INTRODUCTION

Vocal fold injection (VFI) is a minimally invasive procedure that is used to deliver drugs or materials to the vocal folds through a needle. The procedure has been used for over 100 years to treat vocal fold paralysis.\(^1\) In the past decade, it has been re-emerged and highlighted in laryngology with the advance of endoscopic technology and the development of new injection materials.\(^2\) Currently, VFI has rapidly replaced laryngeal framework surgery in the management of vocal fold paralysis and has expanded its indications to include various laryngeal diseases, such as spasmodic dysphonia, vocal fold atrophy, vocal fold scarring, vocal fold papillomatosis, vocal fold nodules, vocal fold polyps, vocal fold granuloma, laryngeal stenosis, and Reinke's edema.\(^2\)–\(^4\)

Several approaches to VFI have been reported, including the transoral and transcutaneous (cricothyroid [CT] membrane, thyrohyoid membrane, and thyroid cartilage) routes.\(^5\) Among these methods, the transoral and CT approaches are the most popular in current practice. Each approach has its advantages and disadvantages. The choice of approach usually depends on the surgeon’s preference, the injection material, and the condition of the patient.\(^5\) The transoral approach is straightforward and easy to learn because the needle tip is clearly visualized under endoscopy. However, it has several weaknesses. The long, curved needle is tricky to handle; and the approach can cause bleeding, laryngospasm, and injectate spillage due to mucosal penetration. Furthermore, some

Objective: The transcricothyroid (CT) membrane approach is a good option for office-based vocal fold injection (VFI). However, because the needle tip is invisible during injection using the CT approach, precise localization requires a high level of experience, and mastering this approach involves a steep learning curve. To overcome current limitations, we conceptualized a novel technique: real-time light-guided VFI (RL-VFI), which enables simultaneous VFI under direct visualization of the lighted needle tip. Herein, we aimed to verify the feasibility of RL-VFI in cadaveric canine model, simulating the setting of office-based VFI, as well as to explore its clinical usefulness.

Study Design: Animal study.

Methods: A customized prototype device was developed. It consisted of three parts: light source, controller, and injector. Light source comprised laser diodes of two wavelengths (635 nanometers [nm], red; 532 nm, green). Four types of injector were developed using 40-mm needles of 23- and 25-gauge and optic fibers of 50 and 100 μm. An ex vivo canine larynx was prepared for the experiment. Flexible laryngoscopy system was used to examine canine vocal folds.

Results: Various routes from three insertion points (3 mm, 10 mm, and 17 mm from the midline) were validated using the device. Regardless of the injection routes, the location of the needle tip was accurately indicated by light. RL-VFI was feasible under light guidance without difficulties. Moreover, precise and simultaneous re-injection could be performed at the intended point using the device.

Conclusion: We introduced RL-VFI using our customized prototype device in an ex vivo canine larynx, simulating the setting of office-based VFI. Clinical application of RL-VFI will improve safety and precision of CT approach, as well as expand its applications in laryngology.

Key Words: Vocal fold, injection, vocal fold palsy, larynx, transcutaneous, light-guided, real-time, device.

Level of Evidence: NA

Laryngoscope, 129:935–942, 2019

Real-Time Light-Guided Vocal Fold Injection: Ex Vivo Feasibility Study in a Canine Model

Wonjae Cha, MD, PhD; Jung Hoon Ro, PhD; Sun Cheol Yang, PhD; Chang Jun Choi, BSc; Inchul Yang, MS; Hochul Kang, MS; Il-Young Cho, MD; Min-gyu Jo, MD; Hyoseok Seo, MD
Laryngologists may have difficulty performing this procedure in patients with a severe gag reflex and in those with anatomical variations of supraglottal structures such as the epiglottis, arytenoid, and false vocal folds.

Seifert first suggested the CT approach as an alternative to the translaryngeal approach, performing VFI via the subglottic airway. Hurst introduced a modification of the CT approach that utilized the submucosal pathway. This modified approach may significantly reduce the risk of bleeding, laryngospasm, and injectate spillage. It also has accelerated the spread of office-based VFI. Currently, some laryngologists favor the CT approach because the submucosal pathway has several advantages: a low complication rate, suitability as an office-based procedure, and good patient compliance. However, despite these advantages, this approach has the inevitable limitation that precise localization of the needle tip is difficult because it utilizes the submucosal route. To overcome this problem, most laryngologists have used physical maneuvers such as CT membrane palpation and the distortion feature of the vocal fold, as well as anatomical references. Nonetheless, a high level of experience is required to perform precise VFI, and a steep learning curve is necessary to master this technique.

The corresponding author (W.C.) also had difficulties mastering the CT approach, which motivated conceptualization of a new technique that allows simultaneous VFI under light guidance: real-time light-guided VFI (RL-VFI). In a previous study, the concept model to implement RL-VFI was developed and its technical feasibility was validated. In the present study, we aimed to verify the feasibility of the new technique in a cadaveric canine model, simulating the setting of office-based VFI, and to explore its clinical usefulness.

MATERIALS AND METHODS

The Prototype Device for RL-VFI

For this study, we developed the new prototype device for RL-VFI upgraded from the previous concept model, which was designed and manufactured by two biomedical engineers (C.J.C. and S.C.Y.). The device comprises three components: the light source, the controller, and the injectors (Fig. 1A). The light source has two laser diode modules of red (635 nanometers [nm], 5 megawatts [mW]) and green (532 nm, 5 mW) color and emits light via a single optic fiber connector using a beam splitter. The controller has dial buttons to control the color and brightness under videolaryngoscopy between the two different types of injectors: 50-μm and 100-μm core diameter optic fiber. The visibility of the injector was validated with the prototype device (Fig. 1C). Hyaluronic acid (HA) (Neuramis Light Lidocaine; Medytox Inc., Cheongju-si, Republic of Korea) was used as the injectate.

Animal Experiment Setting to Simulate Office-Based RL-VFI

A full, high-definition videolaryngoscopy system consisting of a video processor (EPK-i7000; PENTAX Medical, Tokyo, Japan) and video nasopharyngolaryngoscope (VNL-1190STR; 3.7-mm, PENTAX Medical) was used to obtain images of the vocal folds during RL-VFI. The canine larynx was vertically skeletonized by removing the strap muscles to expose the thyroid and cricoid cartilages. Both superior horns of thyroid cartilages were anchored with two safety pins, which were connected to the lateral fixators using rubber bands.

RESULTS

The Visibility of the Injector According to the Color of the Light Source

Visibility of the injector was validated under the two different colors: red and green. The power of the light source was set at the same level (5 mW). The red color light source conferred somewhat better visibility in the canine vocal fold under videolaryngoscopy (Fig. 2).

The Visibility of the Injectors According to the Diameter of the Optic Fiber

The visibility of the injector was validated with the two different types of injectors: 50-μm and 100-μm core diameter optic fiber. There was no difference in visibility or brightness under videolaryngoscopy between the 50-μm and 100-μm optic fibers when a 5 mW red light source was sent to the injector.

Identification of the Needle Tip Pathway From Various Insertion Points Using the Prototype RL-VFI Device

On the basis of the preceding experiment, which validated light color and optic fiber diameter, the following experiments were performed using the red light source, the 23-gauge needle, and the 50-μm optic fiber. The conventional insertion points in VFI using the CT approach are known as 5 to 12 mm from the midline just below the inferior border of the thyroid cartilage. First, the injector was inserted just below the inferior border of the thyroid cartilage at a point 10 mm from the
midline, which simulated the conventional insertion maneuver of the CT approach (Fig. 3). The needle tip was initially identified at the anterior portion of the left vocal fold and then moved along a pathway parallel to the vocal ligament.

To simulate VFI at the insertion points, which are relatively far medial or lateral compared to the conventional insertion point, we performed VFI from insertion points 3 mm from the midline, which is medial to the conventional point; and 17 mm from the midline, which is lateral to the conventional point. When the injector was introduced at the insertion point 3 mm from the midline, the light of the needle tip was initially visualized through the CT membrane without penetration (Fig. 4). Avoiding penetration of the CT membrane, the needle could be rotated laterally and advanced to the vocal fold under light guidance. The needle tip could be positioned at the intended point of the left vocal fold, as indicated by the light.

When the injector was introduced at the insertion point 17 mm from the midline, the light of needle tip was initially identified at the lateral and posterior parts of

Fig. 1. (A) The prototype device for RL-VFI. The device comprises three components: the light source, the controller, and the injectors. The light source has two laser diode modules of red and green color and emits light via a single optic fiber connector using a beam splitter. The controller has dial buttons to control the color and brightness of the light. The injector consists of a commercial needle (1.5 inch, 23- or 25-gauge; Jung Rim Medical Industrial Co. Ltd., Seoul, Republic of Korea) and a custom-made connector with an optic fiber cable (50-μm or 100-μm core diameter). (B) A custom-made larynx fixation device as the simulator for VFI. The L-shape platform of the simulator were made with wooden plates. The silicone cylinder to fixate the canine trachea was attached on the basement, and the lateral fixators were attached on the posterior surface of the wall. Both superior horns of thyroid cartilages were anchored with two safety pins, which were connected to the lateral fixators using rubber bands. (C) A full, high-definition videolaryngoscopy system consisting of a video processor (EPK-i7000; PENTAX Medical, Tokyo, Japan) and video nasopharyngolaryngoscope (VNL-1190STK; 3.7-mm, PENTAX Medical) was used to obtain images of the vocal folds during RL-VFI. The canine larynx was vertically mounted on a custom-made larynx fixation device to mimic the sitting position adopted during office-based VFI. As in real office-based VFI using the CT approach, our otolaryngology residents performed laryngoscopy to confirm the surgical field. A laryngologist then performed RL-VFI using the prototype device. RL-VFI = real-time light-guided vocal fold injection. [Color figure can be viewed in the online issue, which is available at www.laryngoscope.com.]

Fig. 2. The light source has two laser diode modules of red (635 nm, 5 mW) and green (532 nm, 5 mW) color and emits light via a single optic fiber connector. The injectors emit the light of red (A) and green (B) color at the needle tip. The visibility of the injector was validated under the two different colors—red (C) and green (D). The red color light source conferred somewhat better visibility in the canine vocal fold under videolaryngoscopy. [Color figure can be viewed in the online issue, which is available at www.laryngoscope.com.]
vocal fold (Fig. 5). Unlike when using the conventional (10 mm) and the medial (3 mm) insertion points, the needle could be introduced into the vocal fold in various directions of posteromedial, medial, and anteromedial under light guidance. The needle tip could be positioned on the intended point of the left vocal fold.

Regardless of the injection routes, the location of the needle tip was accurately indicated by light, and its depth

---

**Fig. 3.** The conventional insertion points in VFI using the CT approach are known as 5 to 12 mm from the midline just below the inferior border of the thyroid cartilage. Firstly, the injector was inserted just below the inferior border of the thyroid cartilage at the point 10 mm from the midline, which simulated the conventional insertion maneuver of the CT approach. (A) The needle insertion at the point 10 mm from the midline in the cadaveric canine model simulating the setting of office-based VFI. (E) The midline and the insertion point (10 mm) are indicated with the two needles in the canine larynx. (B) The imaginary route of the needle tip is indicated by the black arrow. (C, D, F–H) The needle tips are easily identified by the guiding light on the left vocal fold. The needle tip was initially identified at the anterior portion of the left vocal fold and then moved along a pathway parallel to the vocal ligament. CT = cricothyroid; VFI = vocal fold injection. [Color figure can be viewed in the online issue, which is available at www.laryngoscope.com.]

**Fig. 4.** To simulate VFI at the insertion points that are relatively far medial compared to the conventional insertion point, we performed VFI from insertion points 3 mm from the midline, which is medial to the conventional point. (A) The needle insertion at the point 3 mm from the midline in the cadaveric canine model simulating the setting of office-based VFI. (E) The midline and the insertion point (3 mm) are indicated with the two needles in the canine larynx. (B) The imaginary route of the needle tip is indicated by the black arrow. (C, D, F–H) The needle tips are easily identified by the guiding light on the CT membrane and the left vocal fold. When the injector was introduced at the insertion point 3 mm from the midline, the light of the needle tip was initially visualized through the CT membrane without penetration. Avoiding penetration of the CT membrane, the needle could be rotated laterally and advanced to the vocal fold under light guidance. The needle tip could be positioned at the intended point of the left vocal fold, as indicated by the light. CT = cricothyroid; VFI = vocal fold injection. [Color figure can be viewed in the online issue, which is available at www.laryngoscope.com.]
within the mucosa could be estimated according to the brightness and size of the light.

**RL-VFI With HA Using the Prototype Device**

Before the material was injected into the vocal fold, its ability to pass through the injector was confirmed. HA could pass through all types of the injectors (23-gauge/100-μm, 23-gauge/50-μm, 25-gauge/100-μm, and 25-gauge/50-μm) without resistance. Under the guidance of red light emitted from the needle tip, precise and simultaneous VFI could be performed at the intended point. The vocal fold was gradually medialized as the injectate was instilled into a point just lateral to the vocal process (Fig. 6).

**Identification of the Needle Tip During Reinjection**

During office-based VFI, the patient’s involuntary responses, such as coughing or swallowing, can lead to slippage of the needle from the vocal fold or insertion point. Therefore, we tried to simulate the scenario of reinjection. After the initial injection of HA, the injector was completely removed from the larynx and reinserted into the vocal fold. When the needle tip was introduced into the injected material, the light was dispersed, and its location was easily identified (Fig. 7).

**DISCUSSION**

Despite the advantages, several factors limit application of the CT approach from the perspective of laryngologists. First, the insertion route may be complex because it is comprised of various different anatomical layers: skin, subcutaneous fat, CT membrane, the paraglottic space, and the thyroarytenoid (TA) muscle. Second, the entire route of the needle via the CT approach is basically a blind pathway for laryngologists when performed submucosally. Third, after introduction of the needle into the vocal fold, the most difficult task is the precise localization of the needle tip. In particular, an accurate localization of the needle is very tricky, and most laryngologists identify the position of the needle tip indirectly by the distortion of vocal fold configuration. Finally, because of the above-mentioned reasons, accurate localization requires a high level of experience,11 and there is a steep learning curve involved in mastering this technique.8,12 Also, there are several limitations from the perspective of patients. Obesity might be a problem that is specific to the CT approach because the too long pathway from the skin to the CT membrane would make it difficult to target the proper insertion area. Surgery such as thyroidectomy can result in fibrosis of the tissue around the CT membrane. It is difficult to distinguish the thyroid cartilage, CT membrane, cricoid, and trachea in the fibrotic neck. The loss of external surface anatomy and CT membrane palpation maneuver can lead to numerous tentative and exploratory insertions of the needle. Although penetration of the CT membrane is successful with proper alignment of the superior-to-inferior axis, the needle can be penetrated into the airway or be positioned far laterally to the vocal fold or the TA muscle due to inaccurate identification of the midline.
Previously, various efforts have been made to overcome the limitations of CT approach. Hirano et al. suggested a needle tip localization maneuver in which the initial blind needle insertion is performed during palpation of the cricothyroid membrane and the position of the needle is ascertained by identifying distortion and subtle movements in the vocal folds. This maneuver is still useful and critical in current practice. Laryngologists had numerous clinical experiences relying on only these maneuvers. For this reason, Jin et al.

Fig. 6. RL-VFI with hyaluronic acid using the prototype device. (A) The procedure in the cadaveric canine model simulating the setting of office-based RL-VFI. (E) Before the material was injected into the vocal fold, its ability to pass through the injector was confirmed. (C, D, F–H). Under the guidance of red light emitted from the needle tip, precise and simultaneous VFI could be performed at the intended point. The vocal fold was gradually medialized as the injectate was instilled into a point just lateral to the vocal process. RL-VFI = real-time light-guided vocal fold injection. [Color figure can be viewed in the online issue, which is available at www.laryngoscope.com.]

Fig. 7. Identification of the needle tip during reinjection. During office-based vocal fold injection, the patient’s involuntary responses, such as coughing or swallowing, can lead to slippage of the needle from the vocal fold or insertion point. Therefore, we tried to mimic the scenario of reinjection. After initial injection of hyaluronic acid, the injector was reinserted into the vocal fold. (A–D) When the needle tip was introduced into the injected material, the light was dispersed and its location was easily identified. [Color figure can be viewed in the online issue, which is available at www.laryngoscope.com.]
suggested an anatomical guideline for the CT approach.\textsuperscript{10} They provided anatomical information regarding the depth and angulation of the injection needle during VFIs, analyzing three dimensionally reconstructed CT in patients with unilateral vocal fold paralysis. Hoffman et al. first tried to identify the position of the needle tip using the CT approach.\textsuperscript{12} They demonstrated a technique for transillumination of the vocal folds in humans and in a canine larynx model that used a white-colored light-emitting diode as the light source and a modified 23-gauge needle within the metal sheath of the connector. We consider this concept innovative and commendable because it was the first trial to indicate the needle location using light. However, one limitation of a transilluminated needle tip guidance is that the fiber optic cable must be removed before the injectate can be delivered. This removal-and-reinsertion technique is difficult to apply for “real-world” office-based practice. Thus, we postulated that a new concept (RL-VFI) requires two features to overcome the shortcomings of the CT approach and improve its practicality: 1) the needle tip should be visible to allow identification of its precise location, and 2) the injectate should be instilled simultaneously without manipulation. We then developed a concept model satisfying these necessities and demonstrated its technical feasibility in the previous pilot study.\textsuperscript{5}

In the previous study, we used a single color (red), a single optic fiber diameter (200 \(\mu\)m), and a single needle gauge (25-gauge).\textsuperscript{5} Generally, red light is less absorbed by muscle and mucosa. Nonetheless, we tried to validate the visibility of another color (green) because the tissue around the vocal fold is thin. In this study, red color conferred much better visibility in the canine vocal fold under videolaryngoscopy. In the previous study, we used a relatively large optic fiber (200 \(\mu\)m) and found that the injection pressure was significantly higher than in a needle without any optic fiber. Thus, in the present study, we tried to validate visibility using smaller-diameter optic fibers of (50-\(\mu\)m and 100-\(\mu\)m) to reduce the injection pressure or resistance of the needle. There was no difference in visibility or brightness between the 50-\(\mu\)m and 100-\(\mu\)m diameter optic fibers with the same light power (5 mW). Because smaller diameters would improve the ability of injectate materials to pass through the needle, we chose red color and an optic fiber diameter of 50 \(\mu\)m in subsequent experiments.

The conventional insertion area in VFI using the CT approach is 5 to 12 mm from the midline just below the inferior border of the thyroid cartilage.\textsuperscript{8,13} During VFI via CT approach without the device, the route of the needle tip might be ambiguous and usually is presumed by an anatomical imaginary line or a change of vocal fold configuration. The present study demonstrated that the route of the needle tip could be quite easily identified when inserted on the conventional insertion point (10-mm) using the device for RL-VFI. There are several reasons to avoid points that are far medial or lateral to the conventional insertion area. When the insertion point is too medial (< 5 mm), the needle can easily penetrate the mucosa of the CT membrane and enter the subglottic airway. Conversely, when the insertion point is too lateral (> 12 mm), it is difficult to find the needle tip because it is located in the deep paraglottic space. In this study, we tried to introduce the needle tip in various insertion points (3-mm and 17-mm) other than the conventional point (10-mm).

As mentioned above, when VFI via CT approach without the device is performed on the thick or fibrotic skin, it would be difficult to determine the exact midline despite the CT membrane palpation. Thus, too close or contralateral insertion to the midline may occur under the ambiguous external anatomy and can lead to the penetration of the airway, which results in coughing, gag reflex, and bleeding. To simulate this clinical scenario of inadvertent medial insertion, we performed RL-VFI at a point that was too medial (3-mm) to the midline. During VFI via CT approach without the device, a subtle pinpoint elevation under careful examination implies that the needle is about to penetrate the CT membrane mucosa.\textsuperscript{13} During RL-VFI, we could easily notice the light on the CT membrane mucosa before the change in mucosal configuration and avoid accidental penetration of it.

The superoposterior direction route from the conventional insertion point in the CT approach is widely accepted by laryngologists. However, anterior lesions—or, in extreme cases, anterior commissure of the vocal fold—are considered a blind spot, which is a shortcoming of the CT approach compared to the transoral approach. Due to this shortcoming, anterior lesions such as vocal fold nodules would usually be treated using VFI with a transoral, transnasal, or transhyoid approach.\textsuperscript{14} In this study, we tried to validate the feasibility of the lateral CT approach whereby the needle is inserted at a point far lateral (>12 mm) to the midline. The needle route of the lateral CT approach is very complex because it passes through the skin, the subcutaneous fat, diverse extrinsic and intrinsic laryngeal muscles, the paraglottic space, and then the TA muscle. Also, it is hard for laryngologist to draw the imaginary path because it is not parallel to the vocal fold. For these reasons, the insertion from the points far lateral to the midline is taboo and thus usually would not be attempted by most laryngologists. In the present study, we found that RL-VFI could easily facilitate the lateral CT approach. When the needle was introduced at the point 17 mm from the midline, the needle tip appeared at the posterior vocal fold, just lateral to the vocal process. With the lateral CT approach using RL-VFI, the three directions of the route (posteromedial, medial, and anteromedial) were possible.

Another issue regarding the VFI procedure is reinjection, which could be occasionally encountered in various clinical situations. Injection volume is of vital importance to the configuration of the vocal folds and postoperative voice, especially when using permanent materials such as calcium hydroxypatite (CaHA). For this reason, some laryngologists prefer the gradual injection of material with auditory feedback on intraoperative voice, performing repetitive injections at the same location of the vocal fold. Furthermore, poor patient compliance, as well as coughing or swallowing, can cause the needle to slip from the vocal fold after insufficient injection. In some cases, the vocal fold can transform to unwanted shape due to rapid absorption or migration of
the material. Small volume reinjections or touch-up injections may be necessary in such situations. However, the precise localization is more challenging during reinjection because the vocal fold space filled with the material is not easily distorted by movement of the needle. In this study, to simulate this clinical situation, we performed reinjection using RL-VFI after completing the injection of HA. When the lighted needle tip was located within the prefilled material, the guiding light dispersed and refracted noticeably. The location of the needle tip could therefore be easily discriminated from adjacent muscle and mucosa without physical distortion of the tissue.

Since the concept of the CT approach was introduced, many pieces of knowledge, technical tips, and maneuvers have been accumulated. In particular, the endoscopic system and new biomedical materials contributed to advancements in the CT approach. However, few advances have been made for the delivery method of the CT approach. In this regard, RL-VFI could be a technical boost for the CT approach and influence the clinical practice. First, it is expected to broaden the indications for treatment using RL-VFI. Currently, the major indications for VFI are vocal fold palsy and spasmodic dysphonia, which usually target the deep laryngeal structure. However, RL-VFI will facilitate the treatment of benign superficial lesions, such as vocal fold nodules, vocal fold polyps, vocal fold cysts, vocal fold granulomas, vocal fold scars, Reinke’s edema, leukoplakia, and laryngeal papillomatosis. Second, RL-VFI would improve the safety of the CT approach. It will prevent misinjection into the superficial layer, which can have disastrous effects on the voice; or misinjection into too deep paraglottic space or extra-laryngeal space, which can rarely lead to inadvertent complication of pulmonary embolism. Third, RL-VFI may lower the barrier to entry of the CT approach for physicians. As clinical educators, we have observed that residents and fellows find it difficult to learn the technique because the blind pathway is anatomically tricky and most procedures are performed under local anesthesia. For the same reason, several researchers have attempted to develop the teaching model or trainer for VFI. It is also expected that RL-VFI will help trainees become familiar with VFI as an education tool.

We demonstrated the feasibility of RL-VFI using our prototype device in an ex vivo canine larynx, simulating the setting of office-based VFI. Clinical application of RL-VFI will improve the safety and precision of the CT approach as well as expand its applications in laryngology.

CONCLUSION

In the present study, we introduced RL-VFI using our customized prototype device in an ex vivo canine larynx, simulating the setting of office-based VFI. Clinical application of RL-VFI will improve the safety and precision of the CT approach as well as expand its applications in laryngology.

BIBLIOGRAPHY