Impact of Cricothyroid Muscle Contraction on Vocal Fold Vibration: Experimental Study with High-Speed Videoendoscopy

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Summary: Objectives. The aim of this study was to evaluate the effects of cricothyroid muscle contraction on vocal fold vibration, as evaluated with high-speed videoendoscopy, and to identify one or more aspects of vocal fold vibration that could be used as an irrefutable indicator of unilateral cricothyroid muscle paralysis.

Study Design. This was an experimental study employing excised human larynges.

Methods. Twenty freshly excised human larynges were evaluated during artificially produced vibration. Each larynx was assessed in three situations: bilateral cricothyroid muscle contraction, unilateral cricothyroid muscle contraction, and no contraction of either cricothyroid muscle. The following parameters were evaluated by high-speed videoendoscopy: fundamental frequency, periodicity, amplitude of vocal fold vibration, and phase symmetry between the vocal folds.

Results. Although neither unilateral nor bilateral cricothyroid muscle contraction altered the periodicity of vibration or the occurrence of phase asymmetry, there was a significant decrease in fundamental frequency in parallel with decreasing longitudinal tension. We also found an increase in vibration amplitude of right and left vocal folds, which were similar in terms of their behavior for this parameter in the various situations studied.

Conclusion. Our results suggest that differences in vibration amplitude and phase symmetry between vocal folds are not reliable indicators of unilateral cricothyroid muscle paralysis.

Key Words: Cricothyroid muscle–Superior laryngeal nerve paralysis–Excised larynx–Vocal fold vibration–High-speed imaging.

INTRODUCTION

Cricothyroid muscle paralysis occurs as a result of injury or dysfunction of the external branch of superior laryngeal nerve. From a clinical point of view, it is relatively easy to suspect cricothyroid muscle paralysis from clinical history of affected patients, who invariably complain of difficulty in producing higher registers, either suddenly or after an episode of trauma (surgical or not) in the cervical region. Such paralysis can also manifest as vocal fatigue, hoarseness, and volume changes.[1,2]

The great challenge for laryngologists dealing with cricothyroid muscle paralysis is to diagnose impaired cricothyroid muscle function through laryngoscopy. Various studies in the literature have sought to identify findings that could be used as reliable indicators of cricothyroid muscle paralysis. Unfortunately, there is no consensus among such studies. Among changes in the glottal configuration, a shortened vocal fold on the paralyzed side has been observed in a canine model,[3] as well as during phonation in human patients.[4–6] However, other authors have reported no such changes.[7–9] There have also been reports of rotation of posterior commissure on the paralyzed side during electrical stimulation in dogs,[4,10,11] as well as during phonation in human patients.[4,5,11,12] Again, that finding was not corroborated in other studies.[1,2,6–8,13,14] In addition, a difference in vocal fold height has been observed in unilateral cricothyroid muscle paralysis, in dogs[10] as well as in human patients,[1,2,6] whereas other studies have found no such difference.[7,8,13] Roy et al[15] noted epiglottic petiole deviation on the paralyzed side during phonation. It has also been demonstrated that there is a vocal fold lag on the paralyzed side during speech.[1,2] Some changes in vibration have been observed in unilateral cricothyroid muscle paralysis, such as phase asymmetry that has been reported in dogs,[3,16,17] pigs,[18] and humans.[1,16,17] A decrease in vibration amplitude on the paralyzed side has been observed in patients,[1,14] as has vibratory aperiodicity in dogs[9,16] and humans.[16]

We assume that, in unilateral cricothyroid muscle paralysis, there would be a difference between the two vocal folds in terms of longitudinal tension and, consequently, asymmetry in vibration pattern during cricothyroid muscle activation. The aim of this study was to identify one or more aspects of vocal fold vibration that could be used as an irrefutable indicator of unilateral cricothyroid muscle paralysis. To that end, we used high-speed videoendoscopy (HSV) to evaluate excised human larynges simulated to cricothyroid muscle contraction and induced to artificial vocal fold vibration.

METHODS

The procedures were approved by the Research Ethics Committee of the University of São Paulo School of Medicine (Protocol no. 296/11). This was an experimental study employing 20 larynges from adult male human cadavers, excised at a maximum of 12 hours after death. Supraglottic structures, such as ventricular bands, aryepiglottic folds, and epiglottis, were

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removed for better visualization of the vocal folds. All larynges were submerged in 0.9% saline to hydrate the vocal folds and promote better vibration.[19]

A 3-0 nylon suture placed between the two vocal processes was carried out, involving the arytenoid cartilages to promote posterior glottal closure. Each larynx was mounted onto a suitable support, affixed at the level of the cricoid cartilage. A 5-mm piece of 2-0 nylon suture was positioned laterally to the vocal fold, for later use as reference in making measurements.

To simulate cricothyroid muscle contraction, one 2-0 nylon suture was placed on each side, parallel to the division between *pars recta* and *pars oblique* of the cricothyroid muscle. One end was secured to the midpoint of the cricothyroid muscle attachment to the thyroid cartilage, and the other end to the midpoint of attachment to the cricoid cartilage. The tension applied to each suture was sufficient to achieve maximum shortening of the cricothyroid space without disrupting any of the anatomic relationships. This technique was previously described by Woodson et al.[7] Three conditions were evaluated: bilateral cricothyroid muscle contraction (both sutures remaining in place to simulate normal physiological behavior); unilateral cricothyroid muscle contraction (one of the sutures having been removed to simulate unilateral paralysis); and no cricothyroid muscle contraction (both sutures having been removed to simulate the absence of longitudinal tension).

Artificial vocal fold vibration was obtained by placing a ventilation tube with an inflated balloon into the trachea, with the distal end of the tube at the cricotracheal junction. This tube was connected to a system of compressed air passing through a container in which it was humidified and heated between 36°C and 37°C. A continuous flow of air was maintained at 5–10 L/min, controlled by a calibrated flow meter. For each larynx, the flow volume corresponded to the lowest level sufficient to induce appropriate vocal fold vibration and was maintained at a constant level during all tests.

The following parameters were evaluated with HSV: fundamental frequency (F_0), periodicity, amplitude of the vocal fold vibration, and phase symmetry. We obtained images of vocal fold vibrations using a high-speed camera system (HRES ENDOCAM 5562; Richard Wolf GmbH, Knittlingen, Germany), including a rigid laryngoscope with a 90° angle of view (HRES 8454.002; Richard Wolf GmbH) and a 300-W xenon light source (AUTO LP 5132; Richard Wolf GmbH). Images were obtained at a rate of 4000 frames per second, with a spatial resolution of 256×256 pixels and recorded in a computer coupled to the HSV system.

During the recordings, the laryngoscope was held in parallel with the vocal folds, at 10 cm from the glottis, mounted on a tripod to prevent any change in angle or distance.

Data collection

F_0 was determined with the HRES ENDOCAM program, version 1.10 (Richard Wolf GmbH). After image segmentation had been performed, we used the fast Fourier transform algorithm, which transforms a signal that is in the time domain into a representation of that signal in the frequency domain, expressed in hertz. On the basis of the F_0 values, we calculated the difference between two frequencies (each one corresponding a different studied condition—F_1 and F_2) in semitones (*st*), using the following formula:

$$st = (12 \times [\log_{10}(F_2) - \log_{10}(F_1)]) / \log_{10}(2).$$

Calculations were made in *Microsoft Excel 2016*.

We measured the amplitude of vocal fold vibration during playback of the recordings in the Laryn**X**-**C**omputer **A**ided **D**iagnosis (X-CAD) algorithm, version 1.0.0.0 (Medical Engineering Research Group, Brazilian National Council for Scientific and Technological Development), which allows the use of a reference measure (such as the 5-mm piece of 2-0 nylon suture used in the present study), thus increasing the measurement accuracy, as well as allowing different situations to be compared within the same larynx (Figure 1). Five consecutive images showing the maximum lateral excursion of each vocal fold were selected, and measurements are shown in millimeters.

Phase symmetry was defined as the condition in which both vocal folds reached maximum glottal opening at the same stage of the glottal cycle.[20] Using the HRES ENDOCAM program, we reviewed the HSV images, frame-by-frame, in playback mode. In playback mode, each frame has a duration of 0.25 ms. Therefore, we considered only those disparities with a duration >0.25 ms.

Periodicity of the mucosal wave was classified as regular (quasi-periodic vibration) or irregular (aperiodic vibration). It is known that the vocal folds do not oscillate in a perfectly periodic pattern. Thus, normal vibration is said to be quasi-periodic when the difference from one cycle to the next is sufficiently regular and similar. For that evaluation, we used digital kymography images obtained by the HRES ENDOCAM program. For digital kymography, we used the full video from each experiment. The line for obtaining the kymogram was placed in the middle third of the vocal folds, perpendicular to the anteroposterior axis of the glottis (Figure 2).

The data were statistically analyzed with generalized estimating equations followed by Bonferroni multiple comparisons. The tests were performed with a significance level of 5% ($P < 0.05$).

RESULTS

F_0

Under all conditions evaluated, the right and left vocal folds vibrated at the same frequency. There were statistically significant differences among the conditions for F_0, whether evaluated in hertz (Tables 1 and 2) or in terms of the difference in semitones (Tables 3 and 4). We found that the mean F_0 was lower during unilateral contraction of the cricothyroid muscle than during bilateral contraction, as well as being lower during no contraction than during unilateral and bilateral contraction ($P < 0.001$ for all).

Periodicity

For all studied larynges, the periodicity of the mucosal wave was classified as regular under all conditions evaluated.