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Biomechanical Study of Repair of Tendon Gap by Bovine Fetal Tendon Transplant in Horse

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Abstract: One of the most encountered problems in horses is disorder of locomotion. Injuries to the tendon include large part of orthopedic activitiesin animals. The perpose of this study was to use the tendon xenograft in cases of tendon gap in horses. The study was conducted on 10 clinicaly healthy adult indigenous horses from both sexes. The deep digital flexor tendon of the left fore limb was exposed undergeneral ensthesia and 5-7 cm of middle one third of tendon was cut and removed. Then peices of the bovine fetal tendon were replaced for the tendon gap. The temdon samples were collected from the site of operation on 70th post operative day for biomechanical evaluation. The result of biomechanical testing proved a good tensile strenght on 70 days post operation. The result of the study indicated that fetal tendon graft is a possible treatment fot severe tendon defects or lacerations in the horses.

Key words: Horses, fetal tendon, tendon transplant, xenograft, biochemical study

INTRODUCTION

The treatment of tendon injuries is still controversial. Although, the understanding of the anatomy and biomechanics of the flexor tendon is improving and although a variety of therapeutic options have been described, the results of clinical studies are still conflicting (Barton *et al*., 1984; Bertone *et al*., 1990; Clancy *et al*., 1983; Cross and Powell, 1984; Dandy and Pusey, 1982; Fithian *et al*., 1992; Forslund *et al*., 2003; Fowler and Messieh, 1987; Harner *et al*., 1995; Jaakkola *et al*., 2000; Kannus *et al*., 1991; Lee *et al*., 2000; Taniguchi and Tamaki, 2000). In particular, one of the most encountered problems in horses is disorder of locomotion. Tendon injury can occur in a number of ways. Disruption can occur following an extreme overload, or from traumatic laceration such as car accident in dogs. Blood supply to the tendon is reported to be poor, thereby healing is often protracted (Clancy *et al*., 1983; Dandy and Pusey, 1982; Harner *et al*., 1995; Kuo *et al*., 2003). Vascular supply depends on location within tendon and presence or absence of sheath. Paratenon is most vascular, most vessels run parallel to long axis of tendon. Proximal third is supplied by the musculotendinous junction, middle third is supplied by paratenon vasculature and distal third is supplied by osseous-tendinous junction (Kraus *et al*., 1995). In the past, one point of particular interest was the degree to which collagen fibril organization recovers during autograft and allograft healing following cruciate ligament reconstruction. A shift to predominance of small-diameter collagen fibrils in autografts and allografts was a major finding of several studies. Autogenous skin have been used for repair of cruciate ligament reconstraction in dogs and cattle (Belkoff and Haut, 1992; Bosch *et al*., 1998; Liu, 1994). Autogenous tendon transplant and allogenic transplantation of tendon have been performed with varying degrees of success (Birsh *et al*., 1999; Gillis, 1977; Taniguchi and Tamaki, 2000). Allogenic tendon transplantation has been undertaken experimentally in chicken (Zhang *et al*., 2001). Hamstring tendon graft was used for anterior cruciate ligament reconstruction (Gordia and Grana, 2001). Artificial tendon has been transplanted in man (Dong and Sheng, 1989). There is one report on heterogenic or xenograft tendon transplantation in the literature demonstrating bovine foetal tendon transplantation for repair of tendon gap in rabbit (Dehghani *et al*., 2005). There is no report on heterogenic tendon transplantation on horse. Therefore, the purpose of this study was to use bovine foetal tendon for repair of tendon gap in horses.

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MATERIALS AND METHODS

This study was conducted on 10 clinically healthy young indigenous Iranian horses from both sexes. All the animals were dewormed and examinated clinically. The heart rate, respiratory rate and body temperature were within normal range. During the entire experimental period all the animals were kept under similar management and feeding practices. Deep Digital Flexor (DDF) tendons in the mid third of tibial region of the hind limbs were prepared. The animals were sedated using acetylpromazine (0.05 mg kg^{-1}, Kla laboratoria, Belgian) intravenously. The anesthesia was inducted by injection of ketamine (2.2 mg kg^{-1}, Rotexmedica GMBH, Germany) intravenously following endotracheal intubation maintained by a mixture of halotane and oxygen. The left metacarpal region was prepared for an aseptic surgery. A 10 cm long skin incision on the lateral surface of middle third of metacarpal region was made. The superficial and deep fascia was bluntly dissected to have a good exposure of SDF and DDF tendons. The tendon sheet was incised and the DDF tendon was separated bluntly from the SDF tendon. About a 6 cm long piece of DDF tendon was transected. Both the stumps of each tendon were held in place by the stay sutures with No.1 polyamide. 2 to 3 pieces of the bovine foetal (7 mounts old) tendon were separated aseptically was replaced in the tendon gap by steel strings No.0 using a single locking-loop suture pattern at the graft bed. The tendon sheet was sutured in simple continues pattern.

The surgical site was lavaged by normal saline solution. The subcutaneous tissue in all the animals was sutured by catgut No.1 by simple continuous sutures. The skin was closed with simple continuous sutures using polyamide No. 1 and the wound was painted with povidone-iodine solution and covered with a piece of sterile gauze.

The operated limb in all the animals were immobilized by cast involving 10 cm above the carpal joint include the hoof which was kept slightly flexed in order to reduce tension on grafted tendon. Intramuscular injection of flunexin meglomine (0.02 mg kg^{-1}, Razak Co., Iran) was administered twice daily for three days. As well as ampicilin (Nasr Co., Iran) was given intramuscularly at the rate of 22 mg kg^{-1} body weight twice a day for seven days. All the animals were confined in order to restrict their activity for 10 postoperative days. The cast was removed on 30th post operative day and the horses were allowed to have limited free exercise.

On the day 70th the tendon-graft-tendon unit was collected from the site of operation for biomechanical evaluation. The samples comprised 3 cm above the graft and 3 cm below the graft. The intact samples were collected from DDF tendon of collateral limbs to be compared with tendon-graft-tendon units. Each sample was individually wrapped in normal saline-soaked gauze, placed in a sealed plastic bag, labeled and kept at -20°C. Twenty-four hours before testing, the tendon-graft-tendon units and intact tendons were thawed (Amiel *et al*., 1984; Andrew *et al*., 2004; Blevins *et al*., 1994). Tensile tests were performed by placing wedge-action grips of a servocontroled mechanical testing speed system (Model TT-CM, Instrone, England) (Fig. 1). Statistical analysis was performed by paired T-test using SPSS statistic software and a level of significant of $p<0.05$ was considered.

Fig. 1: Servohydraulic mechanical test system. The tendon-graft tendon unit is under the test

RESULTS

Energy, load, elongation, stress and strain were measured simultaneously on an *in situ* tensile testing apparatus.

No significant difference in tensile strength was found between control and test group in all factors ($p>0.05$) except the load ($p<0.05$).

But when compared to the foetal tendon all parameters in both groups [except the energy ($p>0.05$)] had significant difference with the foetal tendon ($p<0.05$) (Table 1).

Table 1: Biomechanical study of the tendon in the control group, foetal tendon transplanted group and intact foetal tendon

	No.	Energy Cfmm	Load Kfg	Elongation mm	Stress Kfg mm^{-2}	Strain %
Control	5	5718.10 +/-1470.40	221.99+/-21.02 *	14.66+/-2.35	2.66+/-0.10	41.13+/-10.28
Transplanted Tendon -test	5	2930.13+/-773.71	167.74+/-18.79 *	17.76+/-0.70	1.40+/-0.88	37.95+/-4.73
Fetal tendon	5	649.77+/-0.56	48.81+/-1.30 â	14.59+/-0.68 â	3.78+/-0.56 â	205.35+/-2.41 â

(â)- Significant differences between the control and test group with the intact foetal tendon, (*)- Significant differences between the control and the test group

There was no non-weight bearing lameness after splint removal on 70th post operative day, only mild lameness (grade I) was evident for 7 or 10 days after the cast was removed. There was no adhesion, edema, infection or inflammation.

DISCUSSION

One of the most encountered problems in horses is disorder of locomotion. Injuries to the tendon include large part of orthopedic activities in animals. Different treatment techniques have been suggested for repair of tendon injuries. Many researches have been carried out on tendon auto-transplantation. Synthetic ligaments are occasionally used for particular tendon reconstruction, but natural materials are preferred for tendon reconstruction. There is no report of using tendon xenograft transplantation in the horses, but there are some studies on tendon xenograft in other animals. Dehghani *et al.* (2005) used bovine foetal tendon transplant for repair of tendon gap in rabbit, observing no graft rejection (Dehghani *et al.*, 2005).

Our data are in accordance with morphologic findings and mechanical properties of the autograft tendon transplantation previously reported. (Bosch *et al.* 1992 , 1995, 1998; Decker *et al.*, 1991, 1994; Jaakkola *et al.*, 2000; Slade *et al.*, 2001; Mudgal *et al.*, 2000)

Oloumi *et al.* (2002) used greater omentum for repair of tendon defect. They observed high rate of inflammatory cells around the repair site (Oloumi *et al.*, 2002).

The results of this study indicated that bovine foetal tendon graft is a possible treatment for severe tendon defects or lacerations in the horses. There was no rejection and no degenerative process in any cases. The transplanted foetal tendon acted as a good substitute for tendon defects (Dehghani *et al.*, 2005) and the results of biomechanical testing proved a good tensile strength on 70 days post operation. The graft tendon is strong enough to tolerate the projected forces generated during active motion without dehiscence or gap formation at the repair site. The significant difference between load of control and test group is because of incomplete collagen fibrils alignment in 70 days, remodeling of tendon needs more then 1 year to complete. If we could have continued our study for one year, there would have been no significant difference even in load energy at all.

CONCLUSION

The bovine foetal tendon is a good substitute for the cases of tendon gap or severe laceration in the horses.

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