



Laryngeal reinnervation in the horse

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Idiopathic left laryngeal hemiplegia (LLH) is a common cause of poor performance in equine athletes [1] and has been defined as a peripheral axonopathy [2]. Laryngeal hemiplegia results in an inability of the horse to maintain abduction of the arytenoid cartilage under maximal exercise conditions [53]. Consequently, affected horses have airway obstruction, produce an abnormal inspiratory noise, and have a reduced ability to sustain maximal exercise intensity [3,4].

Diagnosis of LLH is routinely made by endoscopic examination of the larynx. A variety of numeric grading systems have been established to help describe the different degrees of paresis or paralysis observed in the resting horse, and a four-grade system is most commonly used [51]. Treadmill studies have clarified that using this system, grade 1 and 2 laryngeal function does not result in laryngeal collapse, whereas most horses with grade 3 and all horses with grade 4 have airway obstruction during strenuous exercise [6]. Indeed, Thoroughbred yearlings with grade 1 or 2 laryngeal function perform better as adults than yearlings with grade 3 or 4 laryngeal function [7]. Most investigators now agree that grade 1 and 2 laryngeal function is normal, whereas grade 3 and 4 laryngeal function results in impaired performance.

In LLH-affected racehorses with grade 3 (showing laryngeal collapse during strenuous exercise) or grade 4 laryngeal function, surgical intervention is necessary. Surgical techniques that have been used for treatment of laryngeal hemiplegia include ventriculectomy, prosthetic laryngoplasty (often combined with ventriculectomy), and arytenoidectomy [4]. Several studies have evaluated surgical success rates using measures of

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racing performance. These studies have been complemented by treadmill studies and quantitative evaluation of upper airway function before and after surgery [8–10].

A review of treatments for laryngeal hemiplegia indicates that the prosthetic laryngoplasty is the most widely accepted treatment [4]. Estimations of success for prosthetic laryngoplasty have ranged from as low as 44% [5] to as high as 87% [11]. This wide variation in reported success is a result of different methods of defining success. In one of the largest follow-up reports on prosthetic laryngoplasty, it was found that 77% of horses raced at least once after surgery. The same study also found that 56% of horses that had raced before surgery had improved racing performance after surgery [12]. In a smaller but more recent review, 94% of horses raced at least once after surgery, 60% won at least one race after surgery, and of those horses that had raced before surgery, 45% showed improved performance based on race times [13].

Many frustrating complications can accompany prosthetic laryngoplasty, and at least 16 different adverse effects have been recorded in the literature, with the most common one being coughing associated with aspiration of food during swallowing [14]. For this reason, research is still being conducted into technique improvements, including assessment of new prosthetic materials [15] and the use of cordopexy [16]. Despite the potential complications, prosthetic laryngoplasty is still the most popular technique to treat LLH, and ventriculectomy is often performed at the same time.

Because of the potentially serious complications associated with prosthetic laryngoplasty, there has been interest in a more physiologic repair. LLH is caused by dysfunction of the recurrent laryngeal nerve; therefore, laryngeal reinnervation has received considerable attention. Laryngeal reinnervation has been successfully used in people with impaired laryngeal function. Early research into the procedure in Standardbreds demonstrated that there was potential for this surgery to allow LLH-affected horses to return to their previous levels of performance [17]. The advantage of laryngeal reinnervation is that there is no alteration of the laryngeal architecture, which significantly reduces the potential for many of the complications experienced with prosthetic laryngoplasty. This article reviews laryngeal reinnervation, describes the results of 146 LLH-affected horses treated with laryngeal reinnervation, and discusses ongoing research aimed at improving the outcome of laryngeal reinnervation in the horse.

Reinnervation of a skeletal muscle

Reinnervation experiments have taken place in a number of species to identify the best method of returning function to a skeletal muscle after nerve damage. The three main methods tested are nerve anastomosis of dissimilar nerves [18], implantation of a transected nerve end into muscle

[19], and the nerve muscle pedicle graft [20]. The latter two techniques have the best success rates. The nerve muscle pedicle graft is considered the better technique, because muscle activity, as determined by electromyography, can be detected as soon as 8 weeks after surgery [21]. This rapid return of function is thought to be a result of the transplantation of motor end plates in the nerve muscle pedicle. Once fibers of the recipient muscle and pedicle graft heal, propagation of depolarization from the pedicle to the denervated muscle is possible [22]. It has also been shown that any transected axons present in a nerve muscle pedicle can aid rapid reinnervation by axonal sprouting, because there is minimal distance for regenerating axons to traverse to reach recipient muscle fibers [23]. The regenerating axons can grow to the original end plate sites of the recipient muscle or form new end plates [24].

Laryngeal reinnervation

Experiments in laryngeal reinnervation were reported as early as 1946 [25]. The most common reason for laryngeal paralysis in people is nerve damage subsequent to thyroidectomy [26]. Head and neck trauma resulting from automobile accidents or tumor removal is also a cause of uni- or bilateral laryngeal paralysis. Early attempts at reinnervation investigated nerve anastomosis of the vagus and recurrent laryngeal nerves [18], recurrent laryngeal nerve implantation [27], recurrent laryngeal nerve section and anastomosis [28], and phrenic nerve implantation [29,30]. This last technique met with some success, and further studies involving splitting the phrenic nerve to preserve diaphragm function took place, but a period of 6 to 8 months was necessary for return of laryngeal function [31]. In 1973, using the ansa hypoglossi nerve and sternohyoid muscle, early signs of laryngeal movement could be detected between 6 and 12 weeks after surgery [21]. Even in muscle that had been denervated for 6 months, it was possible to demonstrate reinnervation using the nerve muscle pedicle graft [32].

Human laryngeal reinnervation

Clinical use of laryngeal reinnervation was first reported in 1976, when five patients with bilateral laryngeal paralysis were treated using the nerve muscle pedicle graft technique [33]. Three of these patients no longer required tracheostomy tubes to breathe as a result of the surgery. Subsequent to this, further reports of rapid reinnervation using the nerve muscle pedicle graft in people appeared in the literature [34,35,52]. The procedure has also been used in unilateral paresis and paralysis to improve voice quality. Success rates of up to 90% in cases of unilateral paralysis and up to 80% in cases of bilateral paralysis have been reported [36,37]. In cases where there was apparent failure of the technique, investigation often revealed poor patient selection, failing to identify other upper respiratory

tract dysfunction before surgery, and poor postoperative assessment of vocal cord movement [38,39]. There is no question that similar causes of apparent surgical failure can be identified in equine laryngeal reinnervation surgery.

Equine laryngeal reinnervation

The first studies on laryngeal reinnervation in horses were reported in 1989 [40–42]. The nerve muscle pedicle graft, nerve implantation, and nerve anastomosis techniques were all investigated in ponies. In these experimental ponies, the recurrent laryngeal nerve was transected at the time of reinnervation surgery. Although the first two techniques demonstrated histologic evidence of reinnervation, minimal endoscopic evidence of arytenoid function was observed. The nerve anastomosis technique demonstrated evidence of arytenoid movement at rest, but the authors concluded that laryngeal function was insufficient to allow for maximal exercise. Importantly, this study identified that the omohyoideus muscle is an accessory muscle of respiration and is therefore suitable for use as a donor muscle along with its innervation: the first or second cervical nerves. Reinnervation has also been attempted using a muscle pedicle graft created from the right cricoarytenoideus dorsalis CAD muscle. It was hoped that muscle-to-muscle neurotization would result in return of function to the paralyzed left CAD muscle; however, this attempt was unsuccessful [43].

In 1990, the nerve muscle pedicle graft technique using the first cervical nerve and omohyoideus muscle was evaluated on experimentally induced cases of left laryngeal hemiplegia in Standardbreds [17]. In that study, histologic evidence of reinnervation was demonstrated [44] as was a return of laryngeal function, verified by upper airway flow mechanics studies [17]. This study demonstrated that the paralyzed CAD muscle could be reinnervated and that upper airway function could return to baseline levels in vigorously exercising horses between 6 and 12 months after surgery.

Since 1991, we have used the nerve muscle pedicle graft technique in selected clinical cases of laryngeal hemiplegia and hemiparesis in 129 Thoroughbred, 10 Standardbred, and 7 warmblood horses. A description of the surgical technique, postoperative care, complications, and follow-up results follows.

Surgical technique

The nerve muscle pedicle graft is performed with the horse under general anesthesia and in right lateral recumbency. An incision is made along the ventral border of the linguofacial vein, and careful blunt dissection is then carried out to identify the first cervical nerve that travels over the lateral aspect of the larynx. There is often a branch of the linguofacial vein that requires ligation to allow access to the nerve. The surgical procedure

requires accurate dissection of the left first cervical nerve as it passes over the lateral aspect of the larynx to where it meets the omohyoideus muscle, an accessory muscle of respiration [40]. The first cervical nerve usually branches into two or three main branches, which are then followed to their point of insertion into the omohyoideus muscle. The main body of the first cervical nerve should be freed of connective tissue as far proximally as possible. This makes repositioning of the nerve over the dorsal aspect of the larynx easier when the pedicles have been created. Often, the cervical lymph nodes are quite large in young horses and can increase the difficulty of dissection of the distal nerve and its branches. Studies using a cholinesterase stain have demonstrated that there are large numbers of motor end plates present in the pedicle grafts used in the horse (IC Fulton, BVSc, MS, MACVS, RF Slocombe, BVSc, PhD, unpublished data) (Fig. 1). Once the points of insertion of the nerve branches are identified, the area is flooded with local anesthetic. This removes the vigorous contraction of the omohyoideus muscle when the pedicle is created, thus avoiding excessive trauma to the fine nerve-muscle interface. A small block of muscle is removed from the omohyoideus muscle with the fine branch of the first cervical nerve attached. In clinical cases, up to five pedicles may be created for transplantation, whereas in the research horses, only a single branch was used. The resulting deficit in the omohyoideus muscle is usually sutured with a cruciate suture of 2-0 polydioxanone to reduce hemorrhage. Any branches of the first cervical nerve that have to be transected to allow repositioning of the nerve are cut as long as possible to allow them to be used as donor nerves as well. The use of these transected nerve ends is based on studies demonstrating that reinnervation from axonal sprouting using nerve implantation is also

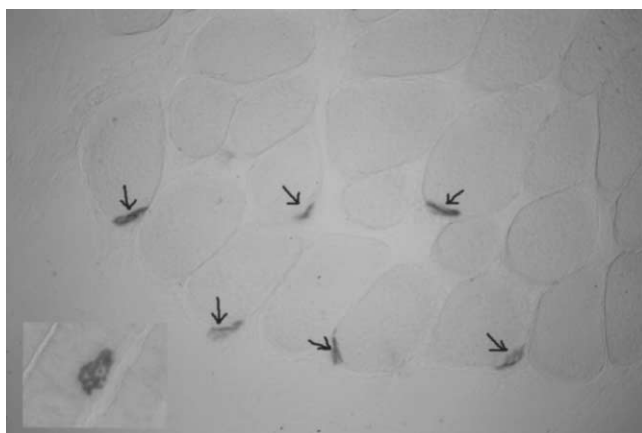


Fig. 1. Section of omohyoideus muscle (original magnification $\times 100$) from a nerve muscle pedicle graft after cholinesterase staining demonstrating motor end plates (small arrows) on individual muscle fibers. Inset: a single motor end plate (original magnification $\times 400$).

possible [19,23,31,45]. The cluster of nerve muscle pedicles and transected branches is carefully placed over the dorsal aspect of the larynx while the recipient muscle is exposed.

The main abductor of the equine larynx is the CAD muscle. Exposure of the CAD muscle is achieved by rotating the larynx laterally using a narrow hooked retractor over the wing of the thyroid cartilage. This retractor is placed through the septum between the cricothyroideus and cricopharyngeus muscles. The use of a 20-mm endotracheal tube rather than the 35-mm tube normally used on an adult horse makes rotation of the larynx easier. Once rotated, dissection through either the caudal portion of the cricopharyngeus muscle (CAD) muscle or fascia overlying the CAD muscle is performed, and using a Weitlaner retractor, a window is created allowing access to the CAD muscle. It is common to find a plexus of vessels in this region, and care must be taken to avoid excessive hemorrhage, which reduces the visibility of the CAD muscle. The CAD muscle in hemiplegic horses is usually pale in color and wasted to a varying degree. The amount of muscle wastage in grade 4 LLH horses is far greater than that in grade 3 horses. Often, in grade 3 horses, the CAD has a striped appearance with pale areas of denervated muscle next to more normal-appearing skeletal muscle. In any case, the overall mass of the muscle is always reduced by at least 50%.

Once the CAD muscle is exposed, the pedicle grafts are inserted into individual pockets in the CAD muscle. Long vascular forceps are used to create a deep slit-like opening in the CAD muscle belly that runs parallel with the fibers of the CAD muscle. A single 4-0 polydioxanone suture is used to hold the pedicle graft in the opening of the CAD muscle. The needle is passed from the muscle surface into the slit opening, retrieved, passed through the pedicle graft (or through the perineurium in the case of an implant), and then passed down into the opening of the CAD, exiting through the surface next to the original needle insertion point. Depending on the exposure of the CAD muscle, the pedicle grafts and nerve implants are spread over the muscle belly as evenly as possible. Nerve branch lengths can sometimes dictate how far apart the pedicles end up in relation to each other, however. Once the nerve muscle pedicles and nerve implants are sutured in place, the larynx retractor is removed and assessment is made of the tension on the first cervical nerve. Closing the incision usually involves a three-layered closure using 2-0 polydioxanone in the subcutaneous and subcuticular layers and 2-0 polybutester in the skin. Care should be taken during closure to inspect for leaking lymphatic vessels, which are common in younger horses. Failure to ligate these leads to significant postoperative seroma formation. A stent bandage is usually sutured over the incision, and an elastic bandage is used to apply pressure over the incision area, minimizing the opportunity for seroma formation.

Since September 2000, the primary author has started performing a left cordectomy using a diode laser in combination with the nerve muscle pedicle graft. Initially, the cordectomy was performed under general anesthesia

immediately before the nerve muscle pedicle graft surgery. Presently, the cordectomy is routinely performed the day after the nerve muscle pedicle graft procedure in the standing and sedated horse. Before September 2000, four LLH-affected horses that had been treated with the nerve muscle pedicle surgery still made excessive noise on return to racing. These horses all demonstrated arytenoid movement consistent with reinnervation, and after ventriculectomy, they continued racing successfully.

Postoperative management

After recovery from anesthesia, the horses are confined to a box for a period of 2 weeks. The neck bandage placed at surgery is removed after 48 hours and replaced with another for a further 48 hours. To avoid trauma to the surgery site, we recommend that horses be in stalls where they cannot put their head over the door and rub the neck incision. Sutures are generally removed after 14 days. The prophylactic antibiotics penicillin and gentamicin are routinely given until 96 hours after surgery. Gentamicin has been added to the postoperative management since laser vocal cordectomies were started so as to reduce the risk of infection. Anti-inflammatory medication, usually phenylbutazone, is administered for 7 days after surgery.

After stall confinement, we normally recommend a further 2 weeks in a day yard followed by paddock turnout for 12 weeks. At this stage, we recommend that horse go into training (16 weeks after surgery). This time frame is based on information from studies on dogs [21], people [33], and horses [17], where the earliest time to see clinical evidence of reinnervation is around 12 weeks after surgery. When the horses are returned to exercise, it is advised that episodes of fast exercise are introduced as early and as frequently as possible. Because the omohyoideus muscle is an accessory muscle of respiration, considerable respiratory effort must be undertaken to activate the first cervical nerve. Therefore, we recommend that Thoroughbreds gallop over 400 m every second day of training.

After 6 weeks of training, trainers/owners are requested to present the horse for endoscopic assessment of the larynx. At rest, the left arytenoid cartilage most commonly looks exactly as it did before surgery. This is because the first cervical nerve is inactive at rest; thus, there is no depolarization of the nerve and no CAD muscle contraction. Two diagnostic reflexes have been developed to stimulate contraction of the omohyoideus muscle and the newly innervated CAD muscle. The first involves stretching the horse's head and neck upward as high as possible while closely observing the larynx through the endoscope (Fig. 2). If reinnervation has occurred, there is often a spontaneous flicker or single abduction of the left arytenoid cartilage. The second reflex involves pulling back rapidly with a finger or thumb on the commissure of the horse's lips (Fig. 3). Again, a sudden



Fig. 2. While standing on a stool, the operator has the horse's head elevated by a handler using a twitch handle as an aid in extension. Moving the horse's head up and down quickly in this manner often produces a spontaneous movement of the left arytenoid cartilage.

abduction of the left arytenoid cartilage occurs if reinnervation has been successful. This reflex can be stimulated from the left or right side of the head.

An abductor movement of the left arytenoid cartilage indicates there has been reinnervation of the CAD muscle. Once this has been identified, the recommendation to the trainer is to continue training toward a return to



Fig. 3. The handler places a finger at the commissure of the horse's lips and pulls back quickly and firmly to stimulate movement of the reinnervated arytenoid cartilage, which is visible via endoscopy. The reflex works on either side of the mouth.

racing. If there is no evidence of movement at the first revisit, the advice is to turn the horse out again for another 8 weeks and then to go through the same program of 6 weeks of training before presenting the horse again for laryngeal evaluation. After this second period of training, at least 9 months have generally elapsed from the date of surgery. Horses can take up to 12 months to show evidence of successful reinnervation [17]. Clinical experience suggests that if there is no arytenoid abduction as a result of reinnervation 9 months after surgery, there is only a small chance of improvement from that point on. Information from stimulation studies suggests that reinnervation probably occurs in nearly all patients by 4 to 5 months. The problem seems to be not whether reinnervation has occurred but whether sufficient regeneration of the CAD muscle has taken place to provide stability of the left arytenoid cartilage during exercise.

The best evidence of successful reinnervation in an individual horse is derived from a treadmill endoscopic or upper airway flow mechanics study. Only a few horses in this series have undergone this evaluation because of the lack of a treadmill facility at the primary author's hospital. Nevertheless, in treadmill studies, we have found that those horses with movement of the left arytenoid cartilage visible at rest after nerve muscle pedicle graft surgery maintained arytenoid abduction during vigorous exercise [6,17]. If movement in the affected arytenoid could not be demonstrated in the standing horse, a dynamic collapse occurred during exercise.

Complications

Complications associated with laryngeal reinnervation are few when compared with prosthetic laryngoplasty. The most frequent complication has been seroma formation 3 to 5 days after surgery. The large potential dead space that exists after the nerve graft procedure lends itself to the formation of a seroma. The use of a compressive neck bandage that encircles the neck rostral and caudal to the poll and is maintained for 4 to 6 days after surgery has reduced this to some extent. It is the opinion of the authors that many of the seromas that form are caused by leaking lymphatic vessels that are inadvertently cut during surgery. Large lymph vessels travel along the lateral border of the omohyoideus muscle and can be transected during the surgical approach. These have the potential to produce large volumes of fluid continually for a number of days after surgery. Some of these seromas have resolved without intervention. Treatment of other seromas has involved open drainage and daily lavage. These horses have been placed on procaine penicillin therapy until the open tract discontinues draining. Some seromas have become infected, and these have been treated with antibiotics selected from culture and sensitivity results. Because of the ventral position of the incision, once opened, the seroma drains easily and requires minimal nursing care apart from daily lavage and attention to skin

around the incision. Horses that have suffered a postoperative incisional infection associated with a seroma have gone on to have successful reinnervation; thus, it would seem that this complication does not necessarily compromise success of the nerve muscle pedicle graft [14].

We have observed several other infrequent complications. Immediately after recovery, one horse suffered a laryngeal spasm that was successfully treated by passage of a nasotracheal tube. One horse developed a large hematoma immediately after surgery that required the incision to be reopened and the vessel ligated. The bleeding had originated from the omohyoideus muscle, where a pedicle graft had been removed.

Follow-up results

Thoroughbreds

For follow-up purposes, horses were placed in two groups: those that had raced before surgery (group A) and those that had not raced (group B). These groups were then further defined as either grade 3 or grade 4 LLH-affected horses.

Group A

Sixty-three horses were included in this group: 39 horses had grade 3 laryngeal function, whereas 24 had grade 4 laryngeal function. The age of the horses ranged between 2 and 6 years (average of 3.2 years). Of the 63 horses, 4 either died or were retired for reasons unrelated to the surgery before resuming racing. Of the 59 remaining horses, 95% went on to start in one or more races. The average length of time from surgery to the first race was 7.5 months for grade 3 horses and 8.6 months for grade 4 horses. Of the entire group of 63 horses, 84% were observed by the primary author to have evidence of laryngeal reinnervation. The earliest that reinnervation was identified was at 4 months, and the latest was at 9 months. After surgery, the group A horses raced an average of 12.5 times each. Of the 59 horses, 32 (54%) won one or more races after surgery.

To analyze the effectiveness of the nerve muscle pedicle graft, the following four variables were calculated for each horse before and after surgery: total performance ranking, total prize money, performance ranking per start, and prize money per start. The performance ranking was calculated on all race starts before and after surgery as follows. First-, second-, and third-place finishes were allocated four, three, and two points, respectively, whereas an unplaced finish was allocated one point. Because both indicators of effectiveness (prize money and performance rank) have a distribution that is right skewed, the median rather than the mean is presented as the primary indicator because it better reflects the “probable outcome” than the mean. In addition to the summary statistics, the Wilcoxon rank test was used to examine potential differences between

indicators of effectiveness before and after surgery. Significance was defined as $P < 0.05$.

1. Total performance ranking: 34 of the 59 (58%) horses had an improved total performance rank after surgery. The median total performance rank was 9 before surgery and 13 after surgery. After surgery, horses had a significantly better total performance ranking ($P = 0.03$).
2. Total prize money: 31 of the 59 horses (53%) earned more prize money after surgery. The median total prize money per horse was \$4080 before surgery and \$8500 after surgery. Although the median prize money earned after surgery was greater than before surgery, the difference was not statistically significant.
3. Performance ranking per start: 34 of the 59 horses (58%) had improved performance ranking per start after surgery. The median performance rank per start before surgery was 1.7, and it was 1.6 after surgery. Performance rank per start before and after surgery was not significantly different.
4. Prize money per start: 34 of the 59 horses (58%) earned more money per start after surgery than before. The median prize money per start was \$625 before surgery and \$783 after surgery. Although median prize money per start was greater after surgery than before, the difference was not statistically significant.

If surgical success is defined as improved performance ranking and more money earned after surgery, the nerve muscle pedicle graft technique was successful in 58% and 53% of affected horses, respectively. When assessed based on grade of LLH, 47% of grade 3 horses had a higher performance ranking and 47% earned more money after surgery. After surgery, 65% of grade 4 horses had a higher performance ranking and 60% earned more money. It stands to reason that the more severely affected grade 4 horses benefited more from the surgery than the grade 3 horses.

After surgery, 53 horses (84%) raced over the same or a greater distance than before surgery. Of these horses, 20 raced and won over equal distances before and after surgery, which allowed comparison of racing speed. Eighteen of these horses (90%) had personal best times after surgery.

Group B

Sixty-six horses were included in this group: 19 had grade 3 LLH and 47 had grade 4 laryngeal function. The age of the horses ranged between 1 and 3 years (average of 22 months). Of the 66 horses, 39 (60%) went on to start in at least one race, 15 were considered to be failures, 2 died, 3 were retired for reasons unrelated to the surgery, 4 were lost to follow-up, and 3 are still convalescing. The average length of time from surgery to the first race was 14.7 months for grade 3 horses and 12.6 months for grade 4 horses. The length of time between surgery and the first race is longer in group B horses, because many were treated at 18 months of age and were not raced until

their 3-year-old year. From the entire group of 66 horses, 76% were observed by the primary author to have evidence of laryngeal reinnervation. The earliest sign of reinnervation was seen was at 4 months, and the latest was seen at 9 months, which was the same as in the group A horses. The average age at their first race was 3.1 years of age. The 39 group B horses that raced after surgery had an average of 10.6 starts each.

For all horses that raced after surgery in group B, we compared money earned per start after surgery with the Australian average, obtained from the Victorian Racing Club. Table 1 demonstrates that LLH-affected horses treated with a nerve muscle pedicle graft performed about the same as the national average each year and that LLH-treated horses earned considerably more money per start than the comparison group as 3-year-olds.

Although some horses that have been treated with left vocal cordectomy in addition to the nerve muscle pedicle graft have resumed racing, there are insufficient numbers to make any conclusions regarding additional benefit of the combined surgeries. There has been a reduction in trainer complaints early in training, however. Many of the horses treated with laryngeal reinnervation alone continue to make audible noise when first put back into training. In cases where reinnervation has been successful, the degree of noise reduces progressively as training continues. This is most likely a result of increased activity in the reinnervated CAD muscle with time.

Standardbreds

A total of 10 LLH-affected Standardbreds have been treated with the nerve muscle pedicle graft. These horses all had grade 4 laryngeal function before surgery, and all had raced. Of the 10 horses, 6 raced after surgery. Of the remaining 4 horses, 2 did not return to racing for unrelated problems, 1 died, and 1 was considered a failure by the trainer. The average time from surgery to the first race was 8.5 months, which is faster than the Thoroughbred grade 4 horses (9.1 months). Three of the 6 horses earned more money after surgery than before. Discussion with trainers of the horses involved in follow-up indicated that in 5 of the 6 horses that raced after

Table 1

Prize money per start for each year of racing of left laryngeal hemiplegia-affected horses treated with a nerve muscle pedicle graft compared with the national average (1996–2001) for Australian horses

Year of racing	Nerve muscle pedicle graft horses (\$/start)	National average (\$/start)
2-year-olds (n = 2)	1895	2038
3-year-olds (n = 34)	2822	1425
4-year-olds (n = 18)	1227	1289
5-year-olds (n = 6)	1054	1167
6-year-olds (n = 1)	1050	1114

surgery, trainers were satisfied with the decrease in noise and return to racing performance.

Warmbloods

Seven LLH-affected warmbloods have been treated with the nerve muscle pedicle graft surgery. Of these seven, five have gone on to perform at a higher level than before the operation. One horse was 12-years-old at the time of surgery and had been affected with LLH for at least 4 years. The owners of these five horses have all reported a marked reduction in noise level during exercise and were satisfied with the outcome. None of these horses underwent vocal cordectomy. One of the horses, a 6-year-old Irish Draught–Thoroughbred cross, is ranked in the top five horses in 3-day event competitions in Australia.

Discussion of results

Eighty-four percent of group A horses and 76% of group B horses showed evidence of reinnervation based on visible movement of the affected arytenoid cartilage. This high level of successful reinnervation explains why as many as 94% of LLH-affected group A horses treated with a nerve muscle pedicle graft raced one or more times after surgery. Furthermore, LLH-affected Thoroughbreds not raced before surgery and treated with the nerve muscle pedicle graft raced as well as the national average after surgery.

Fifty-eight percent of treated horses that had raced previously had an improved performance score. Fifty-three percent earned more prize money after surgery. How does this compare with follow-up results of prosthetic laryngoplasty-treated horses? In the study of Hawkins et al [12], 77% of horses raced after prosthetic laryngoplasty, with 56% racing at an improved level based on a performance ranking system. These investigators only assessed three race starts before and after surgery to calculate the performance ranking. Using the same criteria, laryngeal reinnervation resulted in 54% of horses having an improved performance ranking. In the study of Strand et al [13], 94% of horses raced after prosthetic laryngoplasty, with 45% demonstrating improved performance based on race times. Where comparison was available for wins over the same distance before and after laryngeal hemiplegia, 90% of horses had improved race times. Thus, it seems that laryngeal reinnervation is as effective as prosthetic laryngoplasty in the treatment of LLH.

Failure of the nerve muscle pedicle graft surgery to result in reinnervation could be demonstrated in only 20% of horses. Failure of the nerve muscle pedicles to reinnervate the paralyzed CAD muscle could be caused by a number of reasons. First, the pedicles may be pulled out of the CAD muscle after surgery if an insufficient length of the main branch of the first cervical nerve is provided. Second, if pedicle grafts or transected nerve ends

are placed in the CAD and there is excessive tension placed on them or undue stretching occurs before the pedicles are created, interruption of the intraneural microvascular supply could occur. An 8% elongation of a nerve or section of a nerve results in impaired blood flow, and greater than 10% elongation produces total arrest of microvascular blood flow [46]. The rostral-caudal movement of the larynx during swallowing may also compromise intraneural microvasculature.

Finally, there may be death of the cell body of the first cervical nerve as a result of transecting the nerve branches. When a peripheral nerve is cut, there is degeneration of the distal segment as well as degenerative change for a short distance in the proximal segment, usually up to the point of the first collateral axon. After transecting an axon, interruption of both rapid axonal transport and the slower axoplasmic flow also occurs. Because axonal transport occurs in a retrograde direction, changes in the cell body do occur; if severe enough, they may result in cell death. When the peripheral axon of a spinal motor neuron is cut, characteristic changes occur within the cell body within 2 to 3 days. These changes are referred to as chromatolysis of the neuron. Chromatolysis lasts for 1 to 3 weeks after cutting of a nerve. The process of chromatolysis represents the synthesis of proteins necessary for axon regeneration, and if the regenerating axons do not make new connections distally, the cell body may atrophy or totally degenerate [47]. In most cases, any transected nerve branches are sutured into the CAD muscle, but in some situations, a large caudal branch of the first cervical nerve has to be transected to free up the main body of the nerve for repositioning. In these cases, the potential for failure as a result of nerve damage is increased.

Successful laryngeal reinnervation occurred in 84% of the group 1 horses, but improved racing performance was demonstrated in only 58% of cases. Therefore, it seems that despite successful reinnervation, there is not always sufficient CAD muscle regeneration to sustain abduction of the left arytenoid cartilage during maximal exercise. Other reasons why successful treatment of LLH may not result in improved racing performance include musculoskeletal injury, age, and athletic ability of the horse.

In horses with grade 3 laryngeal hemiplegia, some functioning fibers of the recurrent laryngeal nerve remain. This raises the question of whether a nerve muscle pedicle can reinnervate a partially denervated muscle. There are no studies in horses to answer this question; however, it has been shown in other species that dual innervation of a skeletal muscle can occur after implantation of a second independent nerve [48]. It has been postulated there is a chemotactic factor released in conditions of acute denervation that helps to stimulate reinnervation from an implanted nerve (HM Tucker, MD, personal communication, 1992) [24]. Therefore, it has been suggested that the recurrent laryngeal nerve should be cut at the time of the nerve graft to take advantage of this “trophic” factor. In the first clinical case operated on by the primary author, this was done. That particular horse demonstrated return of arytenoid function and raced for 3 years, winning multiple races.

The only other report that exists in the literature regarding treatment of LLH using the nerve muscle pedicle graft involves 18 event and hunt horses from England. In this series, 14 horses were available for follow-up; of these, 6 (43%) were considered failures. Eight horses (57%) were judged by their owners to have improved performance [49].

Stimulation studies with the nerve muscle pedicle graft

The nerve muscle pedicle graft surgery described in this article results in reinnervation of the CAD muscle in most cases. Reinnervation may sometimes be insufficient for clinical success, however. Reinnervation must result in an early return of function in the reinnervated muscle. Early return to function may be enhanced by peripheral nerve stimulation (PNS). PNS is used in several situations in human surgery in which postoperative muscle strength needs to be enhanced. Examples include dynamic cardiomyoplasty [50], where the latissimus dorsi muscle is used as a supporting muscle in treating cardiomyopathy, and anal myoplasty, which uses PNS to strengthen the anal sphincter. Electrodes are connected to an implantable power source, an internal pulse generator (IPG), which is similar in size to those used for cardiac pacemakers. The IPG is controlled by an external handpiece connected to a computer that adjusts stimulus magnitude and frequency to suit the patient's needs. A pilot project was conducted to assess whether PNS could enhance the success of the nerve muscle pedicle graft in the horse.

Two horses had their left recurrent laryngeal nerve transected and were treated with a nerve muscle pedicle graft 4 weeks later. At the same time, a bipolar platinum electrode (model 3586 lead kit; Medtronic, Minneapolis, MN) was attached to the first cervical nerve after the pedicles had been inserted into the paralyzed CAD muscle (Fig. 4). This lead was then attached to the IPG (model 3203; Medtronic).

The IPG was not turned on until 4 months after the nerve muscle pedicle graft surgery so as to allow time for reinnervation to take place. In both horses, no movement was detected during endoscopy after the IPG was turned on. The two horses underwent a second operation; on examination of the nerve and electrode, it was discovered in both cases that the nerve-electrode interface had not been maintained and therefore no stimulus was being transferred to the first cervical nerve. Also, it was discovered in the second horse that the single platinum wire to one electrode had broken. These two pilot horses demonstrated that the human implant system for peripheral nerve stimulation normally implanted along a straight section of limb nerves or the spinal cord was not easily adaptable in the equine neck region. Both electrode failure and the nerve-electrode connection were problems to overcome. Even if the IPG system had been successful, the cost of an IPG and lead kit was \$5000 and thus possibly not practical for common use.



Fig. 4. The internal pulse generator used in the two pilot horses with the four-point bipolar electrode (inset) to which the first cervical nerve was attached.

As a result, external nerve stimulation has been investigated. In this preliminary study, two cases of LLH were included. One horse had been diagnosed with grade 4 LLH 3 years earlier, and the second horse also had grade 4 LLH diagnosed 6 months before surgery. In this study, two 50-strand, platinum, Teflon-coated wires were tied around the first cervical nerve after the nerve muscle pedicle graft surgery had been performed. These wires were then coiled under the skin just caudal to the angle of the mandible. After 4 months, the Teflon-coated wires were retrieved and left exiting from the skin for a length of 50 cm. The free ends of these electrodes were attached initially to a nerve stimulator to assess whether reinnervation had taken place. In both cases, excellent abduction of the left arytenoid cartilage was observed on endoscopy when a stimulus was applied to the electrode. The stimulus required to induce movement was 0.5 V applied for 200 milliseconds. After identification of reinnervation; a small electric muscle stimulator (EMS-HP; Electro Medical Sales and Rentals, Melbourne, Victoria, Australia) that could be attached to the halter was connected to the electrodes for 30 minutes daily to activate the CAD muscle. After 30 days, the horses were sent home into full training. Subsequently, both of these horses have performed successfully in 3-day events and the dressage arena with minimal noise production. The cost of the external electric muscle stimulator was \$500. The optimal length of time required to

achieve maximal return of CAD function is yet to be established, as are the exact electric stimulus parameters. Long-term stimulation is unnecessary, and once sufficient CAD muscle development has occurred, the external electrodes can be removed. This method of external PNS warrants further investigation as a method to reduce time of return to athletic function and to improve upper airway function further.

Clinical significance

Data presented in this article clearly demonstrate that the nerve muscle pedicle graft technique can be used to successfully reinnervate the CAD muscle of LLH-affected horses. Seventy-six percent of horses not raced and 84% of raced horses demonstrated endoscopic evidence of reinnervation. Because reinnervation may not improve upper airway function in all horses, and because many factors other than upper airway health influence racing performance, not all successful reinnervations have produced successful racing performances. Nonetheless, LLH-affected horses treated with a nerve muscle pedicle graft performed as well as the national average. A question of interest is how the nerve muscle pedicle graft technique compares with prosthetic laryngoplasty for the treatment of LLH. The results presented in this article show that the nerve muscle pedicle graft technique is as efficacious as prosthetic laryngoplasty when racing performance is measured. In the study of Hawkins et al [12], 77% of horses raced after prosthetic laryngoplasty, with 56% racing at an improved level. In the study of Strand et al [13], 94% of horses raced after prosthetic laryngoplasty, with 45% demonstrating improved performance. In the group A horses treated with the nerve muscle pedicle graft technique, 94% went on to race one or more times, 58% had an improved performance score, and 53% earned more prize money after surgery.

The biggest disadvantage of the nerve graft surgery is the time from surgery to the first race. Time to first race for group 1 horses was 7.5 and 8.6 months for grade 3 and grade 4 laryngeal function, respectively, whereas after prosthetic laryngoplasty, time to first race is 5.8 months [12]. The difference in time to first race may not be important to owners, depending on the breed, age, and intended use of the patient. The biggest advantage of the nerve muscle pedicle graft technique is that there are no chronically debilitating or potentially fatal complications as can occur after prosthetic laryngoplasty.

In yearlings or early unraced 2-year-olds, the nerve muscle pedicle graft is a reasonable surgery to offer owners. In the case of a 5-year-old Thoroughbred gelding, prosthetic laryngoplasty could be the logical choice. In the primary author's practice, many clients have refused to have prosthetic laryngoplasty performed because of past experiences with postoperative complications. Many stud masters have complained about the difficulties they have had with some "tie-back" horses long after their

racing careers have finished. In the situation of valuable young female racehorses, where time is not an issue and long-term respiratory health is important, the nerve muscle pedicle graft is a logical option to offer clients.

Based on results from the pilot study, further research into external nerve stimulation has the potential to decrease time from surgery to first race and, more importantly, the potential to increase the percentage of horses that win races, which is the only measure of success that many owners and trainers are interested in.

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