Chapter 39
OCT and malignant lesions of the larynx

Tjoson Tjoa, David Shamouelian, Veronika Volgger & Brian J.F. Wong

Abstract

This chapter describes advantages and pitfalls of OCT in assisting diagnosis of laryngeal carcinoma.

Keywords: *OCT, laryngeal carcinoma, laryngeal tumors, laryngeal malignancy*

Introduction

Cancer of the larynx is the third most common malignancy of the upper aerodigestive tract [1] with 85-95% of these being squamous cell carcinoma, a result of malignant transformation of the epithelial layer of the larynx [2]. The male-female ratio of the disease is approximately 5:1 [3]. With relatively little change in mortality since the 1970's, current clinical focus is not simply on improving survival, but on laryngeal preservation modalities. The key to preserving function of the larynx lies in prevention and early diagnosis. Fortunately, the majority of laryngeal cancer involves the true vocal cords, which tend to be symptomatic early in the course of a lesion. Early symptomatology can result in early diagnosis. When patients have hoarseness persisting for three weeks or swallowing pain/difficulty lasting six weeks, it is important to evaluate for a malignant lesion. [4]

The World Health Organization categorizes lesions of the larynx into pre-malignant and malignant, depending on the degree of histologic abnormality and depth of involvement [5]. Squamous cell dysplasia is a premalignant lesion of squamous epithelium characterized by cellular atypia and loss of normal maturation and stratification short of carcinoma *in situ* (CIS). The cellular abnormalities of mild dysplasia are minor and are limited to the basal one third of the epithelium; moderate dysplasia displays more marked cellular abnormalities involving up to two thirds of the epithelial thickness; and severe dysplasia has cellular abnormalities involving more than two thirds of the epithelial thickness. CIS is an intraepithelial neoplasm in which the full thickness of the squamous epithelium shows the cellular features of carcinoma without violation of the basement membrane and stromal invasion. Severe dysplasia and CIS are classified as different lesions, but clinically they behave in a similar fashion and may be considered as one.

Rates of malignant transformation, or transition to a cancerous lesion, range from 11% for mild dysplasia to 30% for severe dysplasia and CIS. The likelihood of progression is related to the extent of disease. Because all pre-malignant lesions possess some potential to progress to invasive cancer, long-term follow-up of all premalignant laryngeal lesions is warranted [6].

Diagnosis

The diagnosis of laryngeal cancer is currently based largely on physical exam and surgical biopsy. Symptoms at presentation include hoarseness, dysphagia, odynophagia, shortness of breath, and referred otalgia. Once a patient presents with voice or swallowing

complaints, it is important to thoroughly examine the larynx. Indirect mirror laryngoscopy can be used, but often is insufficient for closely examining the laryngeal mucosa and can miss the anterior commissure. Flexible fiberoptic laryngoscopy and video stroboscopy can be used to more closely and thoroughly inspect the laryngeal mucosa, including all subsites of the supraglottis and glottis. These include the epiglottis, arytenoids, ary-epiglottic folds, false vocal cords, and true vocal cords. Assessment for gross vocal cord mobility and mucosa wave can also be performed, which help provide clues as to the extent of any lesions that exist. However, these examination techniques alone cannot provide a specific diagnosis.

For diagnosis of any laryngeal lesions, the gold standard currently involves microdirect laryngoscopy (laryngeal microsurgery) with excisional biopsy, typically performed under general anesthesia, allowing for complete and thorough examination of the laryngeal mucosa with sampling of concerning areas [7]. The difficulty with excisional biopsy lies in identifying the margin of the lesion, and thus the extent of the area requiring removal. It is very difficult to determine where an invasive lesion ends by visualization alone. Taking large areas of the laryngeal mucosa in order to identify and clear margins carries significant morbidity, including swallowing and voice dysfunction. With the push towards organ preservation, it is imperative that adjunctive methods to determine margins are developed.

Imaging modalities such as computed tomography (CT) scan and magnetic resonance imaging (MRI) can be adjunctive, but neither has a diagnostic accuracy greater than 88% [8]. Additionally, both CT and MRI are able to image subsurface tissue architecture, but have very limited resolution for superficial mucosal changes. They can be very useful once a lesion is identified to determine if there is extralaryngeal involvement. The anatomic spaces surrounding the larynx, including the pre-epiglottic, paraglottic, and thyroid cartilage, are difficult to assess on physical examination but are essential in determining the initial stage of a lesion.

Positron Emission Tomography (PET) scanning can indicate areas of high metabolic activity and has been used in conjunction with CT imaging pre-operatively to assess for synchronous and metastatic lesions, but is not sensitive for delineating borders of malignant lesions. PET/CT scanning is commonly used post-treatment for surveillance as it has a sensitivity and specificity greater than 90% for detecting recurrent or residual lesions [4].

The importance of delineating the extent of tumors cannot be underestimated with the current push towards laryngeal preservation. Once a malignant lesion is identified, the goal of surgical excision is to clear all disease while preserving as much healthy tissue as possible. Pathology studies have shown that by leaving positive margins, the risk of local recurrence increases from 32% to 80% [9]. Over the last ten to twenty years, a number of technologies have been developed to image superficial epithelium at tumor margins in an effort to limit the morbidity of excision while adequately clearing disease.

OCT, which combines a low-coherence light source with inferometry to produce high-resolution images of superficial living tissues, is one such promising modality. It is analogous to ultrasonography but instead of sound uses light to discern variation in tissue optical properties. It is an *in vivo*, noninvasive imaging modality that provides information in real time. Images can be obtained in the operating room at video-frame rates. It was initially developed to visualize the retina [10] and subsequently used in other anatomic sites such as the GI tract, urinary system, skin, and coronary arteries. Since then, it has been found to be useful in visualizing epithelial and sub-epithelial tissue to depth 1-1.5 mm [11-12]. OCT usage in head and neck has been developed as part of an effort to bring optical imaging technology from the research lab into the operating room to improve surgeons' abilities to identify tumor margins [13].

OCT in diagnosis and tumor margin identification

At a histological level, stratification of tissue layers is one of the criterion for healthy laryngeal mucosa. In normal laryngeal mucosa, OCT has been shown to correlate well with histological sections allowing identification of the epithelial layer, the lamina propria, and subepithelial glandular structures and vessels [14]. Within the larynx, carcinomatous processes destroy the regular structure of mucous membranes and do not permit typical differentiation of cell layers. Thus, mucosa within a malignant lesion of the larynx has a higher degree of tissue homogeneity than healthy mucosa [11]. OCT has been used to take advantage of this finding.

Shakhov et al. performed a study using OCT to detect changes in the in stratification of tissue layers as a sign of pathologic tissue alterations. Various lesions were examined with OCT and compared with histologic diagnoses after excision. These included CIS and early stage malignant lesions of the vocal cords. The detection of boundaries of malignant lesions based on the absence of normal tissue stratification with OCT was more precise than boundaries detected by gross visualization and endoscopy, based on color and texture changes [15]. Further work in *ex vivo* laryngectomy samples showed that OCT can be used to detect differences in reflectivity of the subglottic mucosa and that a high reflectivity corresponds to hyperplastic epithelium [14]. It is thought that with minimally invasive techniques such as laser excision, OCT can be used as an adjunct to identify margins and preserve as much normal tissue as possible while clearing disease.

Perhaps one of the most useful applications of OCT is to demonstrate the integrity or disruption of the basement membrane. Our group performed OCT imaging of at least one laryngeal sub-site in 82 patients with both benign and malignant processes and found that OCT was able to consistently identify the demarcation between the laryngeal epithelium and the underlying lamina propria in normal patients and most benign pathologic processes [16]. This demarcation corresponds to the basement membrane, which is known to be disrupted in invasive squamous cell carcinoma [17]. This was verified with an OCT imaging study that included 22 patients with laryngeal cancer [18]. In each of the patients, the demarcation between the epithelium and the submucosa tissue was demonstrated. Additionally, in many of the cases, a transition zone was visualized at the periphery of the tumor.

Kraft and Arens compared microdirect laryngoscopy with palpation of lesions with and without OCT imaging and compared their results with histologic specimens taken on excisional biopsy of 193 patients. A total of 217 lesions were examined and palpated, with and without OCT. When compared to histologic specimens, visualization with OCT was able to accurately diagnose 89% of lesions, whereas visualization without OCT revealed an accurate diagnosis 80%. In addition, the measurement of the epithelial thickness and identification of the basement membrane were used to accurately predict the dysplasia grade in 71% and malignancy in 93% of patients. Evidence has shown that even though the maximum depth of penetration 1-1.5 mm limits imaging thicker exophytic lesions, OCT provides extremely precise images of the epithelium/lamina propria interface at the basement membrane.

Limitations of OCT

While the applications of OCT have been shown to be very useful in determining malignant lesions, identifying margins, and even grading dysplasia, the technology is still very new to the field of head and neck and is not without its limitations. As applied to living tissues, where light is scattered and absorbed, OCT has a limited depth of penetration [19]. With current technology, the depth of penetration with OCT imaging is at best 1-1.2 mm. At this point, there is no widely available technology to delineate both the epithe-

lium/superficial lamina propria interface *in vivo* and image the deeper vocal fold (VF) layers (ligament and muscle).

Additionally, bulky hypercellular lesions can block signal penetration. In particular, thick scar tissue, laryngeal papillomas, and blood can all block signal penetration. It is not uncommon for laryngeal malignancies to have areas of scar and or bleeding that can decrease the sensitivity of OCT to detect margins. Additionally, there are other lesions such as granulation tissue (scar) and ulcerative lesions in which the basement membrane is absent or undetectable. This can decrease the specificity of OCT for laryngeal malignancy [18, 20].

As previously mentioned, OCT has been used with great accuracy to diagnose laryngeal lesions and even predict the grade of dysplasia. However, Kraft and Arens also noted in their study microinvasive carcinoma could not be distinguished from severe dysplasia or CIS [21]. Additionally, another group has shown that while both the sensitivity and specificity of OCT in detecting invasive lesions is close to 90%, hyperplastic and dysplastic lesions could not be differentiated based on OCT [20]. Specifically, hyperplastic and hyperkeratotic lesions caused increased light reflection at their surface, which led to less light propagating beneath the epithelium and decreased ability to detect the basement membrane.

A form of OCT that is called Polarization Senstive OCT (PS-OCT) has provided additional information about the structure of laryngeal lesions. PS-OCT delineates both tissue structure and birefringence by measuring intensity and polarization state of reflected light [22]. In particular, molecules such as collagen, which is found in scar tissue, can change the polarization state of reflected light and cause it to appear in specific organized patterns during imaging with OCT. When used to examine benign laryngeal lesions, PS-OCT combined with conventional OCT provides additional information about areas of scar within normal tissue. This adjunctive form of OCT holds promise for delineating scar from malignancy as well.

Future directions

OCT has the potential to become an extremely valuable tool as the push for organ preservation becomes stronger. With the increased usage of transoral endoscopic laser surgery for the excision of laryngeal tumors and the advent of robotics within the field of head and neck surgery, OCT provides the surgeon with an appreciation for the position of the tumor margins in three dimensions intra-operatively. When used as an adjunctive modality, it may streamline, reduce reliance upon, and augment frozen section histologic analysis as a noninvasive method of confirming clear margins. This is image guided surgery at a microscopic level.

Additionally, OCT allows for the monitoring disease over time. There have been studies examining more office-based OCT imaging, which would allow for the outpatient monitoring of individuals' responses to radiation and chemotherapy, or for surveillance of suspicious lesions over time.

References

1. Siegel, R., Naishadham, D., Jemal, A., 2013. Cancer statistics, 2013. CA Cancer J. Clin. 63, 11-30.
2. Barnes, L., et al., 2005. Tumours of the hypopharynx, larynx and trachea: Introduction, in: Barnes, L., Eveson, J., Reichart, P., Sidransky, D. (Eds.), World Health Organization Classification of Tumours. Pathology and Genetics of Head and Neck Tumours. IARC Press, Lyon, pp. 111-117.

3. Jemal, A., et al., 2004. Cancer statistics, 2004. CA Cancer J. Clin. 54, 8-29.
4. Chu, E., Kim, Y., 2008. Laryngeal cancer: Diagnosis and preoperative work-up. Otolaryngol. Clin. North Am. 41, 673-695.
5. Shanmugaratnam, K., 1991. Histological typing of tumours of the upper respiratory tract and ear. World Health Organization. International Histological Classification of Tumours, 2nd ed. Springer-Verlag, Berlin.
6. Armstrong, W., Vokes, D., Maisel, R., 2010. Malignant tumors of the larynx, in: Flint, P. (Ed.), Cummings Otolaryngology: Head & Neck Surgery, 5th ed. Mosby Elsevier, Philadelphia.
7. Kraft, M., et al., 2008. Clinical value of optical coherence tomography in laryngology. Head Neck 30, 1628-1635.
8. Hartl, D., 2012. Evidence-based practice: Management of glottic cancer. Otolaryngol. Clin. North Am. 45, 1143-1161.
9. Batsakis, J., 1999. Surgical excision margins: A pathologist's perspective. Adv. Anat. Pathol. 6, 140-148.
10. Huang, D., et al., 1991. Optical coherence tomography. Science 254, 1178-1181.
11. Sergeev, A., et al., 1997. In vivo endoscopic OCT imaging of precancer and cancer states of human mucosa. Opt. Express 1, 432-440.
12. Wong, B., et al., 2004. Imaging the internal structure of the rat cochlea using optical coherence tomography at 0.827 µm and 1.3 µm. Otolaryngol. Head Neck Surg. 130, 334-338.
13. Hughes, O., Stone, N., Kraft, M., Arens, C., Birchall, M., 2010. Optical and molecular techniques to identify tumor margins within the larynx. Head Neck. 32, 1544-1553.
14. Bibas, A., et al., 2004. 3-D optical coherence tomography of the laryngeal mucosa. Clin. Otolaryngol. Allied Sci. 29, 713-720.
15. Shakhov, A., et al., 2001. Optical coherence tomography monitoring for laser surgery of laryngeal carcinoma. J. Surg. Oncol. 77, 253-258.
16. Wong, B., et al., 2005. In vivo optical coherence tomography of the human larynx: Normative and benign pathology in 82 patients. Laryngoscope 115, 1904-1911.
17. Rubinstein, M., et al., 2010. Optical coherence tomography of the larynx using the Niris system. J. Otolaryngol. Head Neck Surg. 39, 150-156.
18. Armstrong, W., et al., 2006. Optical coherence tomography of laryngeal cancer. Laryngoscope 116, 1107-1113.
19. Mahmood, U., et al., 2006. Evaluation of rabbit tracheal inflammation using optical coherence tomography. Chest 130, 863-868.
20. Volgger, V., et al., 2013. Evaluation of optical coherence tomography to discriminate lesions of the upper aerodigestive tract. Head Neck 35, 1558-1566.
21. Kraft, M., Lüerssen, K., Lubatschowski, H., Glanz, H., Arens, C., 2007. Technique of optical coherence tomography of the larynx during microlaryngoscopy. Laryngoscope 117, 950-952.
22. Burns, J., Kim, K., deBoer, J., Anderson, R., Zeitels, S., 2011. Polarization-sensitive optical coherence tomography imaging of benign and malignant laryngeal lesions: An in vivo study. Otolaryngol. Head Neck Surg. 145, 91-99.