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# APPLICATION OF ALLOGENIC STEM CELLS IN THE TREATMENT OF TENDON INJURIES. CLINICAL CASEREPORTS OF CELL THERAPY IN HORSES

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## INTRODUCTION

In horses, stem cell therapies are a promising tool to the treatment of many injuries, which are common consequences of athletic animals. Tendon injuries are the most common morbidity that often compromising the performance in all types of sport horses and a return to the same level of activity. Although tendon injuries occur spontaneously during exercise, are preceded by progressive degeneration of the tendon matrix brought about by cumulative loading cycles, accelerated by competitive sport. Clinical injury results in a variable disruption of the tendon matrix, which induces an inflammatory response. This response is often short lived. Very soon after the injury, fibroplasia is initiated resulting in the formation of scar tissue within the tendon. Because of the poor functionality of scar tissue new treatments should aim at regenerating the tendon tissue.

## OBJECTIVE

To investigate the effect of the applications of allogenic adipose stem cells (ADSCs) in sixteen horses affected by tendon injuries.

## METHODS

The adipose tissue collected was isolated (Fig. 1). It was also assessed their ability to differentiate into osteogenic, chondrogenic or adipogenic (Fig. 2). All animals with tendonitis received 1 x 107 ADSCs into the injured tissue under local anesthetic and ultrasonographic control (Fig. 3). After one month, ultrasonographic control was performed again (Fig. 4). All procedures were approved by horse owners under signature of a veterinary service contract.

## RESULTS

The data showed that after stem cells from adipose tissue melt, they remained as "fibroblast-like" morphology. Osteogenic differentiation was evidenced by the mineralization of extracellular matrix at day 11, which became stronger at day 21 and by positive Von Kossa staining. After induction of adipogenic differentiation, the cells morphology changed within 24 hours from elongated fibroblastic cells to oval-shaped cells. After 4 days, vacuoles in the cytoplasm of the oval-cells were observed. At the day 6, it was observed an increased number of these cells by positive Oil Red O staining. Chondrogenic differentiation was observed 21 days after induction, visualized by the staining of the extracellular cartilage matrix proteoglicans. Our study was based on clinical cases, the animals were heterogenous for age, weight and sex, but all of them were athletic horses. One month after ADSCs application into the lesion, the formation of healthy tissue has been observed (Fig. 4). All treated horses showed a functional recovery and were able to return to their normal activity, without lesion recurred.

Figure 1. The equine adipose tissue collected was isolated and evaluate the proliferative and differentiative potential of the stem cells. The adipose stem cells were stored frozen with liquid nitrogen.

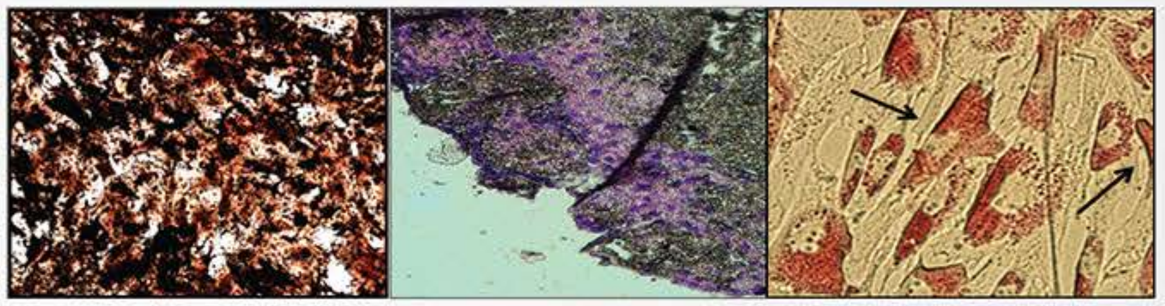
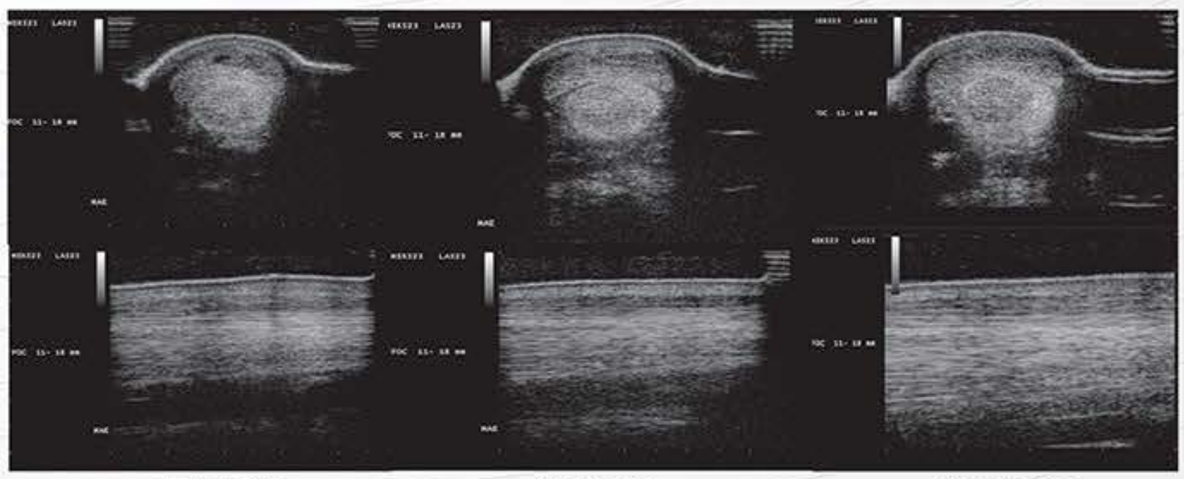


Figure 2. Equine adipose stem cells showed Osteogenic (A), Chondrogenic (B) and Adipogenic (C) differentiation. Objective 20 x (A,B) and 40 x (C).

Figure 3. Sterile application of the allogenic equinie adipose stem cells into the central core lesion under ultrasonographically guided injection.



Pre-injection One month Two months

Figura 4. Sequential tranverse (left column) and longitudinal (right column) ultrasonographs taken from a horse treated by allogenic equine stem cells.

## CONCLUSION

This study demonstrated that, in the case of tendon injury, the application of stem cell therapy in horses provided functional recovery of damaged tendons and treated animals were capable to return to their normal activity.

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