ± 2.71	<0.001*
PTF (mL/s)	0	139.03 ± 33.13	165.82 ± 45.73	0.013*
	10	176.35 ± 51.02	211.77 ± 51.89	0.001*
	20	191.87 ± 50.15	254.27 ± 75.97	<0.001*
PIP (cmH ₂ O)	0	53.91 ± 8.21	41.57 ± 4.68	<0.001*
	10	54.41 ± 7.91	41.68 ± 6.07	<0.001*
	20	55.19 ± 8.52	42.89 ± 5.56	<0.001*
PIF (mL/s)	0	732.02 ± 156.948	604.96 ± 143.23	<0.001*
	10	752.50 ± 183.38	623.86 ± 140.323	<0.001*
	20	732.70 ± 168.08	638.85 ± 160.86	<0.001*
PPR (cmH ₂ O)	0	46.24 ± 8.81	32.70 ± 5.66	<0.001*
	10	45.17 ± 9.07	31.26 ± 6.68	<0.001*
	20	44.77 ± 9.31	30.75 ± 6.65	<0.001*
PFR (mL/s)	0	593.00 ± 158.82	439.14 ± 131.33	<0.001*
	10	576.15 ± 181.08	412.09 ± 113.93	<0.001*
	20	540.83 ± 151.99	384.59 ± 133.41	<0.001*

Note: comparisons with significant results are denoted with an asterisk

Table 3

Within-group comparisons of aerodynamic parameters between different elongation levels.

Dependent Variables	Elongation Levels (%)	Experimental Condition		Control Condition	
		Diff of Means	P	Diff of Means	P
PTP (cmH ₂ O)	20 vs. 0	2.751	<0.001*	3.268	<0.001*
	20 vs. 10	1.173	0.001*	1.717	<0.001*
PTF (mL/s)	10 vs. 0	1.578	<0.001*	1.55	<0.001*
	20 vs. 0	52.843	<0.001*	88.442	<0.001*
	20 vs. 10	15.519	0.437	42.493	0.004*
PIP (cmH ₂ O)	10 vs. 0	37.324	0.012*	45.95	0.002*
	20 vs. 0	1.276	0.249	1.316	0.229
	20 vs. 10	0.777	0.593	1.209	0.287
	10 vs. 0	0.5	0.805	0.107	0.99
PIF (mL/s)	20 vs. 0	0.681	1.000	33.887	0.269
	20 vs. 10	19.796	0.633	14.987	0.769
	10 vs. 0	20.477	0.614	18.9	0.659
PPR (cmH ₂ O)	20 vs. 0	1.474	0.165	1.951	0.046*
	20 vs. 10	0.396	0.875	0.508	0.803
PFR (mL/s)	10 vs. 0	1.078	0.376	1.443	0.178
	20 vs. 0	52.162	0.039*	54.555	0.029*
	20 vs. 10	35.314	0.215	27.506	0.389
	10 vs. 0	16.848	0.699	27.049	0.401

Note: comparisons with significant results are denoted with an asterisk

Table 4

Summary of acoustic data collected for experimental condition

Source of Variation	Experimental Condition			F0
	Jitter	Shimmer	SNR	
15cmH ₂ O + 0% Elongation	0.62 ± 0.37	3.96 ± 1.76	16.42 ± 5.07	192.07 ± 29.94
20cmH ₂ O + 0% Elongation	0.69 ± 0.38	4.12 ± 1.97	16.26 ± 5.48	207.91 ± 32.67
25cmH ₂ O + 0% Elongation	0.75 ± 0.61	4.41 ± 2.05	15.73 ± 4.58	226.74 ± 35.13
20cmH ₂ O + 10% Elongation	0.77 ± 0.50	4.28 ± 2.00	15.62 ± 4.75	262.24 ± 53.86
20cmH ₂ O + 20% Elongation	0.71 ± 0.39	3.97 ± 1.69	15.77 ± 4.50	333.04 ± 87.19

Table 5

Summary of acoustic data collected for control condition

Source of Variation	Control Condition			
	Jitter	Shimmer	SNR	F0
15cmH ₂ O + 0% Elongation	0.70 ± 0.48	4.19 ± 1.89	16.44 ± 5.08	192.70 ± 32.12
20cmH ₂ O + 0% Elongation	0.62 ± 0.33	4.64 ± 3.25	16.64 ± 5.64	209.96 ± 32.87
25cmH ₂ O + 0% Elongation	0.73 ± 0.60	4.63 ± 2.63	15.84 ± 4.42	224.80 ± 36.34
20cmH ₂ O + 10% Elongation	0.79 ± 0.59	4.13 ± 2.03	16.26 ± 4.17	257.31 ± 49.01
20cmH ₂ O + 20% Elongation	0.78 ± 0.45	4.30 ± 1.62	16.39 ± 4.55	326.57 ± 85.40

Table 6

Results from two-way ANOVA with repeated measures for acoustic parameters.

Source of Variation	Jitter		Shimmer		SNR		F0	
	F	p	F	p	F	p	F	p
Condition	0.029	0.867	0.879	0.36	1.447	0.244	3.855	0.064
Elongation	1.019	0.371	0.296	0.745	0.18	0.836	33.577	<0.001*
Condition × Elongation	0.61	0.548	0.662	0.522	0.0756	0.927	0.121	0.887

Note: comparisons with significant results are denoted with an asterisk

Table 7

Results from two-way ANOVA with repeated measures for acoustic parameters.

Source of Variation	Jitter		Shimmer		SNR		F0	
	F	p	F	p	F	p	F	p
Condition	0.0268	0.872	1.269	0.274	0.124	0.728	0.0252	0.876
Subglottal Pressure	0.562	0.575	1.543	0.227	0.621	0.543	71.859	<0.001*
Condition × Elongation	1.497	0.237	0.327	0.723	0.273	0.762	1.808	0.178

Note: comparisons with significant results are denoted with an asterisk