



The role of DMSO and MSM in treatment of tendinopathies affection in equine: A comparative study

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Abstract

Superficial digital flexor tendon (SDFT) injury is a main cause of lameness in polo and racehorses inciting long-term lameness and recovery. This study aimed to compare the efficacy of topical dimethyl sulfoxide (DMSO) and Methylsulfonylmethane (MSM) on bowed tendons in horses. Ten client-owned injured horses, aged between 5-15 years with an average age of 8.5 years and weighing approximately 400-500 kg, suffering from chronic tendinitis were selected and allocated into two groups: group A (DMSO) and group B (MSM). The group A animals (n=5) were treated with 60 mL of topical 90% DMSO aqueous solution, while Group B animals (n=5) received 60 mL of topical 90% MSM aqueous solution on the bowed tendon at days 0, 7, 14, 21, 28, 35, and day 42. Clinical examinations were performed to evaluate the severity of pain and lameness grade. Ultrasound scans of the superficial digital flexor tendon were taken at days 0, 21, and day 42. Images were placed in numerical form, and the fiber alignment score of the fibers (FS) and the echogenicity score (ES) of the superficial digital flexor tendon at the maximum injury zone (MIZ) were recorded. Results showed that pain reduction and lameness significantly ($P<0.05$) reduced from 21d to 42d in the DMSO animals compared with the MSM animals. The echogenicity score didn't differ between the two groups at 0d, 21d, and 42d ($P>0.05$) however, it was significantly declined in group A at 42d than 0d ($P<0.05$). Fiber alignment in group A was ($P<0.01$) reduced at 42d compared with group B. This study concluded that group A horses showed very effective and long-term improvements compared to the group B horses.

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Introduction

Tendonitis of the SDF tendon is most frequent in polo and racehorses. Musculoskeletal injuries in flat racing horses are up to 82%. Injuries from musculoskeletal and tendon injuries are 46% (1,2). There are two kinds of connective tissues tendons and ligaments. Tendons and ligaments are involved in the movement of the skeleton, flexion and

extension of muscles, weight-bearing, and transmitting heat during the stance and swing phase of the horse (3). Tendons have different functions. Based on function, tendons are classified into two groups. First, the positional tendons are involved in the extension, rotation, and flexion of carpometacarpal, tarsometatarsal, and interphalangeal joints. Second, flexor tendons are so-called weight-bearing tendons positioned on the planter or palmer surface of the forelimb

and hind limb. Later, has more elasticity than the former hence functioning to store elastic energy weight (4). In horses, most of the bodyweight is loaded by SDFT, especially during the stance stage of gait. It is a very delicate structure prone to rupture during the gallop of canter (5). Due to improper management of horses after sports events, SDFT gets inflamed, termed as SDFT tendonitis. Tendonitis renders the horses lame for a long period of time (6). A thorough research plan regarding the treatment of tendon injuries has been developed but still, tendon cell responses to combinations of exercise-associated stresses are under investigation. Even though tendonitis results in the permanent end of the carrier in racehorse but some horses with SDFT injury undergo long-term treatment and show good response (7). Tendinitis is treated medically as well as surgically (8, 9). Modern medical treatment includes topical application of DMSO and PRP (10-13). Surgically tendonitis is repaired by desmotomy of check ligament or bifurcation of the tendon (14). Now there is a quest for new treatment protocols for the reduction of tendon inflammation. These treatment protocols would be cheap, easily applicable, and non-invasive. DMSO is readily dissolved in the aqueous medium. DMSO is efficacious in the reduction of musculoskeletal pain and inflammation (15). In equine practice, DMSO is an FDA-approved topical therapeutic agent (16). Therapeutically, it has penetration ability in biological membranes. In addition, DMSO has significance in the removal of free radicals, inflammatory mediators, and oxidants that result in the conversion of normal soft tissue into damaged tissue. Further, the pharmacological characteristics of DMSO are still unknown (17). As DMSO is cheap and easily accessible, it could be topically used for the reduction of pain and inflammation in tendon injuries such as bowed tendons. DMSO also promotes the healing of connective tissue. Above mentioned characteristics make DMSO a drug of choice in the treatment of SDFT injury (15). MSM is a well-known supplement in the diet of animals as it has anti-inflammatory effects, reduces pain, and has antioxidant properties. It is approved in animals as a therapeutic agent (18). MSM is a sanctioned supplement in horses. It is white in color and crystalline powder. Many pieces of evidence support the use of MSM. It reduces several indications of muscle stiffness and joint inflammation in equines (19). When researchers started to investigate MSM more closely; it was related to DMSO (dimethyl sulfoxide, or DMSO, containing one extra oxygen atom attached to a carbon atom). It has the same potential to therapeutically support racehorses (19).

To our knowledge, there is no study yet identified to compare the efficacy of DMSO and MSM for the treatment of SDFT injuries in horses in Pakistan. Therefore, this is the first investigation to characterize the comparative therapeutic efficacy of DMSO and MSM on bowed tendons in horses. We hypothesized that DMSO can be an effective

therapy for general causes of lameness present in the horses as compared to the MSM.

Materials and methods

Ethical statement

This study and all the procedures were approved and conducted following the rules and regulations of the ethical review committee (Ethical Approval No. DR/380; Dated: 08/09/2020) at the Department of Veterinary Surgery and Pet Sciences, University of Veterinary and Animal Sciences, Lahore, Pakistan.

Animals

10 adult athletic horses of Lahore Polo Club, UVAS Surgery Clinic, SPCA, and JHA Stud Farm Lahore suffering from SDF tendonitis were chosen for this study. The age of horses was between 5-15 years (8.55 ± 2.63). All the horses weighed approximately 400-500 kg (4.46 ± 35.33). Before the start of experiments, Horses were acclimatized for seven days to ensure abundant accessibility of grass hay *ad libitum* and sufficient clean drinkable water. All the horses under investigation were kept in separate stalls (20).

Experimental design and treatments

Horses were randomly categorized into two groups: group A (DMSO; n=5) and group B (MSM; n=5). The group A animals received 60 mL of topical 90% DMSO aqueous solution (Dimethyl sulfoxide Reagent Plus[®] SIGMA Life Science, Pvt. Ltd. Pakistan) on the bowed tendon rubbed for 15 min once daily at days 0 to 7. Similarly, group B animals received 60 mL of topical 90% MSM aqueous solution (MSM power Liniment[®] Ghazi Brothers, Pvt. Ltd. Pakistan) on the bowed tendon and rubbed over 15 min once daily at days 0 to 7.

During the pretreatment, horses were examined by lifting up forelimb, palpation of the tendon injury site, and by walking and trotting. Before the start of treatment, the caudal side of the horse's forelimb was shaved with a sterilized razor. During the experiments, all horses were individually put up into stalls on a hard floor with wood shavings and were accessed to a balanced ration concentrate and hay *ad libitum*. After topical administration of DMSO and MSM, the SDFT tendon was wrapped externally without surgical manipulation with a medical-graded plastic sheet and crap bandage. The bandage was wrapped around the tendon for 12 h, So that maximum exposure of DMSO and MSM should be made over a large surface area. The prescribed dosage was measured in a graduated glass container. In clinical cases, it was applied with a cotton-tipped applicator.

Diagnosis and clinical evaluation parameters

SDF tendonitis was diagnosed by visual examination by lunging, locally palpating, and trotting animals in a straight

line in case of bowed tendon conformation. Furthermore, a confirmatory diagnosis of tendon injury was made with ultrasonography on day 0. Lameness and severity of pain were assessed by the simple descriptive scale (SDS) and the lameness scale of the American Association of Equine Practitioners (AAEP). According to AAEP, the lameness scale was described as grade 0 indicated no detectable lameness, grade 1 represented hardly detectable lameness, grade 2 was denoted mild lameness at a walk in the stall, grade 3 designated moderate lameness, but not non-weight bear, grade 4 represented by severe lameness and grade 5 was non-weight bearing 100% of the time. According to a simple descriptive pain scale, the level of pain is graded as 0= no pain, 1= slight pain, 2= mild pain, 3= moderate pain, and 4= severe pain (21). Clinical evaluations were carried out on days 0, 7, 14, 21, 28, 35, and day 42. The degree of lameness in each lame horse was evaluated by walking and trotting the animal on the ground of the University Surgery Clinic. To identify lameness due to tendons, a high palmer nerve block is used.

Ultrasonography

Ultrasonographic evaluations were carried out by using Visionstar® DW-VET6 Veterinary ultrasound portable ultrasound-unit (Chongqing Vision Star Optical Co, Ltd). Ultrasound scans were taken by using a high-frequency 7.5 MHz linear array transducer. Before ultrasonography, the affected limb of each horse was shaved and cleaned with a scrub solution. To achieve aseptic conditions, a coat of methylated spirit was applied along with scrubbing. On the target site, the gel was rubbed to obtain an optimized clear ultrasound scan. At the owner's request, shaving was excluded before ultrasonography (22) and scrubbing and methylated spirit were only used along with gel application on the affected limb area. Before ultrasonography, all animals were brought to an upright standing position for equal limb weight-bearing (23) to minimize changes and differences in ultrasound images (22).

Superficial digital flexor tendonitis ultrasound examination was made by exploiting zone A just proximal to the fetlock joint (24,25). Ultrasound findings were measured by using parameters such as fiber pattern and echogenicity scoring. Echogenicity was scored as 0 designated normal tendon echogenicity, 1 designated 25%-50% tendon has lost echogenicity, 2 stands for half of the tendon appearing echoic and 3 refers to the entire tendon seeming to be anechoic respectively (26,27). Fiber arrangement was evaluated based on either regular or irregular fiber alignment. Homogeneity was measured as either ultrasound scans being homogeneous or heterogeneous (28). All sonographic evaluations were assessed on days 0, 21, and day 42 respectively (29).

Statistical analysis

All data were statistically analyzed by independent-sample t-test using SPSS 16.0 statistical software. All results were expressed as mean \pm SE; $P>0.05$ shows no significant difference, $P<0.05$ shows a significant difference, and $P<0.01$ shows that the difference was extremely significant. All the graphs were formed using the Graph-pad Prism (Version 7.04, Graph Pad Software Inc., San Diego, CA) (30).

Results

Lameness scoring

Comparative statistical analysis of lameness reduction in both groups indicated that lameness reduced significantly ($P<0.05$) on 21d to 42d in 90% DMSO aqueous solution group compared with 90% MSM aqueous solution group as indicated in (Figure 1A).

SDS pain scoring

Pain reduction scores in group A and group B revealed that pain was significantly ($P<0.05$) decreased from 21d to 42d in group A as compared to group B as shown in (Figure 1B).

Echogenicity scoring

In the 90% DMSO aqueous solution group, a reduction in echogenicity was not observed between 0d and 07d, and 21d ($P>0.05$). Furthermore, the echogenicity score significantly declined at 42d than 0d ($P<0.05$). In the 90% MSM aqueous solution group, a non-significant decrease was seen between 0d and 42d ($P>0.05$). Consequently, the difference in echogenicity scoring was not significant ($P>0.05$) between the two groups on 0d, 21d, and 42d as indicated in (Figures 2 and 3).

Fiber alignment

In group-A horses, the change in fiber alignment scoring was not significant between 0d and 21d ($P>0.05$), which means fiber alignment was not observed at day 21, but fiber alignment was evident at 42d ($P<0.01$). In the horses of group B, the arrangement of fibers reduced non-significantly from 0d to 42d ($P>0.05$). However, the change in fiber alignment scoring was not seen at 0d and 21d ($P>0.05$), but it inclined to be decreased ($P<0.01$) in DMSO 90% aqueous solution (group A) than the MSM 90% aqueous solution group on 42d ($P=0.002$) as shown in (Figures 2 and 4).

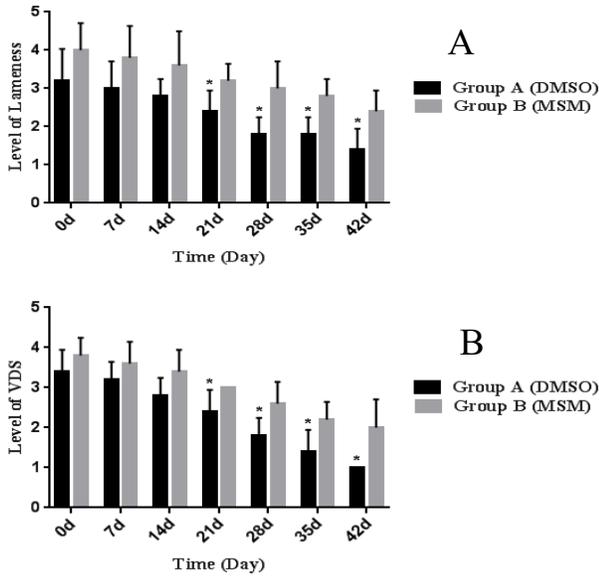


Figure 1: Clinical Evaluation indices: A- level of lameness was determined using AAEP lameness scale. DMSO group had lower level of lameness score than MSM group on days 7-42, *P<0.05, B- level of pain was determined using VDS (Visual Descriptive pain scale). DMSO group had lower level of pain score than MSM group on days 7-42, *P<0.05. Clinical evaluations were carried out on 0d, 7d, 14d, 21d, 28d, 35d, and 42d. Data are presented as mean ± SEM.

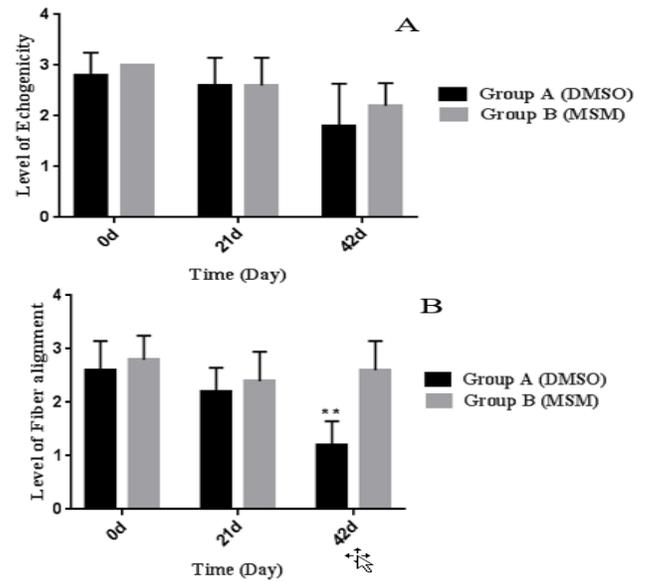


Figure 2: Ultra-sonographic indices: A- Level of Echogenicity was determined using echogenicity scale. DMSO group had lower level of lameness score than MSM group on day 42, P>0.05, B- Fiber pattern was determined using Fiber pattern scale. DMSO group had highly lower level of pain score than MSM group on days 42, **P<0.01. Ultra-sonographic examination was carried out on 0d, 21d, and 42d. Data are indicated as mean ± SEM.

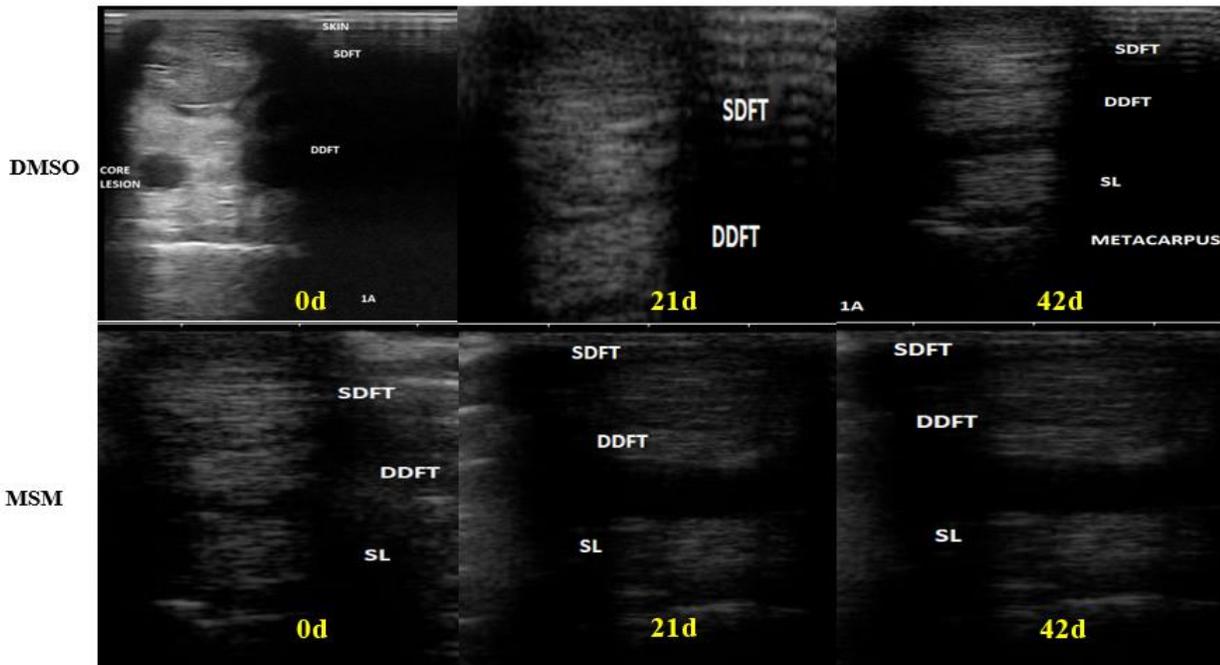


Figure 3: Transverse ultrasound scans of SDFT show changes at 0d, 21d and 42d.

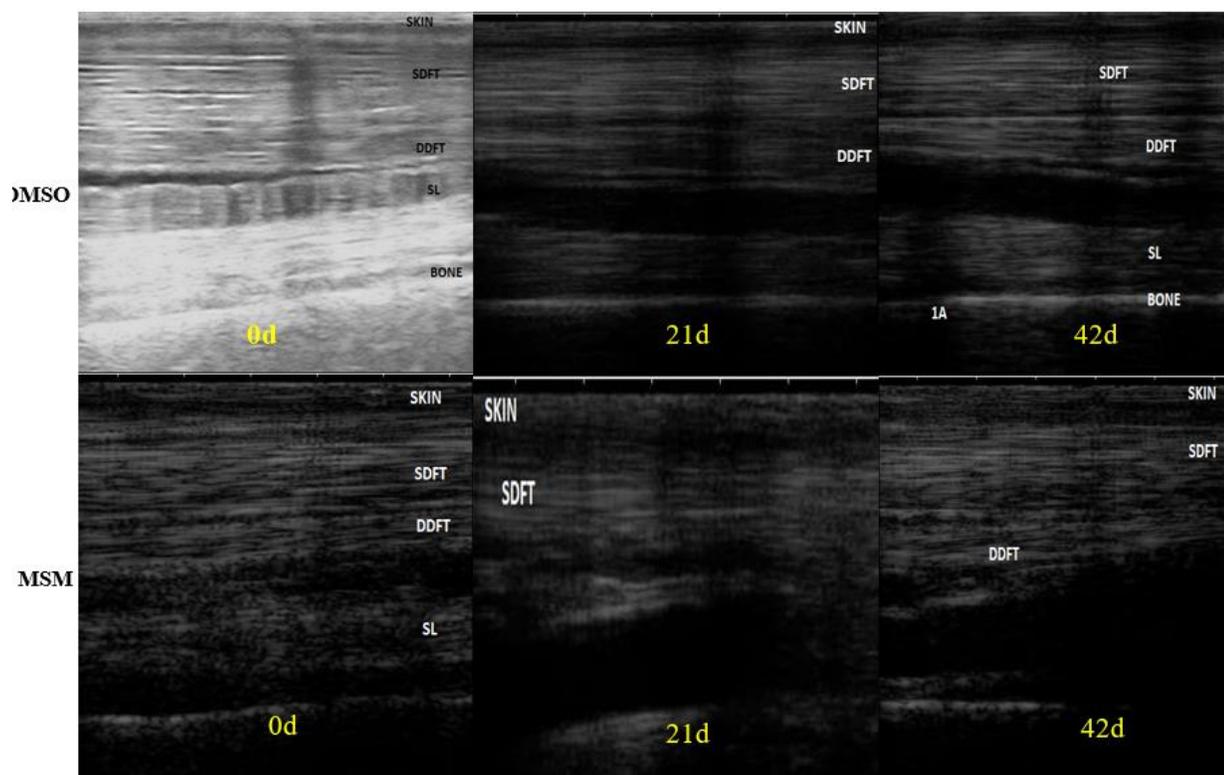


Figure 4: Longitudinal ultrasound scans of SDFT show fiber pattern changes overtime.

Discussion

Superficial and deep digital flexor tendinopathies often lead to joint injuries resulting in severe lameness along with the end of a carrier in the draft, sports, and dancing horses (31,32). Tendonitis is most commonly related to inflamed, bowed, or deformed overextended and painful tendons leading to poor performance and the appearance of abnormalities in sonographic scans (33). Superficial Digital Flexor Tendinopathies (SDFT) particularly in forelimbs are frequently seen (28,31), with an increased rate of recurrence (34). To get remedy these problems many treatment protocols have been formulated. Therefore, it is a need for time to seek novel therapeutic agents in the treatment of SDF tendinopathies.

DMSO is highly water-soluble, dipolar, without proton releasing, and the hygroscopic compound which acts as a good anti-inflammatory agent in case of sprains, tendonitis, bursitis, and disorders related to soft tissue inflammation and hematoma. DMSO is applied topically along with antibiotics and steroids to promote the healing of dermatitis arthritis, mastitis, and tendonitis (35). MSM is used orally as a supplement. It is taken as the therapeutic entity in numerous diseased conditions such as in the reduction of pain, swelling, allergic reactions, arthritis, and bacterial infections (36-38).

DMSO and MSM are very effective therapeutic agents currently known in equine SDF tendinopathies. According to our study, this was the first study to investigate the comparative therapeutic efficacy of DMSO and MSM in the treatment of bowed tendons in equines.

Previous research studies have indicated that lameness is considerably decreased in equines from day 21 to day 42. On 42day significant results are seen. At 60 days, lameness is declined by decreasing tendon inflammation with the topical use of DMSO (39-41). In our study, the lameness score was reduced significantly in the DMSO animal than that of MSM animals on the day 21 to day 42 time points. In the MSM group, the lameness score was reduced non-significantly. Many other research scholars summarized the same results as lameness score is reduced after administering DMSO and MSM agents (14,20,42,43). Likewise, the SDS was built to show that the severity of pain decreased significantly between the two groups on day 21 to day 42 time points. These findings have also been supported by many other research scholars using conventional therapeutic protocols in the treatment of naturally occurring SDF tendinopathies (14,20,44).

In horses, lameness is initially diagnosed by history, clinical examination, and local nerve blocking at the site of tendon injury. Radiography is also a key diagnostic tool for

the evaluation of hard tissue abnormalities; however, soft tissue injuries cannot be diagnosed by radiography (45). This adds to the significance of ultrasonography as a diagnostic tool. As ultrasound is non-invasive and portable, additionally equipped with better visual images and more accurate diagnosis of tendon injuries. These distinctive features of ultrasound outstrip the clinical value of radiography (45,46). Furthermore, the diagnosis of the tendon healing process by ultrasound scanning is an auspicious technique in equine medicine (47).

In our study, ultrasonographic factors were the echogenicity score (ES) of tendon and the fiber alignment of tendon reduced in the study indicating improvement of healing in Group-A and Group-B. Moreover, the difference in echogenicity in both groups was non-significant and ultrasonographic variables were improved. A similar outcome has been reported by other research scholars (20). Furthermore, the fiber alignment score of the fiber declined during day 42 and the change in both groups was significant on day 42 supporting the DMSO group A. The improvement of ultrasonographic variables during trial periods of study has been investigated by other researchers (20).

Conclusion

We concluded that DMSO could decrease the severity of pain and lameness more effectively as compared to MSM during the treatment of bowed tendons in horses. This study also caused an improvement in ultrasound parameters in tendon scanning in the DMSO animals compared with the MSM animals. In the future, further long-term investigations should be planned in a higher quantity of animals to investigate the effect on recurrence rate and develop strategies to control tendinopathies in horses.

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Conflict of interest

All authors declared no any conflict of interest.

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دور الـ DMSO و MSM في علاج اصابات الاوتار في الخيول: دراسة مقارنة

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الخلاصة

تعد إصابة وتر العضلة القابضة الاصبعية السطحية (SDFT) سبب رئيسيا للرج في خيول لعبة البولو وخيول السباق، وان هذا العرج يكون مزمنًا ويحتاج لفترة طويلة للتعافي. هدفت هذه الدراسة لمقارنة فعالية كل من ثنائي ميثيل سلفوكسيد (DMSO) و ميثيل سلفونائل ميثان (MSM) والمستخدم موضعيا على منطقة الأوتار المصابة في الخيول. وقد تم اختيار عشرة من الخيول المصابة والمملوكة لأصحابها المراجعين، تتراوح أعمارها ما بين ٥-١٥ سنة وبمتوسط عمر ٨,٥ سنة ووزنها حوالي ٤٠٠-٥٠٠ كغم، ان هذه الخيول كانت تعاني من التهاب الأوتار المزمن حيث تم تقسيمها إلى مجموعتين: مجموعة أ (DMSO) ومجموعة ب (MSM). عولجت حيوانات المجموعة أ (عددها = ٥) بـ ٦٠ مللتر من محلول مائي ٩٠٪ DMSO موضعيا، بينما تلقت حيوانات المجموعة ب (عددها = ٥) ٦٠ مللتر من محلول مائي ٩٠٪ MSM موضعيا على منطقة الوتر المصاب للأيام ٠، ٧، ١٤، ٢١، ٢٨، ٣٥،

المجموعة أ مقارنة مع حيوانات المجموعة ب. هذا ولم تختلف درجة تجانس الصدى معنويًا عند مستوى ($P > 0.05$) بين المجموعتين للفترات ٠، ٢١، و ٤٢ يوم، ومع ذلك فقد انخفضت بشكل ملحوظ في المجموعة أ عند ٤٢ يوم عنه من اليوم ٠ ($P < 0.05$). وقد تم تقليل محاذاة الألياف في المجموعة أ عند مستوى معنوي ($P < 0.01$) لفترة الفحص ٤٢ يوم مقارنة بالمجموعة ب. خلصت هذه الدراسة إلى أن خيول المجموعة أ أظهرت تعزيزًا فعالة جدًا وطويلة المدى مقارنةً مع خيول المجموعة ب.

و ٤٢ يوم من بدء العلاج. تم إجراء الفحوصات السريرية لتقييم شدة الألم ودرجة العرج، مع إجراء فحص بالموجات فوق الصوتية لوتر العضلة القابضة الاصبعية السطحية في الأيام ٠ و ٢١ و ٤٢. حيث تم وضع صور الفحص بالأمواج فوق الصوتية بشكل رقمي، وقد تم تسجيل درجة محاذاة الألياف للألياف (FS) ودرجة تجانس الصدى (ES) لوتر العضلة القابضة الاصبعية السطحية في منطقة الإصابة القصوى (MIZ). أظهرت النتائج أن تقليل الألم والعرج انخفض معنويًا عند مستوى ($P < 0.05$) من الفترة ٢١ يوم إلى ٤٢ يوم من بدء العلاج في حيوانات