Manuscript Details

Manuscript number	JEVS_2017_309
Title	Ultrasonography appearance of the equine proximal suspensory ligament and proximal palmar metacarpal structures after local anesthetic infiltration
Article type	Research paper

Abstract

Introduction. Suspensory ligament diseases are common sources of lameness in horses. Once injury of the proximal suspensory ligament is suspected, based on the clinical examination and response to analgesia, diagnostic imaging of this region is often performed. The aim was to describe ultrasonographic changes in the palmar metacarpal area after 20 minutes and at 24 hours post infiltration of local anesthetic solution using the lateral approach. Materials and methods. In this study, suspensory ligament and proximal palmar metacarpal structures were ultasonographically evaluated before and after perineural injection of mepivacaine 2% in eight forelimbs. The limbs were prospectically divided into two groups: Group 1: needle not filled with anesthetic (right forelimbs); Group 2: needle filled with anesthetic (left forelimbs). The ultrasonographic examination of the palmar metacarpal structures was performed after 20 minutes and 24 hours post-injection to evaluate qualitative post-injection changes and to perform measures. Results. In this study there were no significant differences in the SL and other palmar metacarpal structures between baseline measurement and pattern and post-injection measurements. No changes in echogenicity or fiber patterns of the tendons and ligaments during the scans were found. Alterations compatible with gas echoes were detected in 1 limb owned to group 1 at 20 minutes post-injection, while no gas patterns were visualized at 24 hours. Discussions. Soft tissue changes, due to local anesthetic infiltration, may be present. This suggest that diagnostic ultrasonography of the origin of the SL should be interpreted with caution if performed within 24 hours after diagnostic analgesia.

Keywords	Horse, Suspensory Ligament, perineural anesthesia, ultrasonography

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Dear Editor,

here is our paper titled "**Ultrasonography appearance of the equine proximal suspensory ligament and proximal palmar metacarpal structures after local anesthetic infiltration"** and authored by Rinnovati et al. The Ethical Committee, University of Pisa, approved this study. The project was supported by funds from the University of Pisa (100%). The manuscript has not been published or submitted for publication elsewhere. Authors' contribution to the manuscript is equally distributed and no conflict of interest exists.

Yours sincerely,

Prof. Micaela Sgorbini

1) The aim of this study was to describe ultrasonographic changes in the palmar metacarpal area after 20 min and at 24 hours post infiltration of local anesthetic solution using the lateral approach.
2) We did not find any differences in measures expressed in cm and obtained in the cross-sectional scan at the origin of the suspensory ligament.
3) No subjective changes noted in echogenicity or fiber pattern of the tendons and ligaments are noted.
4) We detected hyperechoic areas with attenuation resulting in acoustic shadowing and variable reverberation distally, interpreted as gas, only in one limb injected with an empty needle, at 20 minutes, but not at 24 hours post-injection.
5) We did not visualize gas echoes in any of the legs injected with a needle full of anesthetic.

Ultrasonography appearance of the equine proximal suspensory ligament and proximal palmar metacarpal structures after local anesthetic infiltration

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Abstract

Introduction. Suspensory ligament diseases are common sources of lameness in horses. Once injury of the proximal suspensory ligament is suspected, based on the clinical examination and response to analgesia, diagnostic imaging of this region is often performed. The aim was to describe ultrasonographic changes in the palmar metacarpal area after 20 minutes and at 24 hours post infiltration of local anesthetic solution using the lateral approach.

Materials and methods. In this study, suspensory ligament and proximal palmar metacarpal structures were ultasonographically evaluated before and after perineural injection of mepivacaine 2% in eight forelimbs. The limbs were prospectically divided into two groups: Group 1: needle not filled with anesthetic (right forelimbs); Group 2: needle filled with anesthetic (left forelimbs). The ultrasonographic examination of the palmar metacarpal structures was performed after 20 minutes and 24 hours post-injection to evaluate qualitative post-injection changes and to perform measures.

Results. In this study there were no significant differences in the SL and other palmar metacarpal structures between baseline measurement and pattern and post-injection measurements. No changes in echogenicity or fiber patterns of the tendons and ligaments during the scans were found. Alterations compatible with gas echoes were detected in 1 limb owned to group 1 at 20 minutes post-injection, while no gas patterns were visualized at 24 hours.

Discussions. Soft tissue changes, due to local anesthetic infiltration, may be present. This suggest that diagnostic ultrasonography of the origin of the SL should be interpreted with caution if performed within 24 hours after diagnostic analgesia.

Key words. Horse, Suspensory Ligament, perineural anesthesia, ultrasonography.

1. INTRODUCTION

Local or regional anesthesia is commonly used in horses to localize the source of lameness or to desensitize a surgical site [1]. Regional, local and intra-articular anesthesias are all methods to perform diagnostic anesthesia with increasing degrees of specificity, respectively.

Regional anesthesia is defined as the nerve block of local anesthetic solution. Local anesthesia refers to direct infiltration of anesthetic solution within the tissues of interest, and intra-synovial anesthesia is achieved by injecting local anesthetic directly within a joint or other synovial structure [2].

Suspensory Ligament (SL) diseases are common sources of lameness in horses and often occur for SL related problems [1,2]. In particular, injuries of the proximal portion of the SL are common in sports horses and are often associated with chronic continuous injuries [3]. The origin of the SL can be desensitized by direct infiltration or by blocking the lateral palmar nerve before it branches into the lateral and medial palmar metacarpal nerves [1].

Once injury of the proximal portion of the SL is suspected, based on the clinical examination and response to analgesia, diagnostic imaging of this region is often performed [2-5]. Radiography can be used to evaluate the proximal palmar aspect of the third metacarpal bone for evidence of sclerosis, lysis, proliferation, or avulsion fracture at the attachment of the suspensory ligament [6]. Ultrasonography has traditionally been the imaging modality of choice for diagnosis of the proximal portion of the SL injury [5]. To the authors' knowledge, only one study [7] on ultrasonographic changes of SL after peri-neural injection of anesthetic solution have been published. In particular, the authors performed a sub-carpal high palmar and palmar metacarpal, and low palmar and palmar metacarpal nerve blocks. Thus, the aim of this study was to describe ultrasonographic changes in the palmar metacarpal area after 20 min and at 24 h post infiltration of local anesthetic solution using the lateral approach proposed by others [7].

2. MATERIALS AND METHODS

Four adult trotter horses, median age 13 years and weighing 430-550 kg with a median Body Condition Score of 4/5 [8] were included in this prospective study. Horses from the Veterinary Teaching Hospital, University of Pisa herd were used. This study was approved by the Ethics Committee on Animal Experimentation, the University of Pisa (prot n. 14024, 14.02.2015).

Animals were given a brief physical examination with palpation of the forelimbs and examination for lameness at the walk, as a baseline to aid in monitoring for any complications secondary to the injection procedure. The horses were housed in single 4x4 meters boxes for all the study period and were not submitted to exercise.

The palmar aspect of both metacarpal regions was clipped from the accessory carpal bone to the ergot and from the medial and lateral sides distally over the branches of the SL. Animals were sedated with an alfa$_2$-agonist (detomidine 20 mcr/Kg ev) to minimize stress related to the procedure, and no nose twitch was used. Ultrasound examination was performed with a My Lab 30 Gold ultrasound machine (Esaote, Florence, Italy) and a linear probe (7.5 mHz); standoff pad was not used during the ultrasonographic examinations. Each limb was moistened with warm water, and an acoustic coupling gel applied to improve imaging. All proximal palmar metacarpal area and in particular SL was evaluated ultrasonographically, both in transverse and longitudinal scans, from the origin to the bifurcation in the lateral and medial branches to verify alterations. This qualitative evaluation was performed with the limbs fully weight bearing and with the limbs lifted and the hoof fully flexed, as previously suggested [2].

Then, baseline measures (T0) were obtained with the limb weight bearing and in longitudinal scan at the proximal portion of the SL insertion. The measures recorded were: a) distance between third metacarpal bone and the SL; b) dorso-palmar thickness of SL; c) distance between the SL and the accessory ligament deep digital flexor tendon (DDFT); d) dorso-palmar thickness of accessory ligament (AL) of DDFT; e) dorso-palmar thickness of DDFT; f) dorso-palmar thickness of superficial flexor tendon (SFT). The echogenic pattern of the SL was evaluated; special attention was paid to the possible presence of abnormal fluid or gas echoes.

The proximal portion of the SL area of both forelimbs was prepared for aseptic injection technique: a 10-minute scrub in a circular motion using gauze sponges soaked in povidone–iodine 10% was performed. Excess povidone–iodine was removed by a single 70% isopropyl alcohol wipe with a gauze sponge.

The metacarpal nerves were anesthetized using the lateral approach as described [7]: a 3.8 cm, 20 gauge needle was inserted axial to the fourth metacarpal bone perpendicular to the palmar cortex of the third metacarpal bone, until it hits the bone; 2.5 ml of mepivacaine chloride 2% (Industria Farmaceutica Galenica Senese, Italy) were then injected. The needle was partly withdrawn and then redirected in a dorso-medial direction, towards the junction of the second and third metacarpal bones. When the needle was fully inserted (contacted bone), 2.5 ml of mepivacaine chloride 2% (Industria Farmaceutica Galenica Senese, Italy) were injected. Limbs were divided previously in

two groups of four legs each. Group 1: needle not filled with anesthetic (right forelimbs); Group 2: needle filled with anesthetic (left forelimbs).

Then, the ultrasonographic examination of the palmar metacarpal structures was performed after 20 minutes (min) and 24 hours (h) post injection to evaluate qualitative post-injection changes and to perform measures, as already described for baseline assessment.

A routine standing leg bandage was applied to both forelimbs between 20 minutes (T20min) and 24 hours (T24h) ultrasonographic examinations in all horses. Evaluation for any signs of lameness or inflammation in the leg was performed 24, 48 and 72 hours after the nerve blocks.

2.1 Statistical analysis

All the measures recorded were expressed as average and standard deviation. Data were evaluated for distribution using the Kruskal-Wallis test. Data did not show a Gaussian distribution, thus the Friedmann test and the Dunnett's multiple comparison test as *post hoc* were applied to verify differences for all the measures carried out in relation to time (T0 vs T20min vs T24h), both for group 1 and 2. Significance was set at $P<0.05$.

3. RESULTS

A total of 8 forelimbs were examined. No horses developed lameness after injection or during the period of observation. All measures are reported in Table 1, both for group 1 and 2. The baseline qualitative evaluation of the SL showed no alterations in all the forelimbs. No significant differences were found for all the measures at any post-injection ultrasound examinations compared to the baseline, both for group 1 and 2. A qualitative increased definition of the proximal aspect of the SL due to a decreased echogenicity of the space between the third metacarpal bone and the SL itself were found in 4/8 limbs (1/4 for group 2 and 3/4 for group 1) at 20 min post-anesthetic injection, while at 24h ultrasound images were similar to the baseline ones. No changes in echogenicity or fiber patterns of the tendons and ligaments during the scans were found. Alterations compatible with gas echoes were detected in 1/4 limb owned to group 1 at 20 min post-injection, while no gas patterns were visualized at 24 h (fig. 1).

4. DISCUSSION AND CONCLUSIONS

The evaluation of equine lameness often involves peri-neural or local anesthesia. Though complications are uncommon, local anesthetic solutions can cause tissue irritation at the site of injection [10]. Moreover, the deposition of anesthetic solutions in peri-neural soft tissues has been visualized during ultrasound-guided nerve blocks in humans [11].

High quality ultrasonographic images are essential for accurate diagnosis of SL diseases. Large vessels plantaro-lateral to the SL may result in broad linear anechoic artefacts within the SL, which complicate interpretation. In large Warmblood horses in particular the SL is situated deeply and the ultrasound transducer must be focused accordingly. The SL should be imaged in both transverse and longitudinal planes. To examine the proximal portion of the SL in transverse images, it may be helpful to rock the transducer slightly medially and laterally in order to obtain the best quality images. In the healthy horse, the proximal portion of the SL is more uniformly echogenic than in the forelimb. In the hindlimbs, proximal portion of the SL focal anechoic areas are relatively unusual. More commonly there is enlargement of the SL, with poor demarcation of its borders and a diffuse reduction in echogenicity of part or all of the cross-sectional area of the ligament. Ectopic mineralization occurs more often in hindlimbs compared to forelimbs. An irregular contour of the plantar contour of the third metatarsal bone may reflect entheseophyte formation. This explains how important it is to evaluate the structures of this region accurately even after anesthetic infiltration [1].

The aim of this study was to describe ultrasonographic changes in the palmar metacarpal area after T20min and at T24h post-infiltration of local anesthetic solution using the lateral approach. To the authors' knowledge this is the first report that evaluated alterations after T20min and T24h anesthetic injection using one of the approach proposed by others [7] and reported measured of the ligaments. A previous study [7] evaluated the ultrasonographic changes occurred after 10 minutes and 24 hours post-anesthetic injection using the sub-carpal high palmar and palmar metacarpal, and low palmar and palmar metacarpal approaches. The authors found no significant differences in the cross-sectional area of tendons and ligaments between baseline and post-injection measures expressed in mean pixel value. In the present study, the authors did not find any differences in measures expressed in cm and obtained in the cross-sectional scan at the origin of the SL. In this study, no subjective changes noted in echogenicity or fiber pattern of the tendons and ligaments are noted, in line with others [7].

Hyperechoic areas with attenuation resulting in acoustic shadowing and variable reverberation distally, interpreted as gas, were detected only in one limb injected with an empty needle at T20min, but not at T24h post-injection; no gas echoes was visualized in any of the legs injected

with a needle full of anesthetic. In the previous study, hyperechoic areas compatible with gas were reported in all horses in at least one site post-perineural injection at 1 hour post-injection scan, while no changes were found at 24 hours. In the study, the authors did not specify if the needle used for injection was empty or full of anesthetic. Thus, the differences could be due to the different injection approach and to the fully/empty needle.

In conclusion, soft tissue changes, due to local anesthetic infiltration, may be present. This suggest that diagnostic ultrasonography of the origin of the SL should be interpreted with caution if performed within 24 hours after diagnostic analgesia.

GROUP 1	TMB-SL			SL			SL-AL			AL			DDFT			SFT		
Horses	**T0**	**T20min**	**24h**	**T0**	**T20min**	**T24h**	**T0**	**T20min**	**T24h**	**T0**	**T20min**	**T24h**	**T0**	**T20min**	**T24h**	**T0**	**T20min**	**T24h**
1	0.24	0.33	0.29	0.92	0.89	0.87	0.28	0.27	0.24	0.48	0.53	0.52	0.88	0.74	0.61	0.85	0.96	0.80
2	0.20	0.29	0.24	0.99	1.07	0.95	0.27	0.24	0.18	0.47	0.51	0.55	0.76	0.84	0.78	0.70	0.62	0.73
3	0.25	0.30	0.34	1.02	1.08	0.93	0.18	0.21	0.18	0.51	0.51	0.48	0.61	0.61	0.66	0.65	0.68	0.69
4	0.30	0.39	0.20	0.81	0.88	0.97	0.16	0.12	0.15	0.41	0.37	0.38	0.54	0.56	0.56	0.65	0.73	0.60
X	0.20	0.30	0.30	0.90	1.00	0.91	0.20	0.20	0.20	0.50	0.50	0.50	0.7	0.70	0.70	0.70	0.70	0.70
SD	0.00	0.00	0.10	0.04	0.10	0.01	0.10	0.10	0.00	0.00	0.10	0.10	0.20	0.10	0.10	0.10	0.10	0.10

GROUP 2	TMB-SL			SL			SL-AL			AL			DDFT			SFT		
Horses	**T0**	**T20min**	**24h**	**T0**	**T20min**	**T24h**	**T0**	**T20min**	**T24h**	**T0**	**T20min**	**T24h**	**T0**	**T20min**	**T24h**	**T0**	**T20min**	**T24h**
1	0.37	0.39	0.39	0.98	0.90	0.90	0.18	0.24	0.20	0.61	0.56	0.54	0.75	0.87	0.62	0.57	0.74	1.02
2	0.23	0.32	0.23	0.95	0.98	0.97	0.14	0.15	0.10	0.51	0.45	0.45	0.78	0.82	0.55	0.65	0.56	0.74
3	0.29	0.30	0.36	1.05	1.05	0.98	0.19	0.20	0.17	0.53	0.50	0.50	0.66	0.71	0.62	0.75	0.83	0.60
4	0.35	0.15	0.31	0.96	0.98	0.97	0.09	0.09	0.10	0.34	0.51	0.37	0.88	0.84	0.59	0.69	0.67	0.74
X	0.30	0.30	0.30	1.00	1.00	1.00	0.20	0.20	0.10	0.50	0.50	0.50	0.80	0.80	0.60	0.70	0.70	0.80
SD	0.10	0.10	0.10	0.10	0.10	0.00	0.00	0.10	0.10	0.10	0.00	0.10	0.10	0.10	0.00	0.10	0.10	0.20

Table 1. TMB-SL: distance between the TMB and the SL; SL: dorso-palmar thickness of SL; SL-AL: distance between the SL and the AL; d) dorso-palmar thickness of the AL; e) dorso-palmar thickness of DDFT; f) dorso-palmar thickness of SFT.

Legend - TMB: Third Metacarpal Bone; SL: Suspensory Ligament; DDFT: Deep Digital Flexor Tendon; AL: Accessory Ligament of the DDFT; Superficial flexor tendon (SFT); X: average; SD: Standard Deviation.

Fig. 1. Ultrasound images of the proximal palmar metacarpal area obtained in a horse included in group 1. Longitudinal scan. A) Baseline ultrasound examination (T0); B) Ultrasonographic examination performed 20 minutes post perineural anesthesia (T20min): alterations compatible with gas echoes are present between the AL and the DDFT (arrow); C) Ultrasonographic examination performed 24 hours post perineural anesthesia (T24h): no gas patterns are present.

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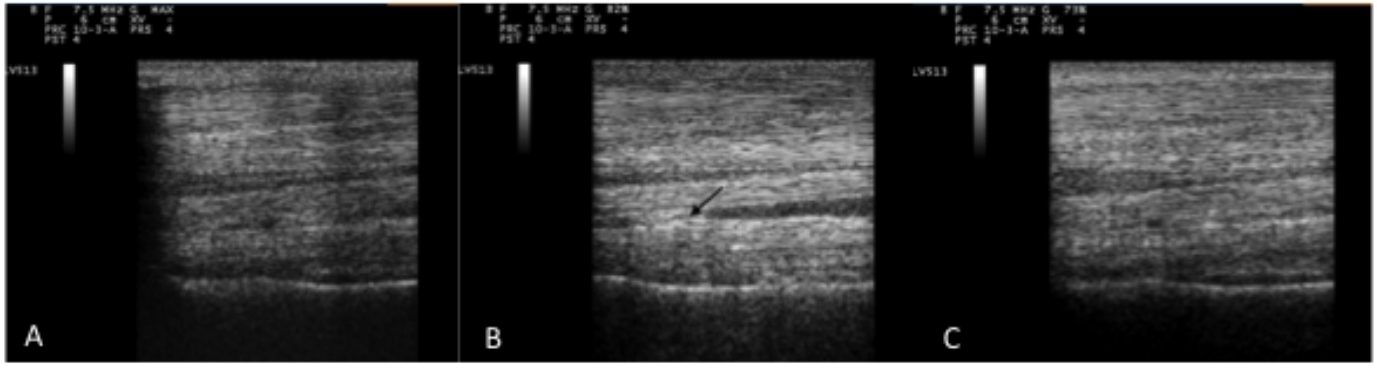
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Dear Editor,

The Authors' contribution to the manuscript is equally distributed and no conflict of interest exists.

Yours sincerely,

Prof. Micaela Sgorbini

Dear Editor,

this *in vivo* study was approved by the Institutional Animal Care and Use Committee of the University of Pisa (D.R. prot n. 14024, 14.02.2015). The University of Pisa owned the horses included in the study.

Yours sincerely,

Prof. Micaela Sgorbini