

A Comparison of Laryngoplasty and Modified Partial Arytenoidectomy as Treatments for Laryngeal Hemiplegia in Exercising Horses

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Objective—To compare upper airway mechanics, arterial blood gases, and tracheal contamination in horses with induced left laryngeal hemiplegia (recurrent laryngeal neuropathy [RLN]) treated by laryngoplasty/vocal cordectomy (LPVC) or modified partial arytenoidectomy (MPA).

Study Design—Repeated measures under the following conditions: Control, RLN, LPVC, and MPA.

Animals—Six horses.

Methods—Two trials were conducted under all conditions at 80% and 100% of maximal heart rate (HR_{max}). In Trial 1, arterial blood gases, tracheal and pharyngeal pressures, and laryngeal videendoscopy were recorded. In Trial 2, upper airway pressure and airflow were determined. Tracheobronchial aspirates were performed after exercise to quantify airway contamination.

Results—Compared with control, RLN significantly increased inspiratory impedance and worsened exercise-induced hypoxemia. At 80% HR_{max} , LPVC restored most variables to control values. At 100% HR_{max} , LPVC improved all variables, but did not restore minute volume, arterial pH, and $PaCO_2$. At 80% HR_{max} , MPA restored all variables except bicarbonate to control values. At 100% HR_{max} , MPA improved all variables, but did not statistically restore minute ventilation or bicarbonate level. Only minor differences were noted between LPVC and MPA. Both resulted in equivalent tracheal contamination.

Conclusions—Airway mechanics and arterial blood gas values were not restored to normal after either LPVC or MPA in horses exercising at HR_{max} . This does not affect ventilation at sub-maximal exercise, but has clinical implications at HR_{max} . Both procedures diminish normal laryngeal protective mechanisms.

Clinical Relevance—At sub-maximal exercise intensities both LPVC and MPA restore airway ventilation to normal. At maximal exercise the superiority of LPVC over MPA is slight.

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INTRODUCTION

SURGICAL THERAPIES used to treat recurrent laryngeal neuropathy (RLN) include ventriculectomy,

ventriculocordectomy, vocal cordectomy, subtotal arytenoidectomy, partial arytenoidectomy, neuromuscular pedicle grafting, and laryngoplasty.^{1–3} The preferred RLN surgical treatment is laryngoplasty, without or

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with (laryngoplasty/vocal cordectomy [LPVC]) ventriculo-cordectomy.^{2,4-6}

Arytenoidectomy was introduced by Günther in 1845,⁷ but was refined by White and Blackwell in 1980⁸ to a partial arytenoidectomy technique. It has failed to gain popularity as an RLN treatment because it is believed to be inferior to laryngoplasty in restoring airway function. It was clearly demonstrated in experimental horses that partial arytenoidectomy improved, but did not normalize, airway mechanics as assessed by variables derived from flow-volume loops, which significantly contributed to the perception of its inferiority to laryngoplasty.⁹

We propose that dynamic collapse of the unsupported ipsilateral aryepiglottic fold is a significant reason upper airway obstruction still remains after partial arytenoidectomy. In this study, we tested a modification of partial arytenoidectomy (MPA) designed to minimize ipsilateral aryepiglottic fold collapse. The modification included caudal traction and fixation of the left aryepiglottic fold, with any remaining loose ipsilateral tissue removed post-operatively.

Our first hypothesis was the difference between LPVC or MPA procedures to correct RLN would be clinically insignificant in horses at sub-maximal exercise. We further hypothesized that ventilation at maximal exercise would be impaired to a greater degree with both procedures. We also hypothesized that by altering airway protection during swallowing, LPVC and MPA would affect tracheal cytology, suggesting increased tracheal contamination compared with control and RLN status.

MATERIALS AND METHODS

Horses

Six mature horses (4 Thoroughbred and 2 Standardbred; mean age = 4.8 years, range 3–6 years; 5 females, 1 castrated male) with a previously exteriorized right carotid artery¹⁰ were used. Horses were determined to be in good condition and athletically fit on the basis of a physical examination. Before study, normal laryngeal function was confirmed by upper airway videoendoscopy at rest (laryngeal grade I or II)¹¹ and during exercise (laryngeal grade A).¹¹

Training Protocol

Horses were trained 5 days/week on a high-speed treadmill for 3 weeks before each condition trial to adapt them to the exercise protocol and instrumentation system and standardize their fitness level.

Experimental Design

Six horses were tested on a high-speed treadmill for 2 exercise trials at 80% and 100% of their maximal heart rate

(HR_{max}) under 4 sequential conditions: (1) control, (2) transection of the left RLN to induce laryngeal hemiplegia, (3) LPVC and (4) MPA. Horses were rested for a minimum of 3 months after each surgical procedure to allow for a steady-state and complete surgical healing. During Trial 1, the following determinations were made: heart rate (HR), videoendoscopy of the upper airway, stride frequency, arterial blood gas, pharyngeal, and tracheal pressures. During Trial 2, airflow as well as tracheal and mask pressures were measured.

Anesthetic and Surgical Procedures

After xylazine hydrochloride (1.1 mg/kg intravenously [IV]) administration, anesthesia was induced with ketamine hydrochloride (2 mg/kg IV) and maintained with isoflurane in oxygen. An oral endotracheal tube was used except for MPA when a mid-cervical tracheostomy tube was used for inhalant delivery. Horses were positioned in right lateral recumbency for transection of the left recurrent laryngeal nerve and laryngoplasty, and dorsal recumbency for MPA.

Transection of Left Recurrent Laryngeal Nerve

Laryngeal hemiplegia was created by transecting the left recurrent laryngeal nerve as previously described.¹² Two stainless-steel clips were placed across the nerve to prevent regrowth.

Laser vocal cordectomy was performed as previously described.¹³ Briefly, a diode laser (Biolitec Inc., East Longmeadow, MA) was used to remove the left vocal cord under standing sedation and local anesthesia using videoendoscopic guidance. Horizontal cuts were first made at the ventral most point of the cord and just below the vocal process of the arytenoid cartilage with the laser fiber directed abaxially. Then, using bronchoesophageal grasping forceps (Richard Wolfe Medical Instruments Corporation, Vernon Hills, IL), the vocal cord was tensed axially and a vertical incision was made to complete removal.

Laryngoplasty was performed as previously described¹⁴ using 2 strands of #5 polyester suture (Ethibond, Ethilon Inc., Somerville, NJ). Sutures were tied to create 80% (grade 2) abduction¹⁵ using videoendoscopic observation.

Grading of Laryngeal Abduction After Laryngoplasty

Laryngeal movements were recorded in unsedated horses using a flexible video endoscope (Olympus GIF-140, Olympus America Inc., Melville, NY) passed into the nasopharynx through the right ventral nasal meatus at rest within 24 hours, at 2 weeks, and 3 months after LPVC. Still images at full inspiration/abduction induced by nasal occlusion were captured and printed. Printouts were randomized and graded independently by 3 observers unaware of the study design to determine the degree of abduction of the left arytenoid cartilage using the Dixon et al¹⁵ grading scale. The median of the 3 observers' values was assigned to each image.

MPA (Figs 1–3)

Horses were positioned in dorsal recumbency and a mid-cervical tracheotomy was performed for intubation. After

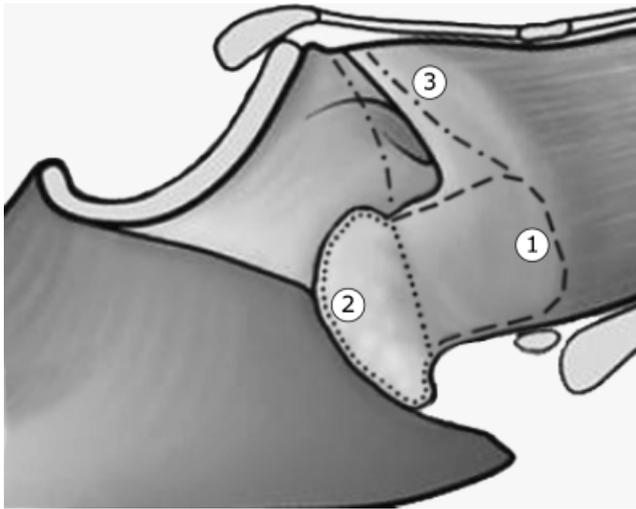


Fig 1. Incisions needed for the modified partial arytenoidectomy procedure. The dashed line (1) indicates the initial incision around the dorsal, caudal, and ventral aspects of the body of the arytenoid. The dotted line (2) represents the incision separating the corniculate process from the body and its abaxial mucosal attachments. The alternating dash/dotted line (3) indicates the incisions required to remove the vocal fold and ventricle.

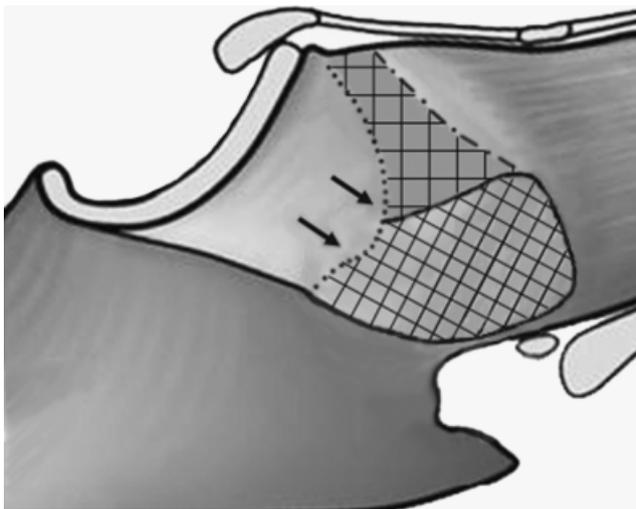


Fig 2. After removal of all arytenoid cartilage except the muscular process, the left aryepiglottic fold was drawn caudally to cover the total defect with diagonal cross-hatching over the cartilage defect and vertical/horizontal cross-hatching over the mucosal defect where the vocal fold and ventricle were removed.

10 cm ventral median laryngotomy, a C-shaped incision was made starting at the dorsal junction of the corniculate process and body of the arytenoid cartilage, extending caudally across the dorsal margin, then tracking ventrally along the caudal edge, and finally rostrally along the ventral border of the body of the arytenoid cartilage (Fig 1). The body of the arytenoid

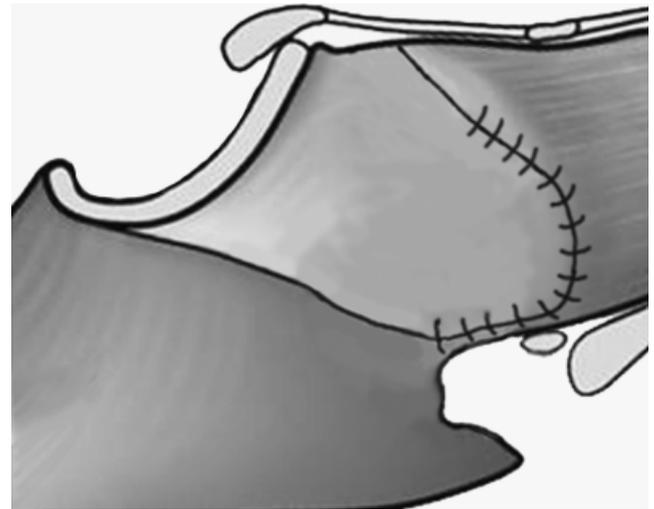


Fig 3. The mucosal defect was closed primarily using the left aryepiglottic fold and leaving the ventral-most 5 mm open to allow for drainage.

was bluntly dissected from its abaxial muscle attachments (ventricularis, vocalis, and cricoarytenoideus lateralis muscles) using a Freer elevator to the level of the dorsal body. Using heavy curved scissors, the junction of the corniculate process and body of the arytenoid cartilage was sharply separated. Using curved Mayo scissors, the corniculate process was separated from the aryepiglottic membrane and removed, as was the remnant of the left ventricle (Fig 2). The arytenoid cartilage, except for the muscular process, was then removed using cartilage scissors. The left aryepiglottic fold was next drawn caudally over the remaining defect and sutured in place using 3-0 poliglecaprone 25 in a simple continuous pattern (Fig 3). The cricothyroid membrane was closed with 0 polyglactin 910 in a simple continuous pattern and the remainder of the incision was left open to heal by second intention. Six weeks after MPA, all 6 horses were evaluated endoscopically under standing sedation with topical anesthesia; any excess ipsilateral tissue along the aryepiglottic fold was grasped with bronchoesophageal grasping forceps and removed using a diode laser.

Grading of Cross-Sectional Area After MPA

A postoperative grading system that paralleled the grading scale of Dixon et al¹⁵ was created to allow subjective assessment of the cross-sectional area of the larynx after MPA. Laryngeal images were recorded, captured, and printed 3 months after MPA. Grading of the cross-sectional area was performed by 3 blinded observers as described above using the grading scheme listed in Table 2.

Postoperative Care

All horses were fed and watered from the ground throughout the study to decrease tracheal contamination expected

after laryngeal surgeries. All horses were administered trimethoprim sulfamethoxazole (30 mg/kg orally every 12 hours) and phenylbutazone (2.2 mg/kg orally every 12 hours) for 5–7 days postoperatively. All horses were examined daily for signs of complications or illness. Horses had 2–3 weeks strict stall rest followed by 8–10 weeks of stall rest with increasing daily hand walking and were then returned to exercise with 3 weeks of training before treadmill testing. All horses had a minimum of 3 months after each surgical procedure before exercise trials.

After laser vocal cordectomy and MPA, horses were administered dexamethasone (0.04 mg/kg IV once daily for 2 days) and 20 mL throat spray solution (glycerin 250 mL, 250 mL dimethyl-sulfoxide [DMSO] 90%, nitrofurazone 500 mL, 50 mL prednisolone [25 mg/mL]) administered through a rubber feeding tube inserted intranasally twice daily for 14 days. After MPA, a tracheotomy tube was maintained for 2 days. It was changed daily and the site was cleaned using moist gauze sponges until healed. Petroleum-based ointment (vitamins A and D ointment, E. Fougera and Co., Melville, NY) was applied around the laryngotomy and tracheostomy to prevent skin excoriation.

Determination of Maximal HR

HR was measured by an on-board HR monitor (Hippocard Systems, Lexington, KY). At time 0, the treadmill was started and accelerated to 4 m/s. At 2 minutes, the treadmill was inclined to a 5% slope. At 4 minutes, the treadmill was accelerated to 6 m/s and kept at that speed for 1 minute. Each subsequent minute, the treadmill was accelerated by 1 m/s until the horse was no longer capable of maintaining its position near the front of the treadmill. HR was regressed against treadmill speed, using the HR for ≥ 3 speeds at sub- HR_{max} . From that regression, speeds predicted to produce 80% and 100% HR_{max} were determined. HR_{max} was used to standardize exercise intensity so comparisons between treatment groups at maximum effort could be made. HR_{max} was determined within 7 days before each trial.

Acclimatization to Treadmill and Training

Horses were shod with flat, aluminum shoes with toe clips. Close-fitting neoprene boots (Professional's Choice, Professional's Choice Sports Medicine Products Inc., Spring Valley, CA) were placed on the lower limbs to prevent injuries from interference. A nylon halter was always used to decrease the chance of breakage. Horses were fasted for 3 hours before any exercise trial.¹⁶ At the end of each workout, horses were bathed and cooled out appropriately.

Exercise Protocols (Trials 1 and 2)

Exercise trials were performed in random order and both included HR, electrocardiogram monitoring, and accelerometer measurements. At time 0, the treadmill was started and accelerated to 4 m/s. At 4 minutes, the treadmill was inclined to a 5% slope. The horses were then exercised on a 5% slope

for 3 minutes sequentially for each speed predicted to produce 80% (sub-maximal exercise) and 100% HR_{max} (maximal exercise). The 80% HR_{max} level was chosen so that conclusions relating to treatment could also apply to horses participating in less physically demanding forms of competition such as showing/hunting.

Airway Mechanics

For Trial 1, 2 Teflon[®] catheters (1.3 mm internal diameter, Neoflon, Cole-Parmer, Chicago, IL) were placed so the ports would lie in the nasopharynx and trachea.^{16,17} Tracheal and pharyngeal catheter positions were confirmed by videoendoscopy. For Trial 2, the tracheal catheter was placed as described above and an additional catheter was placed in the mask to determine mask pressure. This pressure was subtracted from the tracheal pressure during analysis to determine upper airway pressure. Catheters were then attached to differential pressure transducers (Celesco LCVR, Celesco Transducers Products Inc., Canoga Park, CA) referenced to atmospheric pressure and calibrated from -70 to 70 mm Hg (tracheal and pharyngeal) and from -5 to 5 cm of H_2O (mask) using a manometer; catheters were in phase from 1 to 20 Hz.

Airflow was measured by 2 ultrasonic transducers (Birmingham Research and Development Ltd., Birmingham, UK) mounted in the mask, each one in line with one of the nostrils. The ultrasonic transducers were calibrated from -60 to 60 L/s using a rotameter (KDG Flowmeters, West Sussex, UK) and five point calibrations. To determine stride frequency, an accelerometer (Endevco Corp., San Juan Capistrano, CA) was placed securely on the lateral aspect of the left metacarpus. In each horse, a previously exteriorized right carotid artery, (performed before the beginning of the study) allowed aseptic placement of an 18 G, 2.5 in. arterial catheter for arterial blood gas determinations to assess hypoventilation during the 2nd exercise trial. A T-port was placed to introduce a thermistor to measure core body temperature.

Tracheobronchial Aspirate Collection

Samples were aseptically obtained from each horse after it had been cooled out following the HR_{max} determination. This period corresponded to at least 4 hours after last feeding. Horses were restrained in equine stocks and a 3×3 cm square area of hair was clipped from the ventral neck in the mid-cervical region, aseptically prepared, and infiltrated with 2 mL lidocaine hydrochloride. A 5 cm, 14-G needle was inserted between the tracheal rings into the lumen of the trachea through a 5 mm stab incision in the skin over the trachea. A size 5 Fr polyethylene tube was inserted through the needle and advanced ~ 40 cm. Sterile (0.9% NaCl) saline solution (35 mL) was infused and then 10 mL aspirated through the tubing. Aspirates were placed in a tube containing EDTA for cytologic evaluation.

Slides for cytologic evaluation were stained with modified Wright-Giemsa and Gram stains and examined by an individual (D.M.A.) who had no knowledge of the experimental

conditions. Peripheral and central sections of each slide were evaluated and ≥ 300 cells were examined under oil immersion. For each aspirate, the percentages of macrophages, neutrophils, lymphocytes, eosinophils, mast cells, and epithelial cells were determined. Percentages of phagocytes (macrophages, neutrophils) containing intracellular bacteria were also noted.

Data Acquisition

Upper respiratory videoendoscopy was performed on horses exercising on the treadmill using a flexible videoendoscope (Olympus GIF-140, Olympus America Inc., Melville, NY) passed into the nasopharynx through the right ventral nasal meatus, and secured to the horse's halter. Laryngeal activity was recorded using a videotape recorder, (VO 5600, Sony Corporation of America, Park Ridge, NJ). Analog pressure signals (tracheal, pharyngeal, accelerometer) were collected at 256 Hz by a 16-bit A/D data collection board (Data Translation, Marlborough, MA) using a personal computer and LABVIEW software (National Instruments, Austin, TX). The computer software was tailored for this purpose, as well as, for later data analysis.

Arterial Blood Gas Sampling

Three arterial blood samples were rapidly collected in heparinized syringes at the end of each speed interval. Samples were stored on ice and analyzed immediately after the experiment (ABL 30, Radiometer America Inc., West Lake, OH). Blood gases were corrected to body temperature and barometric pressure and mean values for the 3 samples collected during each exercise interval were calculated. The accuracy of blood gas analysis was verified using tonometered equine blood samples.

Data Analysis

Measurements from the last 30 seconds of both the 80% and 100% HR_{max} were analyzed. From Trial 1, tracheal and pharyngeal pressures recorded were used to calculate translaryngeal inspiratory (P_{I}) and expiratory pressures (P_{E}). From Trial 2, mask pressure, tracheal pressure, as well as, peak inspiratory flow (PIF), peak expiratory flow (PEF), tidal volume (V_T), respiratory frequency (f), inspiratory and expiratory time (T_I and T_E) and total breath time (T_{TOT}) from the airflow trace were measured. From those measurements, upper airway inspiratory (Z_I) and expiratory (Z_E) impedance and minute volume (V_E) were calculated. From the accelerometer trace, the gait frequency (G_f) was calculated and reported as a ratio of the respiratory frequency (f/G_f).

Statistical Analysis

A Shapiro-Wilk normality test was performed on continuous variables. Variables were normally distributed except for f/G_f , f , T_I , T_E and T_{TOT} . To determine the correlation between laryngeal grade after LPVC or MPA and inspiratory imped-

ance (Z_I), a Spearman rank correlation was done. To determine the effects of airway conditions (control, RLN, LPVC, MPA) on the various Gaussian variables, 2-way analysis of variance (ANOVA; blocked on horse and the testing condition) followed by Tukey's post hoc comparisons was performed at each exercise intensity. For each of the 5 non-Gaussian variables and the percentage of the respective cells from the tracheobronchial aspirate cytologies, a Friedman 2-way ANOVA (also blocked on horse and testing the condition) followed by repeated Wilcoxon's signed-rank test adjusted for multiple comparisons was done to assess significance at each exercise intensity (when relevant). The level of significance was set at $P = .05$ with Bonferroni's correction for multiple comparisons.

RESULTS

All 6 horses completed all phases of the experiment.

Endoscopic Observations

Horses had a resting laryngeal grade of I or II for control. Laryngeal grade IV was observed after RLN. Several horses coughed and aspirated feed material into the trachea after LPVC and MPA during the immediate postoperative period. Immediately after LPVC, 4 horses developed moderate-to-severe cough with occasional feed material noted at the nostrils while eating. This lasted from several days or for the entire LPVC phase of the study. After MPA, 2 horses developed occasional coughing while eating 4 weeks after surgery. All horses were monitored daily postoperatively for evidence of fever and elevated respiratory or HR. Three months after surgery, videoendoscopy documented feed material in the trachea of 4 horses after LPVC and 2 horses after MPA. All horses were able to successfully complete both treadmill exercise trials under each condition with normal recovery after exercise.

Temporal grading of arytenoid abduction after LPVC (Table 1) revealed a decrease in the extent of arytenoid cartilage abduction when laryngeal grades 24 hours and 3 months after LPVC were compared. Only 1 horse had no decrease in laryngeal abduction grade throughout the 3 month LPVC phase. Based on scoring by 3 independent, blinded observers, the other 5 horses decreased a median of 2 grades¹⁵ in the 3 months from immediately after surgery to treadmill examination. There was no effect of laryngeal grade on Z_I at 3 months.

After MPA, based on scoring by 3 independent, blinded observers, the median cross-sectional area was grade 3 (minimum 1, maximum 3; Table 2) at treadmill examination. Four horses were graded as a moderate cross-sectional area (grade 3); 1 horse each had grade 2 and grade 1 cross-sectional areas. There was no effect of cross-sectional area on Z_I at 3 months.

Table 1. Postoperative Laryngeal Grade¹⁵ in Six Horses After a Laryngoplasty and Laser-Assisted Vocal Cordectomy and Associated Inspiratory Impedance when Exercised on a High-Speed Treadmill at Maximal Exercise Intensity (100% HR_{max})

Horse	Postoperative Laryngeal Grade			Impedance at 100% HR _{max} (mm Hg/L/s)
	24 Hours	2 Weeks	3 Months	
2	2	4	5	0.75
3	2	3	3	0.54
4	2	4	5	0.55
5	2	4	4	0.56
6	1	2	2	0.82
7	2	2	2	0.31

Grade 1, close to or at maximal abduction (80–90° to the sagittal plane); Grade 2, a high degree of arytenoid abduction (50–80° to the sagittal plane); Grade 3, a moderate degree of abduction (45° to the sagittal plane); Grade 4, a slight degree of abduction (slightly more than normal resting position); Grade 5, no detectable arytenoid abduction present.

Table 2. Laryngeal Grades¹⁵ of Cross-Sectional Area in Six Horses, 3 Months After Modified Partial Arytenoidectomy and Laser Resection of Loose Ipsilateral Tissue and Associated Inspiratory Impedance when Exercised on a High-Speed Treadmill at Maximal Exercise Intensity (100% HR_{max})

Horse	Laryngeal Cross-Sectional Area Grade	Impedance at 100% HR _{max} (mm Hg/L/s)
2	3	0.77
3	3	0.81
4	3	0.77
5	3	0.72
6	1	0.82
7	2	0.26

Grade 1 (Excellent), cross-section of the affected side meets or exceeds the fully abducted unaffected side (equivalent to a laryngoplasty with 80–90° abduction); Grade 2 (Good), cross-section of the affected side is equivalent to a laryngoplasty with 50–80° abduction; Grade 3 (moderate) cross-sectional of affected side is equivalent to a laryngoplasty with 45° of abduction; Grade 4 (Slight) cross-section of affected side is equivalent to a laryngoplasty resulting in abduction approximating the normal resting position; Grade 5 (Poor) cross-section of the affected side is equivalent to a laryngoplasty resulting in no detectable arytenoid abduction.

HR_{max}, maximal-heart rate.

Respiratory Variables

Compared with control values, RLN, LPVC, and MPA significantly altered a number of respiratory variables with a greater number of abnormalities noted at 100% HR_{max} than at 80% of HR_{max} (Table 3). These altered mechanical variables did not influence hypoxemia as assessed by arterial blood gases until horses were exercising at 100% HR_{max} (Table 3).

At sub-maximal exercise, 3 horses in the control group had their respiratory rate and gait entrained 1:1 and 3 horses were entrained at 1:2. After RLN, 2 horses had

respiration and gait entrained 1:1, 3 horses were entrained at 1:2, and 1 at 1:1.66. After LPVC and MPA, the 3rd and 4th horses, respectively, were entrained 1:1 with the remainder 1:2. Statistically, no significant difference was found between conditions and respiratory:gait coupling.

The speed at which horses obtained HR_{max} was significantly lower in horses with RLN (mean 10.8 m/s) compared with control (11.7 m/s, $P = .0029$). At sub-maximal exercise compared with control, RLN resulted in a significant increase in Z_I ($P = .022$) and a decrease in V_E ($P = .001$). No differences in inspiratory, expiratory, or total breath duration were noted at either exercise intensity. No effect on blood gases was noted after RLN.

At maximal exercise, 4 horses had respiratory rate and gait entrained 1:1 and 2 horses were unlinked at 1:2 and 1:3, respectively. No change in respiratory:gait coupling was noted in the trials, except the horse that was entrained 1:2 became entrained 1:1 during the LPVC trial. Statistically, no significant difference was found between conditions and respiratory:gait coupling. At maximal exercise, RLN led to a significant reduction in minute ventilation compared with control ($P = .0021$). Neither LPVC nor MPA returned V_E to control values. Compared with control, RLN significantly decreased V_T ($P = .0128$) and PIF ($P = .0355$), increased P_{ui} ($P = .0053$) and Z_I ($P = .0157$) and worsened exercise-induced hypoxemia ($P = .0128$). Both LPVC and the MPA improved these values toward normal, as they were no longer different than control. However, it is more accurate to state that most values were not truly returned to normal because many variables (V_T , Z_I , PIF, and PaO_2) were still not significantly different from RLN. After LPVC, the adjusted mean value was closer to control values than after MPA and P_{ui} was returned to normal while being significantly different from horses with RLN only after LPVC. No effect of RLN was found on variables during exhalation.

Tracheobronchial Aspirate Cytology

For analysis of cytologic data (Table 4), the level of significance set using Bonferroni's correction was $P < .017$. When RLN was compared with control, there was an increase in the percentage of macrophages with intracellular bacteria ($P = .0312$) but no differences were noted in other cellular distributions. Compared with control values, LPVC resulted in a significant increase in the overall percentage of neutrophils ($P = .016$) and a significant decrease in the overall percentage of macrophages ($P = .016$) but macrophages containing bacteria were increased ($P = .016$). Compared with control values, MPA resulted in a significant decrease in the percentage of macrophages ($P = .016$) but an increase in macro-

Table 3. Effect of Laryngeal Condition on Measured and Calculated Variables for Six Horses with Surgically Induced Left Laryngeal Hemiplegia: Before (Control), After Induction of Laryngeal Hemiplegia, After Laryngoplasty and Vocal Cordectomy (LPVC), and Finally After Modified Partial Arytenoidectomy (MPA) During Exercise at 80% and 100% of HR_{max}

Variable*	80% HR _{max} Mean (Range)†				100% HR _{max} Mean (Range)†			
	Control		Laryngeal Hemiplegia		Control		Laryngeal Hemiplegia	
	Control	Laryngeal Hemiplegia	LPVC	MPA	Control	LPVC	MPA	
V _E (L/min)	1013 (763–1311) ^a	793 (639–1115) ^b	948 (761–1152) ^{ab}	888 (590–1086) ^{ab}	1293 (1200–1500) ^a	1128 (930–1400) ^b	1099 (890–1300) ^b	
V _T (L/ breath)	13 (9–16)	11 (9–13)	12 (9–15)	12 (9–16)	14.5 (10.0–26.3) ^a	12.5 (7.8–22.6) ^{ab}	12.5 (7.9–23.5) ^{ab}	
F (breaths/min)	81 (60–110)	54 (54–110)	84 (54–110)	78 (48–110)	120 (48–130)	120 (42–130)	120 (42–120)	
f/G _r	0.8 (0.5–1)	0.7 (0.5–1)	0.8 (0.5–1)	0.8 (0.5–1)	0.8 (0.3–1)	0.9 (0.3–1)	0.8 (0.3–1)	
Z _I (mmHg/L/s)	0.29 (0.21–0.39) ^a	0.46 (0.37–0.63) ^b	0.35 (0.26–0.45) ^{ab}	0.35 (0.27–0.49) ^{ab}	0.46 (0.35–0.57) ^a	0.59 (0.31–0.82) ^{ab}	0.69 (0.26–0.81) ^{ab}	
Z _E (mmHg/L/s)	0.46 (0.20–0.87)	0.52 (0.30–0.80)	0.41 (0.26–0.61)	0.35 (0.24–0.63)	0.38 (0.26–0.64)	0.40 (0.18–0.71)	0.38 (0.20–0.72)	
T _I (seconds)	0.42 (0.26–0.64)	0.51 (0.28–0.66)	0.43 (0.26–0.67)	0.46 (0.28–0.78)	0.34 (0.22–0.68)	0.35 (0.21–0.72)	0.41 (0.23–0.72)	
T _E (seconds)	0.38 (0.24–0.49)	0.42 (0.24–0.52)	0.41 (0.26–0.61)	0.40 (0.25–0.58)	0.35 (0.23–0.63)	0.34 (0.22–0.66)	0.39 (0.24–0.63)	
T _{TOT} (seconds)	0.80 (0.52–1.05)	0.93 (0.53–1.16)	0.84 (0.54–1.27)	0.86 (0.53–1.24)	0.70 (0.48–1.31)	0.70 (0.48–1.37)	0.80 (0.49–1.35)	
PIF (L/s)	–43 (–54.7 to –32.6)	–35 (–49.4 to –22.2)	–39 (–53.1 to –26.7)	–35 (–52.7 to –23.4)	–59 (–67.5 to –46.8) ^a	–49 (–60.8 to –33.7) ^{ab}	–48 (–60.5 to –36.8) ^{ab}	
PEF (L/s)	48 (35.5–65.2)	43 (31.9–56.4)	48 (40.5–63.2)	47 (37.7–53.4)	62 (51.7–66.3)	62 (46.1–82.1)	59 (48.8–65.5)	
Pui (mm Hg)	–4 (–1 to –10)	–6 (–2 to –18)	–4 (–2 to –8)	–6 (–2 to –15)	–13 (–9 to –17) ^a	–17 (–6 to –33) ^a	–23 (–14 to –33) ^{ab}	
Pue (mm Hg)	3 (1–5) ^a	8 (4–16) ^{ab}	9 (5–23) ^b	6 (3–9) ^{ab}	5 (0–10)	11 (6–25)	7 (5–11)	
Speed (m/s)								
pH	7.44 (7.39–7.47)	7.44 (7.42–7.47)	7.44 (7.39–7.48)	7.45 (7.40–7.51)	11.7 (10.4–12.5) ^a	11.7 (9.8–12.4) ^{ab}	11.3 (9.7–12.5) ^{ab}	
PaO ₂	86 (73–94)	84 (68–100)	84 (67–99)	89 (69–105)	73 (64.6–82.9) ^a	66 (60.2–69.3) ^{ab}	69 (61.0–73.1) ^{ab}	
PaCO ₂	37 (34–40)	38 (34–44)	39 (34–43)	39 (36–44)	43 (38.1–47.9) ^a	50 (40.2–59.4) ^{ab}	47 (43.1–54.2) ^{ab}	
HCO ₃	25 (24–26) ^a	26 (24–28) ^{ab}	26 (24.7–28.0)	27 (25.3–28.9) ^b	17 (13.5–20.3) ^a	20 (15.6–24.3) ^{ab}	22 (19.4–27.5) ^b	

*Pui, translaryngeal inspiratory pressure; Pue, translaryngeal expiratory pressure; PIF, peak inspiratory flow; PEF, peak expiratory flow; V_T, tidal volume; f, respiratory frequency; T_I, inspiratory time; T_E, expiratory time; T_{TOT}, total breath time; Z_I, upper airway inspiratory impedance; Z_E, upper airway expiratory impedance; V_E, minute volume; HR_{max}, maximal heart rate.
 †Adjusted mean for all variables except f/G_r, f, T_I, T_E, and T_{TOT} where the arithmetic mean was used.
 Means that are significantly different are noted with different letters.
 Data are expressed as adjusted mean (general analysis of variance [ANOVA]) or mean (Friedman ANOVA) (min–max)

Table 4. Cytologic Assessment of Tracheobronchial Fluid Aspirate Obtained 30 Minutes After Exercise (HR_{max}) in six Horses with Upper Airway Status: Control, After Induction of Laryngeal Hemiplegia (RLN), After LPVC, and After MPA.

Variables	Median (Range)			
	Control	RLN	LPVC	MPA
Macrophage	69 (60–93)	67 (31–91)	49 (27–73)*	51 (37–66)*
Macrophages†	14 (4–30)	30 (25–77)	37 (31–64)*	54 (32–86)*
Macrophages‡	64 (58–90)	30 (3–63)	32 (5–39)	26 (2–52)*
Neutrophil	13 (3–28)	23 (1–54)	38 (11–51)*	24 (14–33)
Neutrophils†	1 (0–4)	3 (2–17)	9 (3–50)	10 (1–18)*
Neutrophils‡	16 (6–23)	8 (2–38)	9 (1–39)	12 (2–25)
Lymphocyte	5 (1–19)	7 (2–15)	7 (2–28)	19 (8–32)*
Eosinophil	0 (0–10)	0 (0–1)	0 (0–2)	0 (0–2)
Mast cell	0 (0–1)	0 (0–0)	0 (0–0)	0 (0–1)
Squamous Epithelial	0 (0–1)	0 (0–1)	0 (0–34)	2 (0–10)
Columnar	3 (0–8)	2 (0–5)	1 (0–6)	4 (0–11)

Results are expressed as percentages.

RLN, recurrent laryngeal neuropathy; LPVC, laryngoplasty and vocal cordectomy; MPA, modified partial arytenoidectomy; HR_{max} , maximal-heart rate.

†With intracellular bacteria.

‡Without intracellular bacteria.

*Significant difference at the $P \leq .017$ level (Bonferroni's correction).

Data are expressed as median (min–max).

phages containing bacteria ($P = .016$). MPA also resulted in an increased percentage of neutrophils ($P = .031$), specifically those containing bacteria ($P = .016$) as well as increases in the number of lymphocytes ($P = .016$) and squamous cells ($P = .031$).

DISCUSSION

Study Limitations

In interpreting the data, one should consider limitations of our study. First, performing MPA after LPVC is a weakness in the study design, because these 2 treatments are not likely to be independent. Some LPVC effects were probably superimposed on MPA effects. For example, stabilization of the muscular process by laryngoplasty might have improved the effect of the arytenoidectomy on airway mechanics. This effect is thought to be small, because only a small area of the arytenoidectomy may have been stabilized. In addition, tracheal aspiration caused by LPVC might have worsened tracheobronchial variables evaluated after MPA. Doubling the sample size would have removed this confounding effect, but would have added the confounding effect of individual horse variation (i.e. horses with same cross-sectional area of the larynx do not have the same athletic performance⁴ and reduction of sounds¹⁸). However, any additive effects would be expected to worsen the variables

measured in this study for MPA instead of improving them. The exercise intensity at which a horse works is another important factor to consider. As observed in our study, conclusions for horses exercising at 80% HR_{max} are different than 100% HR_{max} . This is consistent with the well-known higher success rates after surgery in non racehorses compared with racehorses.^{2–6,19} In this as well as other studies, we used HR_{max} to standardize exercise intensity. We hypothesize from clinical experience that caution should be exercised when comparing data from experimental horses exercising at 100% HR_{max} (usually 10–12 m/s) to racehorses galloping at 18 m/s.²⁰

Respiratory Mechanics and Ventilation

The increased Z_I and decreased P_{ui} associated with RLN were consistent with earlier reports after RLN.^{21,22} In those studies, Z_I and P_{ui} were restored to normal by laryngoplasty (with or without ventriculectomy/ventriculo-cordectomy) at both sub-maximal^{21,22} and maximal exercise intensity.²³ Thus, LPVC returned upper airway function to normal based on airway mechanics data, which is surprising given that the laryngeal cross section is not restored to normal after laryngoplasty.²⁴ In our study, Z_I , but not V_E , was restored to normal with LPVC. It is difficult to reconcile this different finding with previous reports. One possible explanation is a different method was used to measure airflow. Indeed, we used an ultrasonic flow meter to measure airflow, whereas previous studies^{21,22} determined pressure changes across a pneumotachograph. Perhaps the pneumotachograph causes markedly more CO_2 rebreathing, which would lower f and V_E in control horses to a degree that a reduction in V_E would not be detected in the small sample size typical for these types of studies. Given that both methods were calibrated, it seems unlikely there was a direct effect from the measurement system. An alternative explanation may be different breathing frequencies. For example, the mean f was 120 ± 30 breaths/min at maximal exercise intensity in our study versus 77 ± 20 breaths/min.²³ Perhaps at lower f , horses with partial airway obstruction (as seen by inspection after a laryngoplasty) can still maintain peak flow normally.

Arterial blood gases corrected for body temperature may be sensitive indices of impaired ventilation. At maximal exercise, defined as maximal heart and respiratory frequency rate and VO_{2max} , induction of laryngeal hemiplegia resulted in significant worsening of arterial hypoxemia and hypercarbia compared with the control state.^{10,25,26} Christley et al²⁷ reported that minimum PaO_2 and maximum $PaCO_2$ were significantly increased in horses with laryngeal hemiplegia. In our study, arterial blood gases were not affected at sub-maximal exercise, supporting the view that the reduced laryngeal cross-

sectional area seen in hemiplegic horses does not compromise pulmonary ventilation or impact arterial blood gas exchange. However, changes in arterial blood gases are measurable and significantly different from control values at maximal exercise. LPVC did not restore those values to control levels, suggesting that even though Z_I was restored to normal, ventilation was not restored to normal as suggested by V_E measurements. Our results support previous work that determined laryngoplasty improved,^{10,26} but failed to return arterial blood gas values to normal. However, these results must be interpreted with caution, as abnormal ventilatory variables after laryngoplasty may be a direct result of reduced cross-sectional area or indirectly from pulmonary inflammation associated with postoperative tracheal aspiration.

The decrease in PIF and inspiratory tracheal pressure and the increased Z_I after RLN, which was surgically created agrees with earlier studies.^{21,22} Furthermore, normalized Z_I after LPVC is consistent with the findings in those studies. A residual decrease in V_E after MPA and LPVC suggests that airway mechanics have not been fully restored to normal, although improvements are greater after LPVC.

Comparison of LPVC and MPA

Although no significant differences could be detected when comparing horses treated with LPVC or MPA, this could result from the small sample size ($n = 6$) affecting the power of our observations. Indeed, Z_I remained elevated after MPA, thus MPA probably does not improve airway patency as well as LPVC, a finding which has been previously reported.⁹ Yet, the overall difference between treatments was small. The potential for confounding effects of performing the MPA procedure in horses that had already undergone the LPVC procedure must also be considered. Clearly further study of this subject is warranted.

The significant decrease in postoperative laryngeal grade after LPVC observed over the 3-month period is consistent with results reported by Dixon et al.¹⁵ After MPA, the initial postoperative swelling reduces the cross-sectional area of the larynx as seen videoendoscopically. Three months after MPA surgery, the final cross-sectional area of the larynx as graded suggests the final appearance of the larynx is more consistent after MPA than after LPVC.

Airway Protection

Considering morbidity, both procedures interfere with the normal protective mechanisms of the larynx. Using quantitative evaluation of tracheal wash fluid cytology, we demonstrated that both procedures lead to airway

contamination over normal or laryngeal hemiplegic states. There were no significant differences detected in the cellular response or intracellular bacteria in phagocytes, suggesting similar contamination after either LPVC or MPA. The lack of a significant difference between LPVC and MPA is likely because the difference between procedures is small; there is more variation in the post-operative degree of abduction after LPVC compared with MPA and the study's small sample size led to a reduced power of observation.

In summary, both LPVC and MPA improve airway mechanics in induced RLN; LPVC is superior in many regards, but neither technique normalized all aspects of ventilation. Because the difference between the treatments is not large, other factors should also be considered like length of time before returning to work, costs for additional procedures like laser revisions, surgeon's preference, and reversibility of procedure.

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