MECHANICAL EVALUATION OF A MODIFIED PROSTHETIC LARYNGOPLASTY USING A TOGGLE TECHNIQUE FOR THE EQUINE ARYTENOID CARTILAGE

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THESIS

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ABSTRACT

Recurrent laryngeal neuropathy is a frequent cause of poor performance and upper airway noise in horses. This disorder results in varied degrees of upper airway obstruction due to neurogenic atrophy of the cricoarytenoideus dorsalis muscle secondary to degeneration of the recurrent laryngeal nerve. The treatment of choice in horses with recurrent laryngeal neuropathy is the prosthetic laryngoplasty, which utilizes a suture prosthesis to replace the function of the cricoarytenoideus dorsalis and abduct the arytenoid cartilage. The most frequent complication associated with the prosthetic laryngoplasty is gradual loss of arytenoid abduction in the post-operative period. This gradual loss is typically attributed to cyclic fatigue of the muscular process of the arytenoid and partial suture pull-through from the cartilage. A modified laryngoplasty technique was developed to add strength and stability to the prosthesis with the purpose of reducing cyclic fatigue. This modified technique utilizes a titanium suture toggle passed through the muscular process and base of the arytenoid to distribute loading over a larger area of cartilage.

The objectives of this study were to compare the biomechanical properties of the standard technique to the modified laryngoplasty technique in both monotonic and cyclic loading. Larynges from 41 horses were collected following euthanasia and used for mechanical testing. Laryngoplasty constructs were performed using a standard technique on one side and a modified technique on the other. For monotonic loading, the laryngoplasty constructs were prepared and suture ends attached to a load-frame, then distracted at 100 mm/minute until failure. Mean load at failure and failure modes were compared between techniques. For cyclic loading the arytenoid cartilages were maximally abducted and constructs were circumferentially loaded for 10,000
cycles. Loss of arytenoid abduction was evaluated every 500 cycles using both a subjective grading scale and objective change in rima glottidis cross sectional area.

In monotonic loading, the modified laryngoplasty constructs failed at a higher load (191N ± 29N) than the standard laryngoplasty constructs (91N ± 44N, P<0.001). None of the modified constructs failed by suture pull-through from the muscular process of the arytenoid cartilage, whereas most of the standard laryngoplasty constructs failed by suture pulling through the muscular process of arytenoid cartilage. In cyclic testing, 11/20 (55%) of standard laryngoplasty constructs reached surgical failure, considered a Dixon grade 3, while 0/20 (0%) of modified laryngoplasty constructs failed. The modified laryngoplasty constructs lost less rima glottidis cross sectional area compared to the standard laryngoplasty constructs (P<0.001) in circumferential loading. Overall, the modified laryngoplasty technique proved superior to the standard laryngoplasty in both monotonic and cyclic loading.
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CHAPTER 1
INTRODUCTION

Recurrent laryngeal neuropathy (RLN) is a common cause for poor performance in horses worldwide. (1–3) RLN is a progressive demyelinating disease of the recurrent laryngeal nerve that leads to atrophy and paresis/paralysis of the ipsilateral cricoarytenoideus dorsalis muscle. (4,5) Neurogenic atrophy of the cricoarytenoideus dorsalis muscle results in dyspnea, respiratory noise, hypoventilation, severe hypoxia, hypercapnia, hyperlactatemia, and exercise intolerance secondary to upper airway obstruction. (6–8)

Prosthetic laryngoplasty (LP) and ipsilateral vocal cordectomy or ventriculectomy is considered the treatment of choice in horses diagnosed with RLN. (5) The standard LP technique involves placing a prosthetic suture between the caudal border of the cricoid cartilage and the muscular process of the arytenoid cartilage to stabilize the arytenoid cartilage in full-abduction to restore airway diameter and airflow. (5,8,9) However, postoperative loss of arytenoid abduction and subsequent reduction of the rima glottidis cross sectional area is a frequent postoperative complication; (10,11) this is often attributed to partial or complete suture pull-through or breakage of the muscular process of the arytenoid cartilage. (10,12) It has been suggested that failure of the prosthesis to maintain adequate arytenoid abduction is due to the loading exerted on the prosthesis during coughing and swallowing. (12) The recorded load range exerted on the LP suture during coughing and swallowing is 45 to 55 N, (12) which is similar to the one (56 ±13 N) reported in ex vivo LP constructs that failed by suture pull-through the muscular process of the arytenoid cartilage. (13)

Facilitated ankylosis of the cricoarytenoid joint is potentially effective; (14,15) however, loss of arytenoid abduction can still occur in the postoperatively period before ankylosis is complete. Therefore, a highly stable LP prosthesis that can resist loading throughout the period
of ankylosis is required to consistently achieve adequate and persistent arytenoid cartilage abduction and reliably restore upper airway function in horses with RLN. It has been suggested that deeper suture placement into the muscular process of arytenoid cartilage would prevent loss of arytenoid abduction,(16) however, suture placement in the standard fashion further rostral may result in reduced mechanical advantage in fully abducting the arytenoid cartilage.(16) We have developed a modified LP technique that uses a suture button to toggle the suture material to the base of the muscular process of the arytenoid cartilage. By incorporating a much larger portion of the arytenoid cartilage, this modification should reduce suture pull-through from the muscular process of the arytenoid cartilage and progressive postoperative loss of arytenoid abduction in horses with RLN undergoing LP.

The objective of this study was to compare the biomechanical properties of modified and standard LP constructs in both monotonic and cyclic loading using an ex vivo model. We hypothesized that modified LP constructs would have a higher load at failure in monotonic loading than standard LP constructs, as well as a different mode of failure. In addition, we hypothesized that modified LP constructs would maintain a greater rima glottidis cross sectional area in cyclic testing than standard LP constructs evaluated both subjectively and objectively.
CHAPTER 2
LITERATURE REVIEW

2.1 - Anatomy and Physiology of the Equine Larynx

The larynx, as the junction between the pharynx and trachea, acts to serve as a conduit for airflow, protect the airway during swallowing, and provide a means for vocalization.(5,17) An intimate relationship between the laryngeal cartilages, intrinsic muscles, and extrinsic muscles allows for normal function of the larynx. Dysfunction of a single component results in substantial clinical consequences. The larynx consists of five laryngeal cartilages: the epiglottic cartilage, thyroid cartilage, cricoid cartilage, and paired arytenoid cartilages. The arytenoid cartilages play a major role in maintaining rima glottidis area, defined as the opening through the larynx into the trachea.(5) The arytenoid cartilages pivot around the cricoarytenoid joint to reduce the airway lumen during swallowing (adduction), and increase airway diameter during inhalation (abduction).(5) While the other cartilages have distinct roles in normal physiology and disease, the arytenoid cartilages are one of the most dynamic structures within the larynx, particularly with respiration.

The intrinsic laryngeal musculature includes those muscles involved in movement of the laryngeal cartilages or soft tissues in relation to each other. These include the cricoarytenoideus dorsalis, cricoarytenoideus lateralis, ventricularis, vocalis, cricothyroideus, and transverse arytenoideus muscles.(5,17) All but the cricothyroideus are innervated by the recurrent laryngeal nerve, a branch off of the vagus nerve.(5) The cricothyroideus is innervated by the external branch of the cranial laryngeal nerve, also a branch of the vagus nerve.(5) Contraction of the cricoarytenoideus lateralis, ventricularis, vocalis, and transverse arytenoideus cause adduction of the arytenoid cartilages, resulting in narrowing of the rima glottidis. The cricoarytenoideus dorsalis causes abduction of the arytenoid cartilages and increase in rima glottidis area; thus
dysfunction can result in a substantial reduction in inspiratory airflow.(5) The extrinsic laryngeal musculature works to move or stabilize the larynx as a whole. The extrinsic laryngeal muscles include the thyrohyoideus, hyoepiglotticus, sternothyroideus, and pharyngeal constrictors (thyropharyngeus and cricopharyngeus).(5,17) Additional soft tissue structures present in the equine larynx include the paired vocal folds and laryngeal saccules or ventricles.(5,17)

2.2 - What is Recurrent Laryngeal Neuropathy?

Recurrent laryngeal neuropathy (RLN), also known as laryngeal hemiplegia, is a progressive demyelinating disease of the recurrent laryngeal nerve. The etiology of RLN is still unknown, despite years of research, and cases are characterized as idiopathic in nature.(5,18,19) Idiopathic RLN nearly invariably involves the left hemi-larynx.(5,20) Clinical signs of RLN most frequently involve poor performance and/or an upper airway stridor at exercise, often described as ‘roaring’. (5) The recurrent laryngeal nerves are the longest nerves in the horse, and the left nerve is considerably longer than the right due to its course around the base of the heart and aorta.(21) The long length of the recurrent laryngeal nerve has been implicated in the pathophysiology of RLN through excessive stretching of the nerve or ischemic injury due to a tenuous local blood supply.(19,22) Local inflammation of the recurrent laryngeal nerve secondary to inflammation associated with lymphadenopathy, environmental toxins, and genetic factors have all been proposed as possible etiologies for RLN.(22) About 4% of laryngeal paresis/paralysis cases in horses are right sided or bilateral, and are associated with infection, perivascular injections, trauma, or neoplasia.(5,20)

Histopathologic abnormalities of the left recurrent laryngeal nerve of horses with RLN consist of a preferential decrease in large myelinated fibers, particularly in the distal portions of
the nerve. (22) Interestingly, the right recurrent laryngeal nerve in horses with RLN develops mild axonal loss as well, resulting in RLN being characterized as a bilateral mononeuropathy. (22) Curiously the adductor muscles of the larynx typically show more significant histopathologic changes than the cricoarytenoideus dorsalis, despite the fact that the clinical signs of RLN are attributed to abductor dysfunction. (19)

2.3 - Diagnosis and Grading of RLN

Upper airway endoscopy is the gold standard diagnostic procedure for horses with RLN; however, over ground or treadmill endoscopy may be necessary to fully assess the impact of the disease during exercise. (5) A variety of endoscopic grading systems have been developed and validated, and while small variations exist between the different systems, the basic premise and criteria are similar. (23–26) A single 7-grade system, combining both resting and exercising endoscopy, has been developed to standardize the diagnosis and grading of RLN. (5,26) The synchronicity and degree of arytenoid abduction are taken into account in grading RLN, with various subgrades used to more fully describe the clinical scenario. (5,26) However, even with the expanded grading system currently in use, there is only moderate agreement between different examinations of the same horse; this suggests an importance in evaluating the horse and presenting clinical signs in relation to the endoscopic findings. (27) Approximately 7% of horses with normal arytenoid abduction at rest display abnormalities attributable to RLN when exercised. (28) Furthermore, the soft tissues of the upper airway, including the vocal cords, ventricles, and aryepiglottic folds may contribute to dynamic collapse during exercise secondary to the altered airway pressures in horses with RLN. (29) These additional abnormalities require exercising endoscopy for diagnosis and lead to a more complete clinical picture in a given horse.
Laryngeal ultrasonography has recently shown promise as an additional diagnostic procedure for RLN if exercising endoscopy is unavailable. The cricoarytenoideus lateralis is one of the first muscles to undergo neurogenic atrophy in horses with RLN and is easily visualized with transcutaneous ultrasonography. (4,30) Using treadmill endoscopy as the gold standard, ultrasound echogenicity of the cricoarytenoideus lateralis was more accurate in diagnosing RLN than resting upper airway endoscopy. (31) This diagnostic technique may provide clinicians with additional options when evaluating suspected RLN cases.

2.4 - Airway Mechanics in Normal and RLN-Affected Horses

Since RLN is a disorder primarily affecting the exercising horse, a variety of performance variables have been assessed in affected horses. (32) Pulmonary function testing, including evaluation of respiratory flow, upper airway impedance, and pressure differentials through various parts of the respiratory tree, have been used to assess the airflow impact of RLN. (32–35) Inspiratory upper airway pressures can reach -30 cm H₂O at maximal exercise in normal horses. (36) In horses with RLN, the inspiratory upper airway pressure is further decreased, reaching -70-80 cm H₂O, more than doubling the normal exercising inspiratory airway pressure. (36) This increased upper airway pressure resulted in reduced inspiratory airflow in horses with RLN, which has been documented through the use of flow-volume loops. (37) Additionally, evaluation of arterial blood gas parameters (including oxygen and lactate) have been used extensively for assessment of performance limiting disorders in horses. (32,33,38–40) Normal partial pressure of oxygen (PaO₂) in a resting horse is 90-110 mmHg, and mild to moderate hypoxemia can occur with high intensity exercise, resulting in PaO₂ values of 60-70 mmHg. (41) However, in horses with RLN, this hypoxemia drops even further, with PaO₂ values
well below 60mmHg documented.(39) Lactate, a marker indicative of anaerobic cellular metabolism, is also increased between resting and high intensity exercise in normal horses. A difference of 4.3mg/dl was documented in one study between normal and exercising horses.(39) However, an additional increase in lactate occurs secondary to RLN, with an increase of nearly 20mg/dl in horses with RLN at high exercise, likely due to additional hypoxemia.(39)

2.5 - Treatment Options for RLN

Several treatment options have been described for managing horses with RLN, and the optimal treatment depends on a variety of patient and disease factors. The major clinical signs to be considered in selecting the optimal treatment include the presence of inspiratory stridor and exercise intolerance. Described treatments include ventriculectomy/ventriculocordectomy, arytenoidectomy, laryngeal reinnervation, and LP.(5,19,42)

Ventriculectomy was one of the earliest treatment options for horses with RLN and was described in 1845.(42) The procedure is now known as the ‘Hobday’ procedure, and was named after the veterinarian who refined and promoted the use of the procedure.(42) Typically, the ventriculectomy is combined with removal of one or both vocal folds (ventriculocordectomy). Both procedures are described with a variety of surgical approaches and techniques, but all with the same end goal. Dynamic collapse of the laryngeal ventricle and vocal folds in horses with RLN contributes to a reduced airway diameter, turbulent airflow, and upper airway noise.(5,42) Removal of the laryngeal ventricle and vocal fold results in less soft tissue to reverberate in the upper airway, as well as cause peri-laryngeal fibrosis which will assist in stabilization of the arytenoid cartilage.(5) Ventriculectomy and ventriculocordectomy can be performed either standing or under general anesthesia, with the tissue removed either sharply via a laryngotomy
incision or with a transendoscopic diode laser. (5,42–46) Ventriculocordectomy has been shown to reduce airway stridor in horses with RLN, while ventriculectomy alone provides some noise reduction but to a lesser extent. (43–45) Ventriculocordectomy has been advocated as a sole treatment in horses with low grades of RLN; however, while ventriculocordectomy does reduce airway noise, it cannot return airway function to normal in horses with RLN. (43,47) Generally, ventriculocordectomy is viewed as a potential sole treatment option in horses with a presenting complaint of upper airway stridor or as an adjunct to LP, but is unlikely to provide enough improvement in horses with higher grades of RLN or overt exercise intolerance. (42) These techniques are often performed in combination with LP and improves resolution of upper airway noise in horses undergoing LP. (5,10,11,42,44,47–57)

Arytenoidectomy is typically considered the treatment of choice for arytenoid chondritis or following failure of a LP, but can also be used as an initial treatment in horses with RLN. (5,42) This procedure is performed via laryngotomy under general anesthesia or standing sedation. (5,11,42,58–60) Originally, subtotal arytenoidectomy was described, leaving both the muscular process and corniculate process in situ; however, limited improvement was noted in airway flow after the procedure in horses with induced RLN, and the procedure has been largely abandoned. (42,61) Partial arytenoidectomy, which also removes the corniculate process, does improve upper airway flow in induced models of laryngeal hemiplegia, and has therefore replaced the subtotal arytenoidectomy. (42,58) However, airway mechanics improve but do not return to normal at near maximal airflow rates. (58) Horses treated with partial arytenoidectomy have a fair prognosis for returning to racing performance; however, the LP is superior at very high airway requirements. (11,48,60,62)
Several techniques for laryngeal reinnervation have been described, and while these may gain additional popularity as the procedure is refined, they are not currently the preferred technique among most surgeons for treating horses with RLN.(42) Described procedures include nerve anastomosis, direct nerve implantation, and neuromuscular pedicle grafts.(63–66) The neuromuscular pedicle graft procedure has been the most effective technique for restoring function to the cricoarytenoideus dorsalis, and this procedure has been the focus of more recent research on the technique.(66) These procedures are technically challenging, but remain the only methods for truly restoring function to the paralyzed musculature in horses with RLN.(42,66) However, in addition to the technical difficulty of these procedures, they rely on the presence of functional muscle tissue to reinnervate, which may not be the case in horses with advanced RLN.(66) The reinnervation process requires several months to determine if it will be successful, and with the short racing career of many racehorses, this convalescence time is not acceptable.(66) Electronic pacing of the recurrent laryngeal nerve has also been described to augment laryngeal reinnervation, but is still very experimental at this time.(67)

The prosthetic laryngoplasty (LP) was first described in the literature by Dr. Marks in 1970.(9,68) In this procedure a suture prosthesis is used to replace the function of the atrophied cricoarytenoideus dorsalis, resulting in permanent abduction of the affected arytenoid cartilage.(5,42) The LP is superior to the previously described surgical techniques in restoring airway mechanics; however it still cannot establish completely normal airflow at maximal exercise.(39) The LP is also unable to effectively eliminate respiratory noise; therefore, it is typically combined with unilateral or bilateral ventriculocordectomy.(43,53,69) The LP has become the gold standard surgical technique in treating horses with RLN; however, it carries a high complication rate and frequently suboptimal success rates.(5,42,70) When success is
defined as return to intended use, success rates are reported from 78-94%. (49,54,56,70,71)

However, when improved postoperative performance is used as the definition of a successful outcome these rates decrease to 45-60%. (49,54,56,70,71) When racehorses and sport horses are evaluated separately, success is considered much higher in sports horses working at submaximal airway requirements compared to racehorses, where airway requirements are greater. (70)

### 2.6 - Laryngoplasty Complications

Unfortunately, despite its widespread use, the LP procedure is fraught with a variety of post-operative complications, some of which may explain the only fair prognosis for improved performance. In addition to typical intra-operative and post-operative complications that are risks with any surgery, the LP has a variety of complications specific to the procedure as well. The most common complication associated with the LP is post-operative loss of arytenoid abduction. (16,71,72) An acute, complete loss of arytenoid abduction has been reported in 3-11% of horses following LP. (16,49,53-55,71) This acute failure typically results from fracture or suture migration through the laryngeal cartilages, and the muscular process is a frequent site of failure. (16,73) The more frequent complication related to loss of arytenoid abduction is seen as a more gradual loss over the first 6 weeks following the procedure. (16,71,74,75) This gradual loss of abduction is so commonly encountered that a grading scale has been developed to evaluate post-operative arytenoid abduction. (71) This 1-5 grading scale has been used in a variety of studies to describe post-operative arytenoid abduction and is frequently used clinically to document surgical outcome. (56,71,74,75) Up to 95% of horses will lose 1-2 grades of arytenoid abduction following LP, resulting in a median grade 3 abduction in the long term. (56,71,74) The definitive cause for this loss of arytenoid abduction is not typically identified in clinical cases.
unless repeat surgery is performed. However, partial to complete suture pull through or fracture of the muscular process of the arytenoid cartilage is often implicated. (16) The cause for failure of the muscular process has been a long-standing source of debate among veterinary surgeons. (16) Age was long considered a potential contributor to LP failure, with the thought that softer, more flexible cartilage in young horses may be more likely to allow for suture pull through. (16, 76) However, age has been shown in numerous in vitro and clinical studies to not have an effect on the presence or degree of arytenoid abduction lost in the post-operative period. (16, 71, 73) Residual function of the recurrent laryngeal nerve has also been implicated in post-operative cyclic loading of LP sutures through continued action of the cricoarytenoideus dorsalis. (16) However, horses develop loss of arytenoid abduction regardless of the pre-operative grade. (71) In addition, surgical transection of the left recurrent laryngeal nerve at the time of LP has not been effective in improving outcomes in horses with grade 3 RLN. (55) Lastly, blockade of the recurrent laryngeal nerve does not change the forces exerted on LP sutures in horses fit with a suture force transducer. (12) Thus far the only factors that have been shown to cause cyclic loading of LP sutures are swallowing and coughing, likely due to the contraction of the caudal pharyngeal muscles. (12) Since this cyclic loading is inevitable and likely contributes to the progressive loss of abduction seen clinically, a LP method that better resists cyclic fatigue and failure will be pivotal in improving clinical outcomes. While some alternative techniques have been developed, few have been widely adopted clinically. (16) Currently the most common strategy to combat loss of arytenoid abduction is to create mildly excessive abduction at the time of surgery, with the assumption that some loss will occur in the post-operative period. (16) However, excessive abduction can cause a host of alternative complications, including dysphagia
and coughing, and reduces the amount of finesse to tailor surgical abduction to a given individual and airway requirement.

Dysphagia and coughing have been described following LP in 14-43% of cases.(54,71) This complication most often is self-limiting and resolves in the first two weeks following the procedure; however, for some horses dysphagia persists and results in increased patient morbidity.(16,42) Long-term coughing and/or dysphagia has been documented in 2.5-26% of cases following LP.(53,54,71) A low number of horses experience dysphagia and coughing due to excessive arytenoid abduction, which may require repeat surgery to loosen the prosthesis.(71) However, it is recommended to delay this repeat procedure if possible, since some post-operative loss of arytenoid abduction is likely to occur, and may resolve the excessive abduction without an additional procedure.(16) Upper esophageal sphincter incompetence has also been associated with excessive retraction or dissection of the caudal pharyngeal musculature, resulting in coughing and dysphagia without the presence of excessive abduction.(77)

A variety of additional dynamic airway conditions may develop following LP and may contribute to post-operative airway noise and poor performance.(16,42) Up to 78% of horses continue to experience upper airway disorders resulting in reduced airway lumen following LP.(74) Dorsal displacement of the soft palate is a frequent pathology of the upper airway and may occur concurrently with RLN, resulting in continued upper airway noise and poor performance despite adequate treatment of RLN.(74) However, dorsal displacement of the soft palate has also been recognized as a complication following LP in 2-6% of horses.(49,54) Additional types of dynamic upper airway conditions can also be seen following LP, including aryepiglottic fold collapse, vocal cord collapse (if only a unilateral vocalcordectomy has been performed), or axial deviation of the corniculate process.(29,78,79) These clinical studies reveal
that continued poor performance following LP may not be due to surgical failure, but instead could be attributed to development of additional upper airway disorders. However, with the high prevalence of dynamic upper airway obstruction in racehorses, it is difficult to determine whether continued poor performance or airway noise can be attributed to the LP versus natural development of an additional upper airway disorder.

Incisional infection has been reported to develop in 0.5-6% of horses receiving a LP. (49,52–55,71) Many of these cases are due to superficial infection which can be treated with drainage and antimicrobial use alone without long-term complication. (16,71) However, penetration through the laryngeal or nasopharyngeal mucosa during prosthesis placement will result in chronic infection of the suture and requires prosthesis removal. (16,80) If acute failure of the muscular process occurs during placement of the prosthesis, a more cranial placement is required, which increases the risk of pharyngeal penetration during surgery.

2.7 – Evolution of the Laryngoplasty – Suture Placement, Prosthesis Material, and Technique

Several modifications to the LP are described in the literature; most of these modifications aim to improve maintenance of arytenoid abduction in the long-term. Many of the alternatives described involve alterations in the anchoring of the prosthesis to the muscular process, where much of the loosening and failure is thought to occur. (19)

The number and location of standard LP sutures have been extensively studied to determine the optimal number and direction of suture pull to maximize arytenoid abduction. In vitro biomechanical testing has shown that the use of two sutures compared to a single suture increases maximal load at failure and cross sectional area of the rima glottidis. (81–83) Suture
placement more dorsal versus lateral in the cricoid did not appear to significantly influence load at failure. (81–83) However, a recent study designed to assess ideal neuromuscular pedicle location showed greatest arytenoid abduction when the suture pull force was 10-30° axial to the muscular process. (84) This location corresponds to the area at the dorsal spine of the cricoid to 1-2 cm abaxial to midline, suggesting that a more dorsally located suture provides mechanical advantage and therefore superior abduction of the arytenoid cartilage.

Since the muscular process is the weak point in the LP construct numerous modifications to the procedure have been attempted to reduce trauma to the cartilage during suture passage and tightening. Cutting point needles, which are required in order to pass through the thick cartilage of the muscular process, can cause curved fissures. (85) Use of a bone trochar reduced fissure formation; however, no significant difference was noted in maximal load at failure compared to use of a cutting point needle. (85) Excessive force on the suture during tying to create adequate abduction can result in acute failure of the muscular process at the time of surgery; therefore, reducing the force required to create maximal arytenoid abduction may help to reduce partial failure or damage of the cartilage at the time of surgery. (57) Performing a unilateral ventriculocordectomy prior to LP can reduce the force required for arytenoid abduction during suture tightening and increase the maximal amount of abduction obtained. (57)

In addition to modifications of the procedure using the traditional suture, alterations have also been made in anchoring methods and materials. Use of a steel cable and washer system to alter both the arytenoid and cricoid cartilage attachments proved significantly stronger in single cycle to failure testing compared to the standard suture LP. (13) Two separate screw-suture constructs have been used to secure the prosthesis to the laryngeal cartilages, and while significantly less distraction occurred during cyclic loading, no benefit was identified with either
alternative construct over the standard suture approach in single cycle loading. (86,87) A system used originally for the repair of canine cranial cruciate ligaments has also been tested as a LP prosthesis in numerous in vivo and in vitro studies and has appeared to fare well in both short-term and long-term evaluation. (88) However, this system was not compared to the standard LP technique, so a superior procedure cannot be determined.

If adequate arytenoid abduction can be achieved with a prosthesis, additional stabilization of the cricoarytenoid joint could help maintain long-term abduction, even if the prosthesis loses strength and stability over time. Immediate stabilization of the cricoarytenoid joint can be obtained by injecting polymethylmethacrylate into the joint at the time of surgery, which will harden within minutes. In vitro evaluation of this method showed increased stabilization during testing in an airflow chamber after the LP suture was cut. (89) Additional methods of facilitating ankylosis of the cricoarytenoid joint without the use of external substances have also been developed. Use of a motorized burr (14) resulted in fibrous bridging of the cricoarytenoid joint by 3 months following LP, while use of a carbon dioxide laser (90) resulted in progressive capsular and periarticular fibrosis over 2 months. These modified LP approaches resulted in maintenance of arytenoid abduction with or without intact LP sutures at the time of airflow chamber testing, proving improved joint stability following induction of fibrosis. (14,90) The major risk with using an ankylosis technique is if abduction is lost prior to joint fibrosis, the arytenoid cartilage may become stabilized at a suboptimal degree of abduction.

Prosthesis material has been largely based on surgeon preference, and common materials include a variety of large diameter, non-absorbable suture types including high molecular weight polyethylene, braided polyester, nylon, stainless steel, and polyurethane. (42) An ex vivo study compared nylon, polyethylene, and polyester in isolated laryngeal cartilages. The polyester
Suture tested showed some superiority with respect to maximum load at failure and distraction over the other suture materials. (91) Polyurethane, also known as Lycra, was used in the original descriptions of the LP and shows some characteristics that would be desirable in a permanent prosthesis, including elasticity, minimal tissue reactivity, and low tissue drag. (92) Clinical studies have shown similar success and complication rates when polyurethane suture was used compared to previous reports with other types of materials. (92) However, when polyurethane was compared to polyester in isolated laryngeal cartilages the polyurethane sutures pulled through the laryngeal cartilages at lower loads. (93) In addition, a case report described laryngeal fistula formation in two horses in which the polyurethane sutures required removal. The authors noted that the polyurethane suture had become brittle and readily broke apart, therefore making removal difficult and likely not providing significant arytenoid abduction at the time of suture removal. (80)

2.8 - Methods for Testing and Subjective Evaluation of the Prosthetic Laryngoplasty

Modifications to the LP can be tested as either in vitro or in vivo experiments. In vitro experiments include single load to failure testing, which involves applying an increasing force until failure, and cyclic loading, which applies a given force over a set number of cycles. Several variations in testing methods have been documented, each with pros and cons related to ease of use, consistency, and ability to mimic in vivo loading. In addition, a variety of subjective measurements of arytenoid abduction have been evaluated both in vivo and in vitro.

Single load to failure, or monotonic testing, is used to determine the mechanical strength of a construct during acute loading, and is evaluated by distraction and maximal load at failure. Individual testing of anchored cartilage-implant constructs allows for a simple, consistent
loading design.(85,87,93,94) This method allows for a very secure anchoring of the cartilage which resists failure at the attachment point. However, by testing the cartilages in isolation the interaction which would occur in vivo is lost. A clamp system attached to the laryngeal cartilages allows for testing of the entire LP construct.(83) Unfortunately, failure by slipping from the clamp occurs frequently, which reduces the data available and skews the maximal force achieved by failing at the clamp prior to construct failure. The other method described for single load to failure testing involves placing the prosthesis and attaching the suture ends to the load cell.(13) This method of loading mimics the pulley force applied to the LP during suture tying at surgery. While friction at the suture tracts creates an imperfect pulley system and will lower the force required for failure, this is consistent with the in vivo forces. In addition, the speed of loading could alter the stiffness and failure of the construct. A variety of speeds from 20-100mm/second have been reported in the literature, however a value for approximating in vivo conditions for an acute loading situation, such as that occurring during surgery, has not been determined.

Cyclic loading is designed to evaluate the longevity of LP constructs in a method to mimic the physiologic forces in the post-operative period. With the high rate of post-operative loss of abduction, cyclic loading likely provides valuable information regarding the more long-term stability and strength of a given LP technique. Currently, the available research suggests that coughing and swallowing cause an increase in suture tension following LP, likely due to circumferential contraction of the caudal pharyngeal muscles.(12) Cyclic loading of individual laryngeal cartilages in a similar manner to single load to failure testing has been reported numerous times.(91,94) With this method of cyclic loading gradual loss of abduction is unable to be evaluated due to the fixed nature of a single cartilage, so researchers have traditionally performed cyclic testing for a set number of cycles, followed by a single load to failure in order
to compare the constructs. This method of testing has numerous potential factors that prevent translation to an in vivo setting, including the testing of isolated cartilages, the inability to evaluate for loss of abduction, and the performance of cyclic loading followed by an acute high-loading situation. Cyclic testing has also been performed with full laryngeal constructs attached by suture loops to the materials testing machine. (86) This method allows for testing of a complete LP, taking into account interactions between the laryngeal cartilages. The loading in this method occurs on a rostral-to-caudal plane, which may not adequately represent the in vivo loading caused by the contraction of the caudal pharyngeal musculature. Additionally, while the researchers that first reported the technique chose to perform cyclic loading followed by single load to failure, the loss of arytenoid abduction could feasibly be evaluated as the front of the larynx is free in this method. An additional method of cyclic testing involves embedding a complete larynx and LP construct, then utilizing an actuator to depress the arytenoid cartilage axially into the airway. (95) This method of cyclic testing represents a major improvement over the previously described methods to mimic in vivo loading and evaluate progressive loss of abduction. The complete larynx can be tested and the airway is free to evaluate the loss of arytenoid abduction over the cycling period. In addition, depressing the arytenoid cartilage axially, rather than in a more rostral-to-caudal direction, improves the approximation to pharyngeal constrictor muscle contraction, which results in arytenoid loading. However a single point of contact, compared to a wide band of pharyngeal constrictor muscles, may reduce the load distribution over the larynx in situ. Lastly, while the left side could be evaluated for loss of abduction, the right side could not be used simultaneously, which could lead to increased variation and number of samples required compared to a situation in which two LP techniques could be tested simultaneously, one on each hemi-larynx.
A variety of airflow chambers have been developed and used to mimic air flow at exercise in order to test LP constructs.(14,89,90) Intraluminal pressure changes rostral and caudal to a laryngeal specimen are evaluated and compared. These testing methods have been used to evaluate methods for cricoarytenoid joint stabilization or ankylosis, thus allowing for airflow measurements before and after removing sutures to determine the effect of joint stabilization on airway mechanics. While this method allows testing under air flow conditions similar to those that would be encountered in the exercising horse, no external loading occurs with this technique.

Many methods for subjective evaluation of arytenoid cartilage abduction and LP outcome have been described in both in vitro and in vivo studies. The initial grading scale for post-operative arytenoid abduction was developed by Dixon et al, which utilized a 1-5 scale in horses following LP.(71) This grading scale is widely used both clinically and in research settings.(56,74,75) However, this rank scale is somewhat insensitive when evaluating in a research setting due to the wide overlap between cases, and more objective methods to evaluate arytenoid cartilage abduction and its subsequent loss have been described. The cross sectional area of the rima glottidis can be measured in its entirety, or split into left and right sides.(29,57,81,82,96) Some studies using the cross sectional area, particularly those in which a ventriculocordectomy is included in the construct, create the ventral boundary at the top of the vocal fold as the site is a more consistent location to include.(57) Another common method for evaluating arytenoid abduction is the right-to-left quotient angle ratio (RLQ).(29,57,84) This method can be used for in vitro or in vivo studies and uses a ratio measured of the fully abducted right arytenoid compared to the left. A line is drawn along the dorsal-ventral axis of the rima glottidis, then extended by one third further dorsally. Lines are then drawn from the dorsal point
to each arytenoid cartilage, and the angles evaluated as a ratio to compare the left and right sides (Figure 1).
CHAPTER 3
MATERIALS AND METHODS

3.1 – A Priori Sample Size Calculation

A pilot study was performed using a monotonic loading model with 3 standard LP and 3 modified LP constructs, utilizing the previously described clamp method for loading. The modified LP failed at a significantly greater maximal force than the standard LP constructs (234N vs 169N). Our pilot study showed that 10 tests per group in monotonic load to failure would be sufficient to demonstrate a statistical significance difference between laryngoplasty with ≥0.80 power and alpha value of 0.05. A priori sample size calculation was also performed for anticipated results during cyclic loading. Previous studies show a median grade 3 of arytenoid abduction 6 weeks following LP. (56,71,74) This loss of abduction would result in an approximately 50% loss of rima glottidis cross sectional area, which was used as the mean loss for sample size calculation. A more minimal loss of abduction (20%) was assumed for the modified LP. Assuming a variance of 20%, a sample size of 10 tests per group in cyclic loading was found to be sufficient to demonstrate a statistically significant difference between LP methods with ≥0.80 power and alpha value of 0.05. To improve the power of the results and allow for greater statistical confidence 20 tests per construct type (standard vs modified LP) and testing method (monotonic vs cyclic) were used.

3.2 - Sample Collection and Storage

Forty-one grossly normal larynges were collected from horses euthanized for reasons unrelated to the study. Larynges were collected within 4 hours of euthanasia, wrapped in 0.9%
saline soaked gauze, and then stored at -20°C until execution of the study. Specimens were thawed for 16 hours at room temperature (20°C) prior to testing.(13,87)

3.3 - Construct Preparation

Specimens and LP technique (standard or modified) were randomly assigned to testing model (monotonic or cyclic loading) and side (the right and left hemi-larynx), respectively. Randomization at each step was performed by coin flip. The cricoarytenoideus dorsalis muscles were removed bilaterally to facilitate better visualization of the muscular process and similar suture placement between constructs. A 2 to 3 cm incision was made on the cricoarytenoid joint capsule bilaterally prior to suture placement to expose the articular surface and mimic the clinical scenario for surgeons that routinely induce ankylosis of the joint.(14) LP constructs were performed using a standard technique on one side and a modified technique on the other by the same investigator (ES). Standard LP constructs were prepared using a previously described technique.(5) A strand of No. 5 high molecular weight coated polyethylene suture (Fiberwire, Arthrex Inc.) was passed through the cricoid cartilage approximately 2 cm rostral to the caudal rim and 1 cm abaxial to the dorsal ridge of the cartilage. For placement in the arytenoid cartilage, the suture was passed through the muscular process in a caudomedial to rostrolateral direction incorporating approximately 1.5 cm depth of the cartilage. Modified LP constructs were prepared by placing the same type of suture used for the standard LP through the caudodorsal part of the cricoid cartilage as described for the standard LP and through the muscular process and base of the muscular process of the arytenoid cartilage as described below. For the modification the suture was threaded through a commercially available 12 mm titanium button (RetroButton, Arthrex, Inc.) and a 10 mm long tunnel custom-made from a teat cannula (Udder infusion
cannula, Jorgensen Labs). The dorsal aspect of the muscular process was flattened with a #10 scalpel blade, and then a vertical hole was drilled through the center of the muscular process toward the base of the muscular process of the arytenoid cartilage using a 2.7 mm drill bit. The drill was aimed to the point in the cartilage adjacent to the deepest point of the laryngeal ventricle to provide the maximal amount of arytenoid cartilage incorporation. Placing the toggle in the correct position was simple even though the toggle was located under non dissected soft tissues, and appropriate placement was confirmed with dissection on each specimen following testing. Once the hole was drilled through the arytenoid cartilage the prosthesis was passed through, and the suture pulled tight to lock the toggle on the far side of the drill tract (Figure 2). The tunnel was then inserted into the drill tract to provide additional strength to the cartilage.

3.4 - Mechanical Testing

Forty-two LP constructs in twenty-one larynges were tested in monotonic loading until failure, designed to mimic acute loading of the suture, such as at the time of surgery. The LP sutures were passed through the arytenoid and cricoid cartilages and a single surgeon’s throw created. The ends of the prosthetic suture were secured to fixtures attached to the stationary crosshead/load cell and servo-hydraulic actuator of a load frame (Instron 880, Instron Co.) (Figure 3) and loaded at 100 mm/minute until construct failure. Data were sampled at 200 Hz and the load at failure (N) was recorded. The mode of failure (1. suture pull-through or fracture of the muscular process, 2. suture pull-through or fracture of the cricoid cartilage, or 3. suture breakage) was determined after LP construct failure.

Twenty larynges were tested in cyclic circumferential loading, designed to mimic forces exerted on the LP sutures due to swallowing and coughing. Both LP techniques were placed in
each specimen, one in each hemi-larynx, to allow for simultaneous testing. Sutures were tied with a single surgeon’s throw followed by 5 square knots with the arytenoid cartilages fully-abducted. Each larynx was placed in a plastic cradle, and a 2.5 cm wide nylon strap was wrapped around the larynx and cradle. The width of the webbing was chosen to fully cover and engage the muscular process, and was fit to center the apex of the muscular process under the webbing. The nylon strap was cut in the center at the cradle to allow balanced load actuation and the ends of the strap were secured to the load cell and actuator of a servohydraulic load frame (Instron 880, Instron Co.) to create circumferential loading of the larynx (Figures 4 and 5).

An ‘E-buckle’ load-transducer was utilized to determine the loading conditions for the strap that resulted in appropriate LP construct loading. The E-buckle load-transducer was custom-built with similar specifications to a previously reported suture load-transducer. Prior to testing the load-transducer was calibrated with a strand of No. 5 high molecular weight coated polyethylene suture (Fiberwire, Arthrex Inc.). The load-transducer was then placed on the standard LP suture in 3 separate specimens in order to determine the load-frame displacement needed to result in a maximal suture load of 40 to 45 N during cycling, chosen to mimic the maximal post-operative loading of LP sutures during swallowing (Figure 6). A displacement of 25 mm in the load-frame resulted in 40 to 45 N of load on the prosthetic suture. Larynges were loaded for 10,000 cycles at a frequency of 3Hz. Photographs were taken prior to loading and every 500 cycles through the cyclic testing period.

3.5 - Post-Testing Assessment

Photographs obtained from the specimens every 500 cycles were randomized and graded using a previously reported postoperative arytenoid abduction grading system (71) by a blinded...
surgeon (SGN) to evaluate any loss of abduction. Additionally, the rima glottidis cross sectional area of each hemi-larynx was measured using commercial image software (Image J version 1.50b, National Institutes of Health) after calibration of image size using the left corniculate process length. Hemi-larynx cross sectional area was measured to the top of the vocal fold as previously described (Figure 1). (57)

3.6 - Statistical Analysis

Continuous data were evaluated for normality using the Kolmogorov-Smirnov test, kurtosis, and Q-Q plots. Maximal load at failure during monotonic tensile testing was normally distributed and evaluated using an independent sample T-test. Load at failure was expressed as mean ± SD. A χ² test utilizing adjusted residuals (with adjusted residuals ≥ 2 or ≤ -2 determined to contribute to the significance) was used to compare modes of failure during monotonic testing. For cyclic testing, subjective grading scores for loss of abduction were compared between LP techniques with a Kaplan-Meier survival analysis, using a grade ≥3 (74) postoperative arytenoid abduction (according to the Dixon’s grading system) as ‘surgical failure’ in our study. (71) Additionally, hemi-larynx cross sectional area data was evaluated using a repeated measure ANOVA with a Bonferroni post-hoc correction (after log-transformation to allow for parametric testing). A P-value ≤0.05 was considered significant. All statistical analysis was performed using a commercially available statistical software package (IBM SPSS Statistics for Windows version 23.0).
CHAPTER 4
RESULTS

4.1 - Monotonic Load to Failure

Twenty-one larynges were tested in monotonic load to failure. Testing of one of the modified LP constructs resulted in fracture of the cricoid cartilage through the contralateral needle hole of the previously tested standard technique. This data point was not included in the statistical analysis, resulting in 20 modified LP and 21 standard LP constructs. The modified LP constructs failed at a higher load than the standard LP constructs (191 ± 29 N and 91 ± 44 N, respectively, \( P < 0.001 \)) (Figure 7). Nineteen constructs prepared with the standard LP technique failed by suture pulling through the muscular process of arytenoid cartilage, and the remaining 2 constructs failed by the suture pulling through the cricoid cartilage. With the modified LP technique, 12 constructs failed by the sutures pulling through the cricoid cartilage, and in the remaining 8 constructs, the suture slipped from the load-frame clamp. The standard LP constructs failed more often by suture pull-through of the muscular process than the modified LP constructs (\( P < 0.001 \)) (Table 1).

4.2 - Cyclic Loading

Twenty larynges were tested in cyclic loading. Cyclic circumferential loading for 10,000 cycles was successfully completed in all 20 larynges subjected to testing. The modified LP constructs had greater ‘surgical survival’ over 10,000 cycles than the standard LP constructs (Figure 8, \( P < 0.001 \)). We found that 11/20 (55%) standard LP constructs lost abduction to a grade 3, while 0/20 (0%) modified LP constructs reached a grade 3 of arytenoid abduction (Figure 9). In addition, the modified LP constructs lost less cross sectional area compared to the standard LP constructs over 10,000 cycles (\( P < 0.001 \)), with the standard technique losing a
median of 63% (25-75% CI, 12.6 to 80.4%) of the original cross sectional area, compared to a 13% loss (25-75% CI, 4 to 25.6%) with the modified technique over 10,000 cycles.
CHAPTER 5
DISCUSSION

This study demonstrated that a modified LP incorporating a toggle technique to anchor
the suture material to the arytenoid cartilage was biomechanically superior to the standard LP
technique in both monotonic and cyclic circumferential loading of the larynges, both of which
were intended to mimic the forces exerted on the sutures in vivo. The modified LP technique
tested uses a suture button to secure the suture material to the base of the muscular process of the
arytenoid cartilage. We hypothesized that by incorporating more surface area of the arytenoid
cartilage the modification would reduce the occurrence of suture pull-through from the muscular
process of the cartilage, thereby reducing progressive postoperative loss of arytenoid abduction
in horses undergoing LP. This modification also provides a mechanical advantage to the LP by
lengthening the lever arm of the prosthetic suture acting around the cricoarytenoid joint, allowing
more complete and efficient arytenoid abduction. The modified LP technique has been
performed in live horses, and has not proved more difficult than a standard LP to execute in the
authors’ experience. The surgical approach to perform the modified LP technique is similar to
the one described to perform the standard LP technique, including the size of the surgical
incision. Once the muscular process is exposed by transecting the insertion of all the bellies of
the cricoarytenoideus dorsalis muscle, the lateral cricoarytenoid capsule is incised to expose the
articular surface. This step is followed by rotation of the arytenoid cartilage outwards and
drilling of the cartilage toward the base of the muscular process with the drill bit orientated in a
vertical direction. Like any new surgical technique a learning curve is present, but in the hands of
our investigators the modified LP has proved to be of similar difficulty to the standard LP
technique.
Gradual loss of postoperative arytenoid abduction is the most common complication following LP, with up to 90% of horses losing at least one grade (Dixon’s arytenoid abduction grading system) following surgery. (71,72,75) This well documented loss of abduction may partially explain the only fair success rates of the standard LP procedure, particularly in racehorse populations. (49,54,56,71) Typically surgeons attempt to create more abduction of the cartilage than needed at the time of surgery to combat the inevitable loss in the weeks following. (16) Horses with greater abduction at the time of surgery experience a more significant amount of abduction loss in the postoperative period, somewhat negating the benefit of achieving additional abduction at the time of surgery. (75) In addition to the postoperative complications, this method of “planning for loss” minimizes the ability of the surgeon to customize the degree of abduction to the patient in question. However, by incorporating a larger portion of the arytenoid cartilage and eliminating the risk of suture pull-through of the muscular process or breakage of the muscular process, our modified LP technique proved superior in resisting cyclic fatigue compared to the standard LP procedure. By minimizing cyclic fatigue, the modified LP technique may help to reduce much of the speculation in predicting long-term arytenoid abduction in horses undergoing the procedure for the treatment of RLN.

The monotonic and cyclic loading of the specimens used in this study appear to better mimic the in vivo forces placed on prosthetic sutures at the time of surgery and in the postoperative period compared to previously reported methods. Acute, complete loss of abduction due to cartilage failure occurs most frequently in the first week following surgery in up to 10 to 15% of horses undergoing the LP procedure. (12,49,52,54,71) However, this type of failure may also occur at the time of surgery while tightening and tying prosthetic sutures, necessitating a suboptimal prosthesis placement further along the muscular process and often resulting in poor
arytenoid abduction. (16) The monotonic testing used in the present study was designed to mimic acute tightening of the prosthetic sutures, as occurs at the time of surgery or recovery. Unfortunately, this methodology used did not allow for testing of the knot security which could have impacted the results of the monotonic testing. However, this method of monotonic loading appears more reliable than the previously reported clamp method, which tends to result in specimen slippage. Cyclic loading of LP sutures has been attributed to swallowing in the postoperative period. (12) The pharyngeal constrictor muscles reside circumferentially around the equine larynx, and likely contribute to repeated loading of the arytenoid cartilage and larynx, with each swallow resulting in a total load of 40 to 45 N on LP sutures. (12) The cyclic loading model used in this study was designed to mimic circumferential loading of the larynx, in comparison to a single point of loading on the arytenoid cartilage or suture, as has been previously described for similar testing. (86,94,95) Many of the details of pharyngeal constrictor loading of the larynx, specifically the arytenoid cartilage, are unknown, and therefore our model was designed as a best approximation with the current body of knowledge. However, we acknowledge the design of the construct may not fully represent the in vivo forces applied by the pharyngeal constrictor muscles. In addition to improvements in the loading configuration, our cyclic loading model allowed for testing of both hemi-larynges at a time, allowing for an improved head-to-head comparison. Lastly, with having the larynx free it allowed for evaluation of gradual loss of abduction, compared to previous studies which rely on a monotonic loading to failure following cyclic testing to compare methods.

Many of the details regarding signalment of the specimens used in this study were unknown, and therefore conclusions about age regarding LP performance cannot be drawn. Numerous specimens showed varying degrees of ossification of the muscular process of the
arytenoid cartilage, and ossification has been previously documented in older horses. One subjective observation in this study was that ossified cartilages tended to sustain complete pull through or fracture of the muscular process during cyclic loading, more frequently than fully cartilaginous muscular processes. In addition, an adequate suture placement through the muscular process proved more difficult in these specimens due to the ossification. The modified LP, with only requiring a drill bit passage through the muscular process, appeared to result in fewer undergoing complete failure, particularly in ossified cartilages. While older horses are not the typical population undergoing this procedure, the modified LP technique may provide a further improvement over the standard LP in horses with ossified laryngeal cartilages. The presumed distribution in signalment of our specimens could introduce variability in the results, when compared to a more uniform population of young racehorses, which would be more likely to present clinically for LP. However, the differences between the modified and standard LP techniques were consistent across specimens and therefore we feel that this potential variability did not impact our overall conclusion.

Suture choices have been largely left to surgeons preference, with minimal research to evaluate different prosthesis materials in a direct comparison for LP. For this study high molecular weight polyethylene suture was used as it was donated in conjunction with the suture toggles. Additional research comparing different suture types would add substantial knowledge to the body of research and help improve surgeon’s choices when performing LP.

Numerous limitations arise with ex vivo studies when crossing over to an in vivo application. The modified LP technique was simple to perform and did not appear to take a significantly longer period of time to perform, however this was not objectively measured in this study. The ability to perform the technique in a patient is certainly a concern when testing a new
surgical procedure on isolated specimens. To begin to address this concern, we have performed the modified LP technique both in cadaver animals and live patients in addition to ex vivo testing. While new mechanical loading methods were developed and used as part of this study to more closely mimic in vivo conditions, ex vivo results often do not fully replicate the clinical scenario. However, the biomechanical superiority of the modified LP technique noted in this study was significant enough to likely have a substantial benefit over the standard LP technique in a clinical patient. Further studies involving follow-up of the modified LP technique in live horses are planned to evaluate the long-term stability and strength of the implant.
CHAPTER 6
CONCLUSIONS

Based on the results reported in this study, the proposed modification to the LP technique proved to be biomechanically superior to the standard LP technique in both monotonic load to failure and cyclic loading over 10,000 cycles. In monotonic loading the modified LP technique had a significantly higher maximal load at failure. Additionally, the modified LP technique failed either through the cricoid cartilage or by suture failure, compared to the standard LP technique, which tended to fail through the muscular process. Failure of the modified LP technique occurred at supraphysiologic loads in this in vitro model. The modified LP technique also lost significantly less abduction than the standard LP technique over the cycling period. The use of two separate loading models in this study fully characterized the biomechanics of the modified LP technique. These results support the use of the described modified LP technique to reduce the incidence of post-operative failure. Further studies evaluating the long-term arytenoid cartilage maintenance in horses is warranted to ensure the results reported here carry over into an in vivo situation. Ultimately this technique may help to combat the common complication of gradual loss of abduction so frequently noted in horses undergoing prosthetic LP, which may help to improve success rates in the post-operative period.
CHAPTER 7
TABLE AND FIGURES

Figure 1: Photographs showing appropriate measurements for attaining the hemi-larynx cross sectional area (left) and right-to-left quotient angle ratio (RLQ, right). The most frequently used method for evaluating complete or hemi-larynx cross sectional area is shown below, with the lower bound at the level of the top of the vocal fold, which reduces some variability related to visualization below the epiglottis. This method was used for evaluation of hemi-larynx cross sectional area in the present study. For the RLQ a is drawn along the dorsal-ventral axis of the rima glottidis, then extended by one third further dorsally. Separate lines are then drawn to the arytenoid cartilages, and a ratio created (a/b) to compare abduction of the 2 arytenoid cartilages.
**Figure 2:** Photographs documenting the modified LP technique. First the 2.7mm drill bit is passed vertically through the flattened muscular process to exit at the level of the laryngeal ventricle, rostral is to the left of the image (A). The appropriate orientation of the drill bit in the vertical plane is shown in (B). Appropriate toggle placement is at the base of the muscular process to incorporate the maximal amount of cartilage area (C). The left corniculate process is marked with an asterisk.
**Figure 3:** Photographs obtained from the side (A) and top (B) of one of the specimens during monotonic loading of the standard LP technique. Note the surgeon’s throw created on the LP suture and the ends of the suture attached to the clamps of the load frame.
**Figure 4:** Photographs obtained from the front (A) and top (B) of a specimen during cyclic loading of both LP techniques simultaneously. Note the larynx placed in a blue cradle with nylon strap wrapped around the larynx and secured to the load cell and servohydraulic actuator (white arrows) of the load-frame (A). Distraction of the nylon straps resulted in circumferential loading. Also, note the lead wire attached to the ‘E-type buckle’ load-transducer placed on the suture of the standard LP to monitor loading on the prosthetic sutures (arrowhead) (A).
**Figure 5:** Cyclic loading of both the modified (right side of the specimen) and standard LP (left side of the specimen) techniques simultaneously. Photographs obtained from a specimen while unloaded (A) and loaded (B) in a cycle. Both techniques were placed, the specimen mounted in a cradle, and a nylon strap wrapped around the larynx to create circumferential loading.
**Figure 6:** Photograph obtained from a specimen prior to cyclic loading. The rostral aspect of the larynx is on the bottom of the image and the asterisks represent the muscular processes of the arytenoid cartilages. Note the stainless steel ‘E-type buckle’ load-transducer placed on the suture of the standard LP on the left of the image and the modified LP on the right of the image.
Figure 7: Maximal load at failure during monotonic testing of the modified and standard LP constructs. Modified LP constructs failed at a higher load than standard LP constructs (191 N and 91 N, respectively). Bars show the mean load at failure, with standard deviation depicted by lines.
Table 1: Mode of failure during monotonic testing of the modified and standard LP constructs. The standard LP constructs failed significantly more frequently at the muscular process compared to modified LP constructs, which failed only at the cricoid cartilage or the suture ($P < 0.001$).

<table>
<thead>
<tr>
<th></th>
<th>Modified Laryngoplasty</th>
<th>Standard Laryngoplasty</th>
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<tbody>
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<td>Muscular Process Pull-Through</td>
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<tr>
<td>Suture Failure at Clamp</td>
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Figure 8: Kaplan-Meier survival analysis comparing the modified and standard LP constructs, using grade 3 arytenoid abduction as the end event. The standard LP constructs had significantly less survival over 10,000 cycles compared to the modified LP constructs ($P < 0.001$), with 55% of the standard LP constructs cycling to at least a grade 3 abduction over the duration of the testing.
**Figure 9:** Bar graph showing the grades of arytenoid cartilage abduction (using the Dixon grading system) after cyclic loading (10,000 cycles) for the modified and standard LP techniques.
REFERENCES


