QUANTITATIVE MEASUREMENTS OF THE MEDIAL SURFACE OF THE VOCAL FOLDS IN AN IN VIVO CANINE MODEL

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ABSTRACT
Medial surface dynamics of the vocal folds are important for an understanding of sound generation within the larynx. Such dynamics illustrate the propagation of the mucosal wave which makes voice production possible. Almost universally, in vivo investigations of vocal fold dynamics are performed using endoscopic techniques which allow only a superior view of vocal fold vibration. Hence, the view onto the medial margin is blocked which leads to an observation leak within the vocal fold cycle. In this study, an excised hemi-larynx set-up, is adapted for use with an in vivo canine model. In vivo models allow us to examine effects of intrinsic muscle contraction on the medial surface dynamics of the vocal folds, and provide greater insight into mechanisms of vocal control. Dynamics, of the vocal folds with varying levels of stimulation to the recurrent laryngeal nerve (RLN) will be shown. In particular, quantitative measurements such as fundamental frequency, vocal intensity, and the dynamics of up to 15 different positions along the medial surface of the vocal folds will be presented as a function of RLN innervation. To capture the vibrations, the vocal folds were imaged with a sampling frequency of 2000 Hz, and a spatial resolution of 1024 x 1024 pixels

INTRODUCTION
Medial surface dynamics of the vocal folds are important for an understanding of sound generation within the larynx. Such dynamics illustrate the propagation of the mucosal wave which makes voice production possible (Titze 1984). Almost universally, in vivo investigations of vocal fold dynamics are performed using endoscopic techniques which allow only a superior view of vocal fold vibration (Doellinger et al., 2002). Hence, the view onto the medial margin is blocked which leads to an observation leak within the vocal fold cycle. To overcome these difficulties, an excised hemi-larynx set-up was adapted for use with an in vivo canine model. The subject of the investigations was the variation in dynamics as a function of stimulation to the recurrent laryngeal nerve (RLN).

METHOD
The experimental set-up in this study is based upon studies using an excised larynx methodology (Berry et al., 2001). Within this study, a canine model (mongrel, male, 25 kg) was used. First, the creation of an in vivo hemi-larynx requires the opening of the neck to excavate the larynx. Then, the trachea was cut below the larynx, to enable the positioning of the glass plate and the prism. The in vivo model was artificially respirated. To expose the medial surface of one vocal fold, the other one had to be removed, Fig. 1. To track the movements during vibration, microsutures were placed on the remaining vocal fold by an experienced...
phonosurgeon. They were positioned to penetrate only the outer layer of the vocal fold tissue (mucosal epithelium) and therefore did not disturb the natural dynamics. To capture the dynamics of the entire medial surface, 5 vertical rows with 5 sutures per row were placed on the medial surface of the vocal fold (Fig. 2). The positions of the sutures were computed, using a semi-automatic algorithm. The trachea was mounted over a stainless steel cylindrical tube. A wedge was mounted within the tube, to smoothly channel the airflow beneath the vocal fold. A glass plate was attached at the top of the tube (Fig. 1). To prevent air leaks, vacuum grease and gauze were applied between the anterior and posterior parts of the larynx and the glass plate. To modify the degree of vocal fold adduction, the recurrent laryngeal nerve (RLN) was stimulated. Former studies showed, that increasing stimulation yields to more adduction, i.e. decrease in glottal area (Bielałowicz et al., 1999). The superior laryngeal nerve (SLN) was stimulated at a constant voltage.

Schematic View from the Top

![Schematic View from the Top](image)

Figure 1. Detailed schematic view from the top of the experimental set-up.

For later calibration and computation of the physical coordinates of the microsutures, a brass cube (5mm$^3$) was glued to the glass plate above the superior part of the vocal fold, Fig. 2. A right angle prism was placed on the other side of the glass plate, see Fig. 1. This served to simulate two different camera views, which was necessary for the computation of three-dimensional vocal fold movements (Berry et al., 2001). The requirements of the present experimental set-up suggested a linear approximation (Wood & Marshall, 1986) instead of the Direct Linear Transform (Berry et al., 2001) for computing the three dimensional coordinates, which yielded improved accuracy. The linearization error was between 7.1% and 12.5%.

Vibrations of the vocal fold were induced by passing airflow up through the trachea and through the area between the glass plate and the vocal fold (i.e. hemi-glottis). The vibrations were
recorded with a digital high-speed camera (Fastcam-Ultima APX, Photron Unlimited, Inc.) with a 200mm lens at a frame rate of 2000Hz and spatial resolution of 1024x1024 pixels. Three 150 Watt lamps served as light sources. The emitted acoustical signal was simultaneously recorded with a condenser microphone and digitized directly at 44.1kHz.

![Image](image.jpg)

**Figure 2.** Split view through the digital high speed camera. The numeration of the 25 mounted sutures is shown. At the superior part of both views, the calibration cube can be seen. On the lower left side, gauze is noticeable.

**RESULTS**

Four different recordings (E1-E4) with varying RLN stimulation were investigated. The different settings and the number of investigated sutures are shown in Tab. 1. Due to the stimulation of the RLN and the necessity of closing the anterior and posterior leaks with gauze, certain sutures were concealed during most of the time within one vibration cycle, Fig. 2. The extracted sutures are specified in Tab. 1.

Quantitative results of vocal fold dynamics were reported using Empirical Eigenfunctions (EEF) which are a powerful tool for investigating mechanisms of regular and irregular vibration (Berry et al., 1994). Conceptually, they may be viewed as the basic building blocks of many simple and complex vibration patterns (Berry et al., 1994). In previous computational studies, just a few EEFs created a variety of periodic and aperiodic vocal fold vibrations, where the principal difference between such vibration patterns was the entrainment or lack of entrainment of the
spatial EEFs (Berry et al., 1994; Alipour et al., 2000). Studies using an excised canine larynx yielded similar results for vibrations along the coronal axis (Berry et al., 2001). Within Tab. 2, the percentages of the three largest EEFs are shown along with the corresponding fundamental frequencies. In Fig. 3, the directions of the two largest EEFs and the corresponding deflection of the sutures for E1 – E4 along the middle vertical column can be seen.

Table 1. The experimental settings, the fundamental frequency and the stimulation level of the RLN are shown.

<table>
<thead>
<tr>
<th></th>
<th>E1</th>
<th>E2</th>
<th>E3</th>
<th>E4</th>
</tr>
</thead>
<tbody>
<tr>
<td>RLN [mA]</td>
<td>0.04</td>
<td>0.05</td>
<td>0.07</td>
<td>0.08</td>
</tr>
<tr>
<td>Flow [ml/s]</td>
<td>675</td>
<td>675</td>
<td>650</td>
<td>660</td>
</tr>
<tr>
<td>Sub. P. [cmH2O]</td>
<td>16</td>
<td>22</td>
<td>21</td>
<td>16</td>
</tr>
<tr>
<td>F0 [Hz]</td>
<td>182</td>
<td>120</td>
<td>218</td>
<td>202</td>
</tr>
<tr>
<td>Sutures</td>
<td>2-5,7-10,12-15</td>
<td>1-15</td>
<td>1-15</td>
<td>2-5,7-10,12-15</td>
</tr>
</tbody>
</table>

Table 2. The percentage part of the three largest EEFs within the entire dynamics as well as their fundamental frequencies are shown.

<table>
<thead>
<tr>
<th></th>
<th>E1</th>
<th>E2</th>
<th>E3</th>
<th>E4</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. EEF [%] – Hz</td>
<td>60.2 – 180</td>
<td>55.0 – 60</td>
<td>69.5 – 220</td>
<td>73.8 – 200</td>
</tr>
<tr>
<td>2. EEF [%] - Hz</td>
<td>33.1 – 180</td>
<td>28.3 – 120</td>
<td>23.6 – 220</td>
<td>22.0 – 200</td>
</tr>
<tr>
<td>3. EEF [%] - Hz</td>
<td>2.6 – 70</td>
<td>6.3 – 60</td>
<td>1.6 – 440</td>
<td>1.1 – 410</td>
</tr>
<tr>
<td>1.-3. EEF [%]</td>
<td>95.9</td>
<td>89.6</td>
<td>94.7</td>
<td>96.9</td>
</tr>
</tbody>
</table>

DISCUSSION

Despite E2, where the hemi-larynx didn’t phonate properly, some conclusions can be made. First, the fundamental frequency tended to increase as a function of RLN stimulation, Tab. 1. As in excised hemi-larynx experiments or in theoretical computer models (Berry et al., 1994, Berry et al., 2001), almost 97% (except of E2) of the vocal fold dynamics were captured by a few EEFs, Tab. 2. Also, the fundamental frequency of the vocal folds is reflected in the first two EEFs (except E2). As observed in other models, the movement of the first EEF generated an alternating convergent/divergent shaping (lower and upper parts of the folds moved were 180 degrees out of phase), and the second EEF primarily captured in-phase lateral movements. However, maintaining constant experimental conditions within in vivo experiments was crucial.
Figure 3. The directions and the elongations produced by the EEFs are shown at the middle coronal suture line for sutures 3,8,13 (see Fig. 2). The arrows determine the directions. The solid line represents the mean value, the dotted line the maximal elongation, and the dashed line the minimal elongation. Zero at the lateral axis indicates the glottal midline.

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REFERENCES
