Ex vivo high resolution imaging with a miniaturized microendoscope to discriminate between benign and malignant mucosa in the upper aerodigestive tract

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Introduction

- Surgical treatment is an important component of multidisciplinary care for patients with laryngeal squamous cell carcinoma.
- Effective surgical treatment requires complete removal of diseased tissue with clear margins in order to prevent recurrence.
- Optimal, minimal amounts of benign mucosa should be removed which will decrease treatment-associated morbidity and mortality.
- Current practice requires intraoperative margin determination by frozen section which adds to the complexity of the procedure.
- High resolution imaging is a relatively new modality which allows for the acquisition of real time images of the epithelial tissue and has been previously evaluated to detect mucosal changes in numerous sites along the upper aerodigestive tract, including the oral cavity and the esophagus.
- This study is the first to determine the feasibility of using high resolution imaging to discriminate between non-neoplastic and neoplastic mucosa of the larynx.
- The ability to discriminate between normal and cancerous mucosa at the time of tumor resection will allow for more precise tumor ablation while minimizing the amount of healthy tissue removed.
- The use of optical imaging to determine intraoperative margins provides a potential mechanism by which tissue morphology can be non-invasively visualized at the time of surgical treatment for laryngeal cancer.

Methods

- Four patients with biopsy-proven squamous cell carcinoma of the larynx consented to imaging of the specimen after standard-of-care surgical resection.
- Following resection for biopsy-proven squamous cell carcinoma of the larynx, sites of both grossly “normal” and suspicious mucosa were imaged with the fiber optic HRME after surface staining with the nuclear dye proflavin.
- Imaged areas were either marked with India Ink or subjected to punch biopsy to allow for histopathological correlation.
- Images obtained with the high resolution microscope were compared to the goldstandard of histopathologic diagnosis.

Results

- Areas of histopathologically confirmed carcinoma had an increased density of nuclei and more disorganization when compared to normal mucosa.
- Lesion margins could be visualized by moving the probe over the transition zone between benign and cancerous mucosa.
- In specimens that were heavily keratinized, autofluorescence artifact limited the ability to visualize cell nuclei.

Figure 1. High Resolution Microendoscope (HRME). The device consists of a fiber-optic bundle which can deliver high resolution narrow field images of tissue morphology. The image is magnified by a microscope objective. Surface staining with a nuclear dye is required to display the morphology of the epithelial tissue.

Figure 2. Representative images obtained with the HRME (Left) with corresponding histopathological images (Right). A. Benign squamous epithelium of the larynx. B. Dysplasia C. Squamous cell carcinoma. D. Ulcerating squamous cell carcinoma. Note that HRME images of malignant mucosa feature increased nuclear density, and nuclear disorganization.

Figure 3. Limitations of imaging with high resolution microendoscopy. HRME (Top) and corresponding H&E histopathology (Bottom). A. Hyperfluorescence in areas of hyperkeratinization. B. Inability to detect submucosal tumor spread. C. Benign histologies which feature increased nuclear density (such as pseudostratified ciliated “respiratory” epithelium pictured above).

Summary

- The HRME can image epithelial changes present in squamous cell carcinoma of the laryngeal mucosa.
- The presence of autofluorescence artifact and the inability to visualize submucosal tumor spread are significant limitations to be overcome.

Conclusions

- High resolution optical imaging provides non-invasive visualization of laryngeal epithelium in real-time and shows promise as a new technology for determination of intraoperative tumor margins.
- The ability to determine optical margins during cancer surgery has the potential to reduce the number intraoperative frozen-section pathology consultations.
- Continued refinement of this optical imaging technology may lead to improved ability to accurately determine optical margins, facilitate the sparing of uninvolved normal tissue, and find application to novel technological platforms, such as robotic surgery.

References


