Comparison of Thyroid, Auricular, and Costal Cartilage Donor Sites for Laryngotracheal Reconstruction in an Animal Model

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**Objective:** To evaluate and compare the use of autogenous thyroid cartilage with that of auricular and costal cartilage in laryngotracheoplasty (LTP).

**Design:** A blinded comparison of LTP techniques using anterior thyroid, auricular, or costal cartilage as graft material in a rabbit model. Histological and anatomical analyses were performed on the laryngeal specimens 1, 4, and 6 weeks after surgery. The following factors were analyzed in each specimen: graft viability, cartilage proliferation, perichondrial viability, degree of necrosis, inflammatory response, and degree of epithelialization.

**Subjects:** Fifty-seven New Zealand adult male rabbits, aged 6 months, were divided into 3 study groups (19 animals in each group) initially and equally into the 3 time periods.

**Results:** No episodes of respiratory compromise occurred in any of the animals in the 3 study groups. Gross inspection of the laryngotracheal complex in the thyroid cartilage group revealed no evidence of laryngeal structural compromise. There was no statistical difference between the 3 types of cartilage used for reconstruction for the variables of graft or perichondrial viability, degree of necrosis, or inflammatory response at 1, 4, or 6 weeks. Cartilage proliferation in the thyroid cartilage group was decreased compared with that in the other 2 groups at 1 week. The amount of proliferation increased in this group and was equal to the amount present in the other 2 groups 4 and 6 weeks after surgery. Complete epithelialization of the graft material was present in all 3 groups at 4 and 6 weeks after reconstruction.

**Conclusions:** The use of autogenous thyroid cartilage for LTP compares favorably with that of other methods of reconstruction that use either auricular or costal cartilage in the rabbit model. This technique is a viable alternative for single-stage LTP and has the added advantage of using a single incision.


During the past 30 years, the treatment of severe subglottic stenosis has evolved into the now widely accepted concept of surgical expansion of the stenotic laryngeal lumen. Early methods, pioneered by Rethi in 1956, focused on splitting the cartilaginous skeleton. In 1969, Lapidot et al reported on a method of repairing subglottic stenosis in piglets that used an interposition technique. Fearon and Cotton extended the experimental work of Lapidot et al and popularized the method of anteriorly splitting the thyrocricoid complex and placing autogenous graft material in children. Their technique—laryngotracheoplasty (LTP)—has become an accepted method for treating severe subglottic stenosis.

The best augmentation material for LTP remains controversial. Many autogenous grafts have been described. Ideally, the graft material should be malleable to permit carving and shaping, but firm enough to stent the anterior cricoid and tracheal cartilage open. The autogenous material should be similar to tissue in the area. It should hold sutures well and be easy to harvest. The graft harvest should be associated with minimal donor site morbidity and preferably be obtained through the same operative incision used for graft placement.

Costal and auricular cartilages are the most common augmentation materials used in the LTP procedures. Donor site discomfort and morbidity are well-documented problems when either rib or ear cartilage is used in LTP procedures. To our knowledge, no one has described the use of an anterior thyroid lamina cartilage graft in single-stage laryngotracheal reconstruction. This type of graft offers several advantages, particularly the use of a single operative inci-
ANIMALS AND METHODS

Fifty-seven white New Zealand adult male rabbits, aged 6 months, were used in this investigation. Their preoperative mean weight was 4.0 kg (range, 3.3-4.4 kg). The animals were divided into 3 groups of 19 animals each. All animals underwent anterior LTP procedures with the use of auricular, costal, or TAC grafts.

SURGICAL TECHNIQUE

On the day of surgery, each rabbit received premedication consisting of subcutaneously administered atropine sulfate (3 mg), followed by apropazine (0.18 mg/kg) and xylazine hydrochloride (0.18 mg/kg) administered intramuscularly. The animals were intubated, and anesthesia was maintained with the use of 2% halothane. Sterile conditions were maintained throughout all operative procedures. Auricular and costal cartilages were harvested using the methods of Cotton11 and Wiatrak et al.12 A 20-mm section of cartilaginous rib was harvested. A section of auricular cartilage 10 x 10 mm was removed. The ventral perichondrial layer was included and preserved on each graft.

The TAC graft was obtained through the LTP incision. A vertical midline incision was made over the larynx and upper trachea, and the airway was exposed. The TAC graft was harvested as follows: the anterior aspect of the thyroid cartilage was exposed and examined. A 5 x 8-mm section of cartilage was removed from the superior aspect of the left thyroid lamina (Figure 1). Care was taken to leave the medial, or inner, layer of the perichondrium intact. The ventral, or lateral, surface of the perichondrium was harvested with the graft. After the grafts were harvested from 1 of the 3 locations, the material was then sculpted into a 5 x 8-mm piece with tapered ends (Figure 2).

The laryngotracheal complex was exposed for the costal and auricular graft models using the incision described for the TAC graft. Then a vertical midline incision was made in the cricoid cartilage and first tracheal ring, and the pre-shapened graft was interposed between the cut edges of the cricoid cartilage and tracheal ring. The graft was positioned with the perichondrial layer facing the airway lumen (Figure 2). The graft was fixed in position with 5-0 polydioxanone sutures. The neck incision was closed in layers using 4-0 Vicryl sutures. Each animal was extubated at the conclusion of the operative procedure.

The animals in each group were then evenly distributed within 1 of 3 subsets based on the time to laryngeal harvest: 1, 4, and 6 weeks. At the various time points from the date of the surgery, the animals were killed. The larynges were harvested and transversely sectioned through the region of the cartilage graft. The sections were stained with hematoxylin-eosin, and histological analysis was performed.

All procedures were conducted humanely and approved by the Institutional Animal Care and Use committees of the University of Toronto School of Medicine and the Hospital for Sick Children, Toronto, Ontario.

HISTOLOGICAL ANALYSIS

Six histological variables were observed and graded in each specimen. The pathologist (M.R.S.) was blinded to the graft material and to the duration of graft placement in each animal. The following variables were analyzed: graft viability (nonviable = 0 and viable = 1), cartilage proliferation (none = 0 and active proliferation = 1), perichondrial layer (not intact = 0 and intact = 1), degree of necrosis (minimal = 1, moderate = 2, and severe = 3), inflammatory response (minimal = 1, moderate = 2, and severe = 3), and epithelialization of the intraluminal graft surface (<25% = 0, 26%-50% = 1, 51%-75% = 2, and >75% = 3).

We used the Kruskal-Wallis test for the statistical analysis of all histological variables with the aid of a statistical software program (Minitab version 12.1; Minitab, Inc, State College, Pa), with P < .05 considered significant.

RESULTS

OBSERVATIONS

No episodes of respiratory compromise occurred in any of the animals studied. Seven animals died in the early postoperative period. Three animals in the group having costal cartilage graft died (2 in the 1-week and 1 in the 4-week subsets), and 3 animals in the group having auricular cartilage graft died (2 in the 1-week and 1 in the 4-week subsets). An additional rabbit from the group having TAC graft (1-week subset) also died. All animals that died prematurely failed to maintain oral feeding, with no evidence of respiratory distress in any of these animals. Autopsies were not performed.

Gross inspection of the laryngotracheal complex at the time of harvest revealed a widely patent airway in all animals studied in all 3 graft material groups at all intervals (Figures 3, 4, and 5).
tilage formation increased from 1 to 4 weeks in the TAC graft group. The difference between the 3 graft material groups was not significant by 4 weeks. A trend toward earlier graft epithelialization was observed in the group having the costal cartilage graft compared with the other 2 groups. Complete graft epithelialization was present in most of the animals in all 3 groups by 6 weeks after graft placement. No viable graft cartilage was found in 3 of the animals in the group with the costal cartilage graft (Table 2, specimens F, N, and O) and 2 of the specimens from the group with the TAC graft (Table 1, specimens A and O). Although new cartilage formation was seen in 3 of the 4 animals, the new cartilage was found in association with the cut edges of the native laryngo-tracheal cartilage.

**COMMENT**

No single, consistently successful, method of repair for subglottic stenosis has been devised. Several methods that use TAC have previously been described. Pedicled TAC was proposed as a graft material for cricoid expansion, first by Fearon and Cotton\(^3\) and then later by Fry et al.\(^{15}\) Whereas the idea of a muscle pedicled graft with its own blood supply and mucoperichondrium is attractive, in practice, it is not a good option in small children due to limitations in size. Free TAC grafts in the posterior cricoid lamina were evaluated in dogs by Strome et al.\(^{8}\) and found not to be beneficial. Rice and Colman\(^{16}\) described

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**Figure 1.** The thyroid alar cartilage graft procedure showing harvesting of the superior aspect of the thyroid ala. Note that the inner perichondrium is preserved.

**Figure 2.** The thyroid alar cartilage graft procedure showing graft preparation and application. The graft outer perichondrial layer is placed facing the subglottic lumen.

**Figure 3.** A transverse section of thyroid alar cartilage graft 6 weeks after graft placement showing the graft anteriorly (hematoxylin-eosin, original magnification ×10).

**Figure 4.** A transverse section of costal cartilage graft 6 weeks after graft placement showing the graft anteriorly (hematoxylin-eosin, original magnification ×10).

**Figure 5.** A transverse section of auricular cartilage graft 6 weeks after graft placement showing the graft anteriorly (hematoxylin-eosin, original magnification ×10).
the use of a free thyroid alar perichondrial graft in dogs with and without stents. They found that in 9 of 10 animals studied, the amount of cartilage formed from the free perichondrial graft was adequate to bridge the airway. The shape of the subglottic lumen, however, was unpredictable.17

This animal study was undertaken to evaluate the use of a TAC graft in laryngotracheal reconstruction compared with 2 other well-accepted graft materials (auricular and costal cartilages). The TAC graft compared favorably with both the auricular and costal cartilage graft models.

One problem with the present study is the limited sample size in each group. The small number makes statistical evaluation difficult and could also explain why no significant difference was found between groups.

Using an autogenous TAC graft offers many theoretical advantages compared with other techniques. Superior thyroid cartilage lamina is nearly identical in thickness to the cricoid cartilage.
Viable perichondrium, however, is well known to superior portion of TAC does not affect laryngeal growth. A recent animal study indicates that removing the bidity was observed up to 6 weeks after cartilage harvesting of the thyroid lamina. No evidence of donor site mor-

Bible decreases the possibility of prolapse of the graft material into the tracheal lumen. The perichondrium is harvested with the lateral surface of the thyroid cartilage and can be positioned facing intraluminally when the graft is sutured in place. This provides a surface for epithelialization that limits the potential for the formation of granulation tissue. Most important, the graft is harvested through the same neck incision used to expose the cricoid cartilage, eliminating the need for a second operative site. Possible disadvantages of the TAC graft include limitations of the size of the graft, destabilization of the laryngeal skeleton, and alteration in the growth of the thyroid lamina. No evidence of donor site morbidity was observed up to 6 weeks after cartilage harvest. A recent animal study indicates that removing the superior portion of TAC does not affect laryngeal growth. Viable perichondrium, however, is well known to induce neocartilage formation. The inner perichondrium at the superior aspect of the TAC donor site is carefully preserved to promote the generation of new cartilage. Although the present study is applicable only to an animal model, future investigations will focus on the application of this technique in the pediatric airway.

In conclusion, the use of autogenous TAC graft represents a viable option and alternative for single-stage cricoid expansion procedures and has the added advantage of using a single incision.

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REFERENCES