

ON-LINE APPENDIX

ROI Analysis: Spectral Attenuation Characteristics of Normal Nonossified Thyroid Cartilage

To evaluate the consistency and reproducibility of spectral attenuation characteristics of nonossified thyroid cartilage, we performed an evaluation in 3 different patient groups. First, normal/unaffected cartilage was evaluated in all patients with HNSCC, except 3 patients with completely ossified cartilages. In addition to providing an evaluation of normal NOTC in patients with HNSCC, this approach also enabled calculation of NOTC-tumor contrast-to-noise ratios in the same patient later on. Furthermore, NOTC was evaluated in 10 healthy patients. Healthy subjects were selected ad hoc from the same period and consisted of those with normal or near-normal scan findings with minor incidental findings (dental periapical lucencies, benign reactive lymph nodes or tonsillar enlargement, and incidental cutaneous lesions such as sebaceous cysts). To be included, all normal or near-normal cases must have had NOTC on at least 3 separate 1.25-mm sections. The clinical indications for the normal study findings are provided in On-line Table 1. The NOTC attenuation of the 3 groups was compared at each kiloelectron volt across the spectral attenuation curve evaluated with 1-way ANOVA with Tukey multiple comparison tests.

We have already demonstrated that other than for a small lucency at the junction with ossified parts in some cases, the NOTC itself is homogeneous with uniform attenuation on conventional CT.²⁴ In this study, the spectral attenuation characteristics of NOTC and tumor on DECT were evaluated by using circular ROIs. We elected the largest diameter ROI that would avoid volume averaging with adjacent structures. For this study, the minimum ROI diameter used was 1.5 mm (corresponding to a sampled area per ROI of 1.77 mm²) and the maximum diameter used was 5.5 mm (corresponding to a sampled area per ROI of 23.76 mm²). Our goal was to obtain sufficient coverage and representation while avoiding volume averaging with adjacent structures, heterogeneous/necrotic tumor parts, and so forth. Thus, a range was used rather than using the smallest common circular ROI that could be placed on the usually thin NOTC. For a representative assessment, multiple ROIs were used per patient for analysis. First, in the healthy patient group, 18 ROIs were used per patient. In this group, 9 ROIs were placed on each side, on at least 3 different sections. An example of ROIs placed is shown in On-line Fig 1A. For optimal positioning, images were zoomed in before ROI placement. The normal cartilage from the 10 patients in this group was sampled with 180 (18 × 10) ROIs.

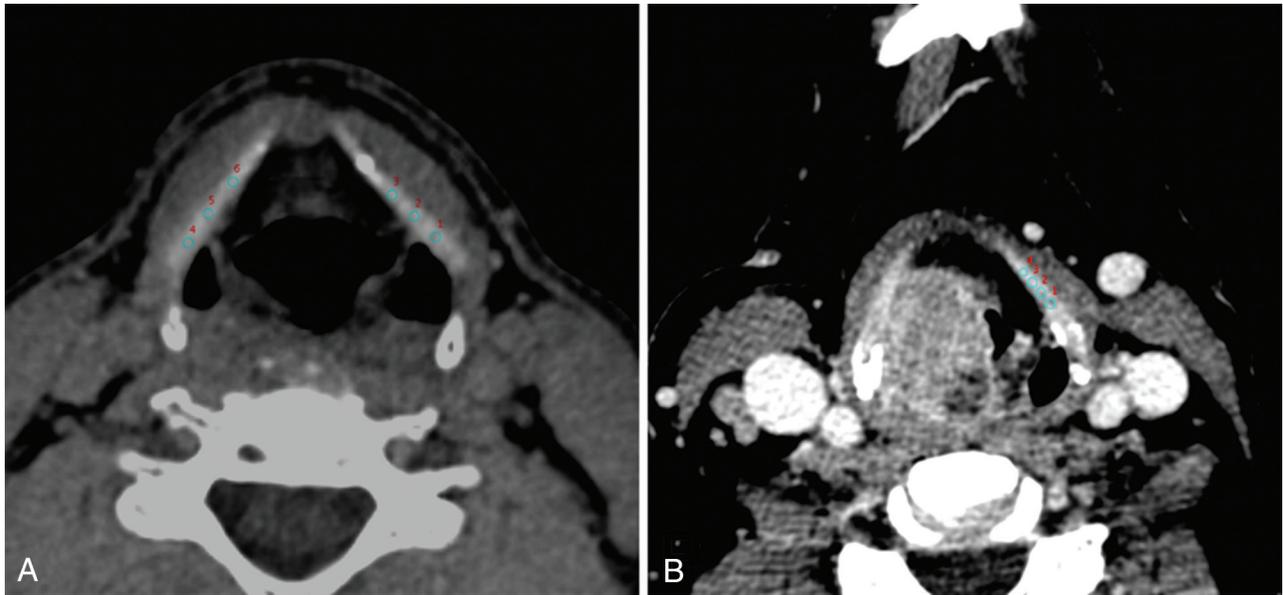
Thereafter, the normal/unaffected cartilage in the tumor group (the Table and On-line Table 2) was evaluated. Of 30 patients with head and neck squamous cell carcinoma, in 3 cases, the thyroid cartilage was completely ossified. Therefore, in these cases, the NOTC could not be evaluated. The normal/unaffected

NOTC in the other 27 patients with HNSCC was evaluated with 9 ROIs per patient, for a total of 243 ROIs (9 × 27). For tumors outside the larynx, none invaded or abutted the thyroid cartilage, including the 1 case of hypopharyngeal tumor. For cases of laryngeal cancer, in all but 1 case, the contralateral NOTC was completely unaffected and therefore was used for analysis. In 1 case, there was small bilateral focal paramedian anterior thyroid cartilage invasion. In this case, the posterior part of the uninvolved cartilage, distant from the level of invasion (by at least 5 contiguous sections where there was no cartilage invasion or abutment) was used. An example of normal NOTC ROI placement in a patient with laryngeal cancer is shown in On-line Fig 1B. For evaluation of tumors, 9 ROIs were used per tumor/patient, for a total of 270 ROIs (9 × 30).

The homogeneous enhancing part of the tumor was evaluated, and areas of cystic change/necrosis were avoided. At the end of the article, in Fig 5, we provide an example of potential future applications of our work, such as placing ROIs in the invaded cartilage. However, for the purpose of this study, which was to compare characteristics of tumor with those of NOTC, all quantitative evaluation of tumor was performed in the part of the tumor not invading the cartilage. Furthermore, ROIs were not placed in areas of visible artifacts, for example, secondary to dental fillings for tumors involving the oral cavity or oropharynx. Typically, larger ROIs could be placed in tumors rather than in the thin cartilage. Combined, 693 ROIs were used for evaluation of NOTC and tumor in this study. The average ROI area evaluated per cartilage or tumor in each patient was 86.8 mm² (range, 21.2–212.1 mm²). In calculating averages among cases, each NOTC or tumor was considered equivalent (ie, we did not correct/provide different weighting based on the area covered, which could have biased the analysis in favor of larger NOTCs or tumors).

Differences in Attenuation of NOTC Compared with Tumors on High Kiloelectron Volt VMIs

At kiloelectron volts of ≥ 95 , there was no overlap between individual mean tumor and NOTC attenuation values, as shown in the scatterplot in Fig 3B. At 95 keV, the mean attenuations of tumor and NOTC were 63.4 ± 9.2 HU and 103.9 ± 8.6 HU, respectively. The highest measured individual tumor attenuation was 86.2 HU, and the lowest measured individual NOTC attenuation was 88.4 HU (see scatterplot in Fig 3B). The difference between these groups further increased with increasing kiloelectron volt, with the greatest difference at 140 keV. On 140-keV VMIs, the mean tumor and NOTC attenuations were 48.0 ± 8.1 and 97.3 ± 9.0 , respectively. The highest measured individual tumor attenuation was 64.3 HU, and the lowest measured individual NOTC attenuation was 79.3 HU, with no overlap between tumor and NOTC attenuations in our study population (Fig 3B).



ON-LINE FIG 1. Examples of ROIs placed for analysis of normal NOTC. A, Example of ROI analysis in a case from the healthy patient group. B, Example of ROI analysis of the normal/unaffected contralateral (left) NOTC in a patient with a right supraglottic cancer. At least 9 ROIs were used per cartilage or tumor in each patient (and 18 ROIs per NOTC in healthy subjects, 9 on each side).

On-line Table 1: Clinical indications for neck CTs in the healthy patient group^a

Patient No.	Clinical Indication
1	Axillary lymph node; rule out lymphoma
2	Recurrent right parotid swelling; rule out stone
3	Rule out right level 5 lymphadenopathy
4	Bilateral parotid hypertrophy, left more than right
5	Patient with severe otalgia, with pain referred to right ear
6	Globus, not responding to proton pump inhibitors, left-sided throat discomfort; rule out mass
7	Rule out lymphoma
8	Rule out lymphadenopathy
9	Feeling of fullness in the throat at the level of base of tongue
10	History of globus sensation, increased dysphagia, worsening sensation of pain; rule out soft-tissue mass

^aNone of the patients had a known malignancy or major systemic disease.

On-line Table 2: HPV status of untreated tumors (ie, excluding recurrent or metastatic tumors)

Primary Site	HPV Status
Larynx	5/7, HPV-negative; 2/7, HPV status not available
Hypopharynx	1/1, HPV-negative
Retromolar trigone, anterior tonsillar pillar	3/3, HPV-negative
Oral cavity, other	5/5, HPV-negative
Oropharynx, other	2/3, HPV-positive; 1/3, HPV-negative
Sinuses, nose	1/3, HPV-positive; 1/3, HPV-negative; 1/3, HPV status not available

Note:—HPV indicates human papillomavirus.