Magnetic Resonance Imaging Application to Live Horse for Diagnosis of Tendinitis

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ABSTRACT. Six live horses with various stages of acute to chronic superficial digital flexor (SDF) tendinitis were examined using magnetic resonance imaging (MRI). In each case, MRI findings were compared to the corresponding ultrasonographic (USD) and histologic findings, to establish the usefulness of MRI. In the acute cases, lesions characterized by hemorrhage were well defined as high signal intensity on MRI and hypoechoic regions on USD. Chronic tendon fibrosis was slightly hyperechoic and difficult to distinguish from the normal tendon tissue around the original injury by using USD. In contrast, MRI visualized the chronic lesion as a low intensity signal, which could be distinguished from the black background of the normal SDF tendon tissue. This study clearly demonstrated MRI was the better imaging modality for the objective detection of chronic scar tissue in live horses. These findings, from living horses, suggest an advantage of MRI in the clinical application to diagnose tendinitis in cases where there is chronic scar tissue that is difficult to discern on USD.

KEY WORDS: equine, MRI, tendinitis.

Superficial digital flexor (SDF) tendinitis is a common musculoskeletal disease, occurs with high incidence in racehorses required to run at full speed [5]. The histopathologic characteristics at the onset of tendinitis include hemorrhage and inflammation. After initial inflammation, damaged tendon is replaced by granulation tissue which then remodels into fibrous scar [6]. At present, ultrasonography (USD) is a very valuable for the evaluation of equine tendinitis[4]. USD depicts such inflamed tendon areas as anechoic or hypoechoic areas. However, it is difficult for the clinician to diagnose SDF tendinitis during healing as the injured region is progressively replaced with echogenic connective tissue which can be difficult to differentiate from remaining tendon tissue [12]. To reduce this limitation, experienced ultrasonographers evaluate the tendon using a combination of fiber alignment and echogenicity on the ultrasonograph during this phase. But such diagnoses lack the objectivity in comparison to the diagnoses in the acute case has clear hypoechoic findings. Magnetic resonance imaging (MRI) has the ability of depicting soft tissue and is widely used to diagnose diseases of joints, tendons, and ligaments in human medicine [11, 17]. Crass et al. [2] first reported the usefulness of MRI in diagnosing equine SDF tendinitis, using cadaver samples. They suggested that MRI made it possible to distinguish between tendons containing scar tissue and tendons in which inflammation remained in progress. Therefore, MRI may make it possible to monitor more accurately the state of tendon repair than USD. Consequently MRI may have clinical benefits in preventing the recurrence of tendinitis caused by inadequate rehabilitation or imprudent return to training. However, the application of MRI to large animals is limited by costs and the size of the animal and most reports on the application of MRI to equine limbs have involved studies using cadaver limbs [7, 15]. In this study, we applied MRI systems with a permanent magnet to live horses affected with various stages from acute to chronic SDF tendinitis. In each case, MRI findings were compared to the corresponding ultrasonographic and histologic findings. The purpose of this study was to investigate the advantage of MRI over USD for the diagnosis of tendinitis and its potential clinical application to horses.

MATERIALS AND METHODS

Horses examined: Six racehorses (Thoroughbred, five males and one female, ages ranging from two to five years) were the subjects of this study (Table 1). All these horses suffered from SDF tendinitis during training or racing. Horses 1 and 2 had acute tendinitis with obviously swollen SDF tendon, while horses 3 and 4 were recurrent acute cases with previous histories of chronic SDF tendinitis. Horses 5 and 6 were chronic cases which had been stall rest given for three to four months respectively since the onset of tendinitis.

Ultrasonography: An ultrasound machine (LogiqTM500 MD, GE Yokogawa Medical Systems, Japan) with a 7.5 MHz transducer was used. After the palmar aspect of the third metacarpal bone was clipped, transverse images of seven regions (1A, 1B, 2A, 2B, 3A, 3B and 3C) [3] of the SDF tendon, from immediately below the accessory carpal bone to the level of the proximal sesamoid bones, were recorded with the horse standing.

MRI diagnosis: A 0.2-tesla MRI system with permanent magnet (MRP-20, Hitachi Medico, Japan) and a human flexible body coil with 20 cm diameter were used for this study. This system has the relatively large gantry bore size (38 × 100 × 133 cm, rectangular) to enable imaging the equine distal limb. After removal of the shoes, horses were
sedated with xylazine (1.0 mg/kg). General anesthesia was induced by ketamine hydrochloride (2.0 mg/kg) or 1g thiopental combined with 5% guaiacol glyceryl ether (GGE, 1,000 ml), and maintained with isoflurane anesthesia with positive pressure ventilation. Because of the metallic components of the anesthetic equipment influencing the magnetic fields, the anesthetic equipment was approximately 3 m away from the gantry [8]. The horses were positioned in left lateral recumbency on a movable table, then inserted into the MR gantry up to the mid-position of the radius. The flexible body coil was placed at the mid-position of the metacarpus. The pastern of the forelimb being examined was fixed with a rope to minimize limb’s movement associated with positive pressure ventilation. Spin-echo sequences were used and T1-weighted images (TR 500 ms, TE 25 ms) and T2-weighted images (TR 2000 ms, TE 110 ms) were obtained. Transverse images of the SDF tendons at seven identical portions to the ultrasound were obtained. All images were in 256 × 256 matrix format. The time required to complete the MRI examination was about 60 min. In all cases, MRI was performed within 72 hours of ultrasonography.

**Histologic examination:** Following the MRI, the horses were euthanased in a humane manner without recovering from anesthesia, in strict adherence with the protocol established by the Ethics Committee for Laboratory Animals of JRA Equine Research Institute. Tendon tissue was then collected from the same seven regions imaged, and preserved in 10% neutral buffered formalin. Following gross photography, sections of tendon containing representative areas of pathology were processed for routine histology, cut at 6 µm, and stained with hematoxylin and eosin (H & E). The tendon tissue collected from horses 5 and 6 (chronic tendinitis), were also stained with Alcian blue (AB; pH 2.5) in order to identify the presence of highly negatively charged matrix proteins (most frequently the large proteoglycans containing numerous glycosaminoglycan (GAG) side chains [10]).

**RESULTS**

Table 2 summarises the ultrasonographic, MRI, and associated histological findings, with respect to each case. In those cases of acute tendinitis, two separate well defined anechoic core lesions were identified ultrasonographically in horse 1 (Fig. 1-a). At the same site high signal intensity was obtained in both T1- and T2-weighted images (Fig. 1-c and d), precisely delineating the shape of the hemorrhage observed in the gross tissue (Fig. 1-b). In addition, in horse 2, hypoechoic areas on the ultrasonograph also corresponded to high signal intensity regions on the MR images. Histologic examination in horse 1 and 2 showed many red blood cells between the collagen fibers which had lost alignment, indicating of acute injury (Fig. 2).

In those cases of superimposed acute tendinitis on...
chronic damage, horse 3 showed several anechoic areas within a hypoechogenic area occupying ≥11% of the cross-sectional area of the tendon on ultrasound (Fig. 3-a). Correspondingly, high signal intensity was observed in T1- and T2-weighted images (Figs. 3-c and d). The ultrasonograph of horse 4 showed hypoechoic areas in the center and medial aspects of the tendon where high signal intensity was also observed in MR images. Gross findings of tendon tissue collected from horse 3 and 4 verified the presence of extensive hemorrhage in white hyperplastic granulation tissue (Fig. 3-b). Histologic characteristics of the hyperplastic granulation tissue included proliferation of fibroblasts and marked microvascularization (Fig. 4). These findings suggested that these cases had both chronic and acute inflammatory characteristics.

In the chronic tendinitis cases, the lesions in both horses 5 and 6 at the onset of SDF tendon injury had been recorded ultrasonographically as a large central hypoechogenic area (Fig. 5-a and Fig. 6-a) and also lacked a parallel fiber pattern in longitudinal images (Fig. 5-b). However, the echogenicity and the parallel fiber pattern of injury area had increased when ultrasound examinations were performed in this study after three and four months stall rest respectively (Figs. 5-c, -d and Fig. 6-b). Mildly hyperechoic areas indicating replacement with scar tissue were observed (Fig. 6-e). MR signals were observed in the same areas in both T1- and T2-weighted images, although the signal intensity was weaker compared to horses 1–4. Signal intensity observed in T1-weighted images of horses 5 and 6 were higher compared to those in T2-weighted images of the same horses (Figs. 5-e, -f and Figs. 6-c, -d). Scar tissue was found grossly (Fig. 6-e), with an irregular arrangement of collagen fibers and slight neovascularization observed microscopically (Fig. 7). In addition, the scar tissue was stained positively with Alcian blue (Fig. 6-f).

**DISCUSSION**

We applied MR imaging to six live horses with various stages of SDF tendinitis. The MRI system (0.2 T) used in this study was a low-field MRI that has the major advantages of being less costly and requiring less space. However, the image quality can be inferior to high field MRI in many, but not all, circumstances. In spite of the theoretical disadvantages, low-field MRI has been reported to be promising in the evaluation of human tendon disease [16].

Horses were immobilized with isoflurane general anesthesia. In this study, horses were euthanased after the MRI study to allow sample collection. However, for the clinical application of MRI in tendinitis, careful recovery techniques would be necessary to prevent possible further injury to the
SDF tendon during recovery.

In this study, the acute injuries (Horses 1 and 2) were delineated as areas of very high intensity MRI signal in both the T1- and T2-weighted images. In recurrent cases (Horses 3 and 4), which included both hemorrhage indicative of early inflammation stage and fibrosis indicative of chronic change, high intensity MRI signal in both the T1- and T2-weighted images were observed in those areas of recent injury. Even though it was possible to delineate the injured area precisely with MRI, there appeared to be little clinical benefit of MRI in the acute lesions because clearly hypoechoicity obtained by USD enable diagnosis of tendinitis.

In contrast, the healed areas of the tendons (Horses 5 and 6), were detected as weak MRI signals in both types of images. Scar tissue was visualized by MRI, even though the signal was low intensity, and it was easily distinguishable from the black background of the normal SDF tendon tissue. This study clearly demonstrated the presence of scar tissue with MRI where there were no clearly defined hypoechoic regions on ultrasonography.

The decrease in MRI signal intensity may indicate the progression of tendon fibrosis, as does increased echogenicity in ultrasonography. However, Crass et al. [2] reported that early fibrogenesis had significantly low signal in MRI although ultrasound findings still showed lucency, similar to the earlier more inflammatory phases of the healing process. Since our samples did not include same classifications of histologic findings in the healing tendon, a similar comparison could not be made. They also reported increasing MRI signals in later stages of fibrosis, in terms of scar tissue, which had very similar histologic findings to those observed in chronic cases in this study. The change of MR signal intensity obtained in the different healing stages (between early fibrogenesis and scar tissue) was less clear. We hypothesized the difference in GAG levels in the area of the lesion would influence MR signal intensity in the later stages of healing. There is an accumulation of large proteoglycans containing numerous GAG chains in healing scar tissue [9] and in the central core lesions of the SDF tendon [1]. GAGs play an important role associated with collagen synthesis and controlling the architecture of scar tissue [14]. These proteoglycans have a strong negative charge which is capable of binding water. In this study, the scar tissue in healing process was positive for Alcian blue, consistent with a greater GAG content than in surrounding normal tissue. MR findings also depicted precisely the dis-
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Therefore, GAG may influence water content of scar tissue and contribute to the change of MR signal intensity in the later stages of healing.

Movin et al. [13] reported that the contrast agent, gadolinium, binding with GAG by its negative charge enhanced the MR image contrast in human Achilles tendon core lesions. Accumulation of GAG in the scar tissue during the healing process of equine tendinitis was observed histologically in this study. Therefore, it is possible that the use of a contrast agent in the clinical application of MRI for diagno-

Fig. 5. The hypoechoic core lesion and lost fiber alignment were recorded at the onset of SDF tendinitis of horse 5 (a and b; white arrowhead). The echogenicity and the parallel fiber pattern of injury area had increased after three months stall rest (c and d; star). MR signals were observed in the same areas in both T1- and T2-weighted images (e and f; white arrow), the signal intensity was weaker compared to horse 1–4. S: Superficial Digital Flexor Tendon, D: Deep Digital Flexor Tendon, I: Inferior Check Ligament, SL: Suspensory Ligament, MC III: Third Metacarpal Bone.

Fig. 6. The hypoechoic core lesion was recorded at the onset of SDF tendinitis of horse 6 (a; white arrowhead). The echogenicity had increased after four months stall rest (b; star). Signal intensity observed in T1-weighted image was higher compared to that in T2-weighted image (c and d; white arrow). The hyperplastic granulation tissue was found glossy (e) and stained positively with pH 2.5 Alcian blue (f). S: Superficial Digital Flexor Tendon, D: Deep Digital Flexor Tendon, I: Inferior Check Ligament, SL: Suspensory Ligament, MC III: Third Metacarpal Bone.
ysis of chronic tendinitis, would greatly enhance the ability to visualize the scar tissue, especially where there is no clearly defined hypoechogenicity on ultrasonography.

One of the interesting findings in this study was that the MRI signals indicating injured tissue in all cases were stronger in T1-weighted images compared to T2-weighted images. The MRI signal obtained from the acutely injured tendon results from the presence of hemorrhage, inflammatory fluid and other water molecules within the damaged tissue. Thus, the stronger MRI signals in T1-weighted images were obtained in acute and acute recurrent cases. However, the reason why the MRI signals in chronically scarred tendon, where hemorrhage and hypervascularization typically is absent, were also stronger in T1-weighted images compared to T2-weighted images is unclear. The slight movement of the forelimb associated with mechanical ventilation during general anesthesia, although the distal limb was fixed in position with a rope around the pastern, was seen in this study. Thus the T1-weighted imaging, which has a shorter imaging time, may have provide an advantage for obtaining clear images of live animals under the MR conditions in this study. Further investigation of the ideal imaging conditions to obtain the clearest MR signals is warranted.

This report supports the findings of Crass et al. [2], which suggested that tendons containing mature scar tissue and inflamed tendons might be distinguished by MRI. In this study, the injured areas in the SDF tendon of all the horses, verified by gross and histological evaluation, were also observed as abnormal findings in MRI images. Our results therefore indicate that low-field MRI may be a useful imaging modality for the diagnosis of tendon injuries, particularly chronic lesions in horses.

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REFERENCES