

# Enhanced Suspensory Ligament Healing in 100 Horses by Stem Cells and Other Bone Marrow Components

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Suspensory ligament injuries are challenging because healing is slow and reinjuries are common. Healing requires growth factors that stimulate angiogenesis, mitogenesis, and matrix formation. Bone marrow contains growth factors, cells, and fibrinogen, which facilitate healing of damaged tissues. In 100 horses with ligament damage, bone marrow seems to enhance healing. Author's address: Alamo Pintado Equine Medical Center, Los Olivos, CA 93441. © 2001 AAEP.

## 1. Introduction

Suspensory ligament damage is a major cause of lameness and loss of use in sport horses. Natural healing of the suspensory ligament is often slow and of poor quality and results in long layoffs and a predisposition to reinjury. Many different therapeutic approaches have been used to promote ligament healing. However, none of these techniques have proven to be effective by double-blind or clinical studies. The results of anecdotal reports on ligament splitting, hyaluronic acid injections, iodine injections, blistering, pin firing, acupuncture, cold laser, rest, and rehabilitation have not been encouraging.

For the past 6 years, we have used a novel biological approach to facilitate suspensory ligament healing. This approach involves the intralesional injection of autologous stem cells and associated bone marrow components to stimulate natural ligament regeneration.

The use of autologous bone marrow to enhance bone healing is not new and has been described

in numerous veterinary and human orthopedic texts. Percutaneous injection of bone marrow for ligament healing was reported in 1987 by Pierce of Amgen, Inc. Pierce reported that platelet derived growth factor and transforming growth factor  $\beta$  in bone marrow cells enhanced wound healing in rats by stimulating collagen synthesis.<sup>1</sup> In 1998, Young at Osiris Therapeutics reported in the *Journal of Orthopedic Research* that seeding cultured mesenchymal stem cells into a tendon gap resulted in significantly improved repair biomechanics.<sup>2</sup>

This report summarizes the results of a retrospective study of the first 100 cases of suspensory ligament injury that we treated with intralesional injection of autologous bone marrow. Follow-up information for 2–6 years is available for these cases. The success rate in these first 100 cases has led to further use of this procedure in treatment of over 400 other ligament and tendon injuries.

This report also summarizes the comparison of outcomes of 66 proximal suspensory lameness cases not bone-marrow-aspirate treated from 1991 to 1996

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vs. the outcomes of 100 cases of suspensory ligament injuries that were treated with intralesional stem cell and marrow components from October 1995 to December 1998.

Of the 66 cases seen between 1991 and 1996 that were not treated with stem cell and bone marrow, 84.8% did not go sound or did reinjure within a year. Of the 100 cases that were treated with autologous bone marrow injection (ABMI) between 1995 and 1998, 84% went sound within 6 months and were back at work. Another 8% improved enough to return to competition but were not 100% sound. A total of 92% went back to work compared with 84.8% of the 66 cases diagnosed and monitored that did not go sound or go back to work between 1991 and 1996.

Since 1990, the use of diagnostic ultrasound to detect, quantify, and qualify ligament injuries and healing has improved our ability to evaluate the efficacy of treatments designed to promote ligament healing. In the past 6 years, diagnostic ultrasound, nuclear scintigraphy, computer tomography, clinical exam, and history have been used to evaluate the efficacy of intralesional injection of autologous bone marrow in horses with ligamentous injuries. Comparisons of ultrasonic healing between similar high-suspensory lesions not treated and treated with stem cells and bone marrow aspirate are significant. The improved quality of fiber alignment, decrease in cross-sectional area, and decrease in echolucency occurs more rapidly in the treated ligament vs. the non-treated ligaments. Serial ultrasound photos will be used to demonstrate these results.

Improving the quality and rate of suspensory ligament healing in the sport horse is critical to the economics and health of the showhorse and racehorse industry. After injection of bone marrow components into damaged suspensory ligaments, ultrasonic, clinical, and historical evidence indicated that 84% of treated horses responded exceptionally well to this mode of treatment. The use of stem cells and associated bone marrow components result in high-quality suspensory ligament healing, which is a practical, effective, economic, and safe procedure available to veterinarians dealing with sport horse injuries.

### 2. Materials and Methods

Between October 1995 and December 1998, 100 horses were treated for suspensory desmitis using the intralesional autologous bone marrow procedure. The patients that were treated had to satisfy one of the following entrance criteria: 1) clinical and ultrasonic evidence of a serious acute suspensory injury or 2) moderate to severe lameness caused by chronic suspensory desmitis. Ultrasonic changes included circumscribed peripheral or central hypoechoic lesions, significant increase in cross-sectional area, loss of ligament margins, associated MT III or MC III avulsion fractures, or combinations of the above. Some cases were selected based on

significant scintigraphy findings along with *grade 2+* lameness and significant abnormal ultrasonic findings.

Nine breeds of horses were represented in the following order of prevalence: Warmblood, Thoroughbreds, Quarter Horses, Andalusians, Peruvian Pasos, Arabians, Standardbreds, Saddlebreds, and Morgans. Ages ranged from 2 to 24 years of age. There were 19 hindlimb suspensories treated and 81 forelimb suspensories treated. The most common equine sports represented in this study were dressage, jumping, and racing.

All patients were anesthetized with intravenous or inhalant anesthetics, and positioned in dorsal recumbency. A surgical preparation of the sternum and affected leg was performed. A 60 ml syringe attached to a bone marrow needle was used to aspirate 20–30 ml of bone marrow from the sternabrae. The bone marrow was immediately transferred to 6 ml syringes, and the lesions were injected through ultrasound guided preplaced 18 gauge needles. Samples of the bone marrow aspirates were examined for cytology and cell counts.

Follow-up examinations were performed at approximately 60, 120, and 180 days for clinical and ultrasonic healing and return to soundness. Two patients that did not become sound with one treatment were retreated in the same manner.

### 3. Results

The results of 100 autologous bone marrow treatments are summarized below. After 6 months, 84 of 100 horses had returned to full work and soundness; an addition 8 horses were improved and went back to work with a very mild lameness. Five horses remained too lame to return to normal work. Three horses died of other causes and were not followed past 6 months. None of the horses had the degree of lameness increase, and only 2 horses developed evidence of soreness for 2–3 days after the treatments. Fifteen horses had concurrent forelimb or hindlimb retinaculum resections with open bone marrow injections. The horses that had retinaculum resections had greatly enlarged proximal suspensory ligaments.

### 4. Discussion

The results of this study indicate that intralesional injection of autologous bone marrow components is a safe, effective, and economical way to treat recalcitrant and serious suspensory ligament injuries. Nearly 90% of the treated horses responded favorably to this treatment technique, and the complications were limited to transient localized soreness in 2 horses. This study is the first to document the use of this technique in a large population of horses with ligament injuries, over a long period of time. The author recommends this procedure for the treatment of horses with injured suspensory ligaments. This study showed the rate of return to

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soundness and lack of evidence of reinjury was significantly higher than previously reported by other techniques, and significantly higher than healing rates previously documented at Alamo Pintado Equine Medical Center for the 5 years before the use of the intralesional stem cell and bone marrow aspirate procedure.

**References**

1. Pierce GF, Mustoe TA, Lingelbach J, et al. Platelet-derived growth factor and transforming growth factor- $\beta$  enhance tissue repair activities by unique mechanisms. *J Cell Biol* 1989; 429–440.
2. Young RG, Butler DL, Weber W, et al. Mesenchymal stem cell-bases repair of rabbit Achilles tendon, in *Proceedings*. 43rd Annu Meet Orthopaedic Res Soc 1997;249–342.