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Ultrasonography of the cervical articular processes in donkeys and mules: technique and reference images

[Ultrassonografia dos processos articulares cervicais em asininos e muares: técnica e imagens de referência]

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ABSTRACT

In veterinary medicine, there are few descriptions in scientific literature referring ultrasound techniques in the cervical region of mules and donkeys. Based on patterns observed in horses, ultrasound-guided infiltrations are used for diagnostic or therapeutic purposes. In this study, cervical joints were evaluated in 13 donkeys (12 females and 1 male) with mean weight of 133kg and ages between 4 and 16 years, and additionally 05 mules and 09 donkeys, with mean weight of 393kg and ages between 4 and 15 years, all of them were mixed breed. The ultrasound examinations were performed using a variable frequency linear transducer 3 to 11 Mhz. The scan protocol was performed starting on the dorsal aspect of each vertebra, followed by an assessment of the cranial region to the caudal portion of the vertebra, towards the joint processes, longitudinal and in the caudal direction of the joint. Variations of the angulation allowed the exact location of the joint processes and the corresponding references images. The results observed show that these ultrasound techniques can guide cervical intra-articular puncture in donkeys and mules, and it is possible used for therapeutic or diagnostic purposes, and it is suggested that more studies in this sense be performed.

Keywords: cervical vertebra, articular processes, ultrasound technique, equidae

RESUMO

Há poucas descrições na literatura cientifica veterinária de técnicas ultrassonográficas da região cervical de muares e jumentos. Com base nos padrões observados em cavalos, as infiltrações guiadas por ultrassom são usadas para fins diagnósticos ou terapêuticos. Foram avaliadas as articulações cervicais em 13 jumentos (12 fêmeas e um macho), com peso médio de 133kg e idades entre quatro e 16 anos, e ainda cinco mulas e nove burros, com peso médio de 393kg e idades entre quatro e 15 anos, todos sem raça definida. O exame ultrassonográfico utilizou transdutor multifrequencial de 3 a 11Mhz, inicialmente com varredura do aspecto dorsal de cada vértebra, seguindo-se avaliação da região cranial à porção caudal da vértebra, em direção aos processos articulares, longitudinalmente e em sentido caudal para localização da articulação. Variações na angulação do transdutor permitiram a localização exata dos processos articulares e da articulação correspondente, do espaço articular entre as vértebras cervicais C2-C3 ao espaço articular entre C6-C7, e as respectivas imagens de referência. Os resultados observados mostram que essas técnicas ultrassonográficas podem orientar a punção intra-articular cervical em burros e mulas e que é possível utilizá-la para fins terapêuticos ou diagnósticos. Nesse sentido, sugere-se que mais estudos nesse sentido sejam realizados.

Palavras-chave: vértebra cervical, processos articulares, técnicas ultrassonográficas, equídeos

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INTRODUCTION

Donkeys can suffer from pathological cervical conditions similar to those of other equidae; however, due to evolutionary anatomical and physiological differences, they can lead to incorrect diagnoses, inadequate treatment, unnecessary expenses and a negative result (Toribio, 2019). It can also be observed in relation to the mules.

Commonly experienced professionals can face difficulties in evaluating the images obtained in imaging examinations of the cervical region of donkeys and mules, since there is no scientific evidence that cervical ultrasound imaging of horses can be used as references to cervical scan of those equidae.

Not only the anatomical complexity involved in the laborious ultrasound diagnosis of this region in equidae, but also the results obtained in imaging examinations sometimes do not show subtle lesions, but capable of producing clinical presentations that are difficult to understand, which makes the study of cervical vertebrae essential by using different diagnostic imaging techniques that can assist in the clinical understanding of this region.

The ultrasound examination complements the radiographic examination and is the essential and primarily option of choice for diagnosis, as can be observed in studies carried out in (Gollob *et al.*, 2002), (Audigié *et al.*, 2004) and (Pease *et al.*, 2012) in the atlanto-occipital (AO) joint.

In Audigié *et al.*, 2004 described two ultrasoundguided procedures for puncture of the atlantooccipital subarachnoid space with collection of cerebrospinal fluid and injection of contrast medium to perform myelography to reduce complications associated with the technique performed blindly, which can be performed with standing horse.

Similarly, Pease *et al.* (2012), performed a similar technique for collecting cerebrospinal fluid (CSF), without complications and allowing blood contamination to be minimized. Although the technique of intra-articular puncture in the cervical region is more accurate and requires less experience from the professional who will perform it, it can have the same success as the

technique performed without using the ultrasound, blindly, depending only on the experience of those who are performing the procedure (Chope, 2008).

In Gollob *et al.*, 2002 used anatomical specimens and ultrasound as a complementary exam to diagnose diseases in the AO region. In this study, the authors conclude that research on live horses should confirm the usefulness of this diagnostic tool.

And so it was in 2017 when Mackenzie *et al.*, in which measurements in the AO joint were performed to establish a standard of values in healthy neonate PSI foals comparing them with foals diagnosed with neonatal maladjustment syndrome (NMS), with significant differences in some of the measures obtained by the ultrasound assessment of the AO space between the two groups, where the dimensions of the spinal cord appear to be reduced in foals with NMS.

Chope (2008) reported the usefulness of performing radiography and nuclear scintigraphy, if available, prior to ultrasound examination to assist in the location and interpretation of osteoarthritis in cervical vertebral joints in horses.

Also, as a diagnostic aid, ultrasound was used in the diagnosis of cervical vertebral osteomyelitis in a horse with a history of multiple cervical abscesses (Alonso *et al.*, 2019), with the identification of areas with accumulation of secretion, in addition to certifying that there were no foreign bodies related to the injuries.

Ultrasonographic examination of the cranial nuchal bursa of horses was used in (Abuja *et al.*, 2014) to compare anatomical aspects and images obtained by magnetic resonance imaging of this structure.

In Berg *et al.*, 2003 a group of researchers stated that there was no detailed ultrasound description of the cervical spine of normal horses (without clinical alteration) and therefore they carried out the study to describe the normal ultrasound aspects of the transversal and longitudinal images in the cervical region, mainly of the articular processes, cervical vertebrae, and paravertebral structures. To this end, they compared ultrasound images with macroscopic sections of bones obtained in transverse and longitudinal planes in frozen specimens and observed a good relationship of these aspects, with small variations that were possibly attributed to transducer angulation when performing measurements. (Berg *et al.*, 2003).

There was a great variation in the contour of the articular processes, both in the ultrasound images and in the anatomical specimens, presenting irregularities, which were attributed to the insertions of the epaxial muscles (Berg *et al.*, 2003).

Ultrasound-guided intra-articular puncture has the advantage of more easily locating and positioning the needle in the joint of interest, whether for diagnostic or therapeutic purposes in cases of degenerative joint disease or osteochondrosis of the cervical vertebrae joint processes. These applications were described, in 2004, with anatomical specimens (Johnson *et al.*, 2017; Mattoon *et al.*, 2004), with the recommendation of clinical evaluation of such procedure (Mattoon *et al.*, 2004).

This same group of researchers, also in 2003 (Nielsen *et al.*), assessed the accuracy of the ultrasound-guided intra-articular puncture technique in 8 specimens of euthanized horses due to other non-orthopedic causes, in the 5 joints between C2 and C7, where 72% of the injections performed were intra-articular, with a marked increase in this efficiency as the operators acquired experience, resulting in 92% from the fifth to the eighth.

In addition, 17% of intracapsular injections (between the fibrous portion and the intimate synovial region) and 12%, periarticular injections (1mm from the joint capsule) were observed. They concluded that this technique can be applied to the diagnosis, associating with the radiographic alterations eventually obtained, as well as to the treatment of diseases with thirst in the cervical joints from C2 to C7 (Nielsen; *et al.*, 2003).

However, they warn that such a study was performed on anatomical parts and its practical application involves the use of restraint of the horse with the use of sedatives, as well as the movement of patients during its performance will make it difficult to direct the needle due to displacement of the transducer and may even bring problems when the needle is inserted in this region (Nielsen *et al.*, 2003).

In the study by Johnson *et al.* (2017), different techniques were performed, with the combination of some variables: angulation of the transducer in relation to the axial axis of the cervical spine, positioning it at 90° to this axis, a position called dorsal (D) or in 45° angled, cranio-dorsal (CrD); use of 2 transducers (micro convex 10 Mhz and linear 13 Mhz), 2 examiners and 2 needles (18 and 20 Gauge).

After examining 14 cervical columns in the different combinations of the variables presented, when the micro convex transducer was used, the examination was faster and more accurate, however there was no significant difference in accuracy between the 2 transducers used (Johnson *et al.*, 2017).

Considering the real possibility of producing some nerve damage, the CrD approach can be advantageous, avoiding the dorsal and ventral branches of the cervical nerves. It must be considered that this study was performed on anatomical parts, however, the techniques described are performed on live horses and in standing, routinely, without complications (Johnson *et al.*, 2017).

The ultrasound-guided intra-articular cervical puncture technique is easy to learn, with high precision in D or CrD approaches, and with linear or micro convex transducers, which can be used with equipment commonly used in the field (Johnson *et al.*, 2017).

In 2014 (Aleman *et al.*), for the first time, an intra-articular injection guided by ultrasound in the atlantoaxial joint (AA) was performed, aiming to perform myelography for the diagnosis of AA fusion, with success in the procedure.

In view of the considerations presented, the present study provides reference and parameters capable of contributing to the ultrasound interpretations of the cervical spine of donkeys and mules.

Ultrasound interpretations of the cervical spine of donkeys and mules.

To characterize the articular processes of the cervical vertebrae and their respective articulations in donkeys and mules, establishing the ultrasound technique and the capture of reference images that can assist in the more objective and accurate evaluation and interpretation of the ultrasound images, enabling the development of reliable interpretations.

MATERIALS AND METHODS

This study was approved and carried out in accordance with the local Animal Ethics and Use Committee (protocol number: 0018/2020).

13 donkeys (12 females and 1 male) with mean weight of 133kg and ages between 4 and 16 years (average of 9.5 years), and additionally 14 mules (05 females and 09 males), with mean weight of 393kg and ages between 4 and 15 years (average of 9.7 years), all of them were mixed race. Before, during and after the study, they were provided with water *ad libitum*, balanced feed for horses prepared at the institution, offered in the proportion of 1% of the

live weight per day, remaining in horse grass paddocks.

Previously to the ultrasound exams described below, it was inspected that the animals did not show any signs of swelling in the cervical region or lameness or motor incoordination, through a physical examination (palpation and inspection of the horse in the standing position and in the walk and trot, on soft and hard surfaces).

They were performed in a specific restraint stalls for equines (Fig. 1), within which the animals remained in standing, contained only by halter, with the participation of three professionals: one holding the halter and containing the equine; another capturing the images by positioning the transducer in the cervical region using the MyLab70 - ESAOTE[®] (Genoa - Italy) ultrasound devices on donkeys and MyLab30 - ESAOTE[®] (Genoa - Italy) on mules, and a 3 to 11 MHz variable frequency linear transducer 3 to 11 MHz. A third professional providing support and assisting in the handling, containment, and handling of equipment.



Figure 1. Mule positioned on the restraint stalls to perform the cervical ultrasound examination.

The procedure was preceded by regional trichotomy (Fig. 2) to mitigate artifacts and application of 70% isopropyl alcohol to remove dirt and oil. Ultrasonic gel was also used to minimize the artifacts produced by the contact of the transducer with the skin.

The ultrasound examination of each animal started with a scan of the dorsal aspect of each vertebra, first in C2, followed by an assessment of the cranial region to the caudal portion of the vertebra, towards the joint processes, longitudinally and in the caudal direction to locate the joint C2-3.

The scan was started with the transducer in a transverse plane (90° with the longitudinal axis of the neck), with the reference setting in the dorsal position (D), moving it in the cranial-dorsal direction (CrD), reaching to position it in

the longitudinal plane of the cervical spine (Fig. 3).

Thus, by scanning with variations in the transducer angulation in relation to the longitudinal plane of the neck, the exact location of the articular process of each joint was possible, since there was variation in the positioning of the transducer depending on the joint under study.

During the ultrasound examination, joint spaces C2-3 to C6-7 were shown and as the thickness of the neck increased in the caudal direction the lower frequency and an increase in the depth was observed. The scan started in C2-3 through the joints successively to C6-7 and always on the left side of the neck of the patients for standardization of the procedure.



Figure 2. Region of interest: from the caudal end of the transverse process of the atlas, two straight lines are projected. One in a ventral direction reaching the dorsal region to the jugular and dorsally to the base of the neck. The other, dorsally, extending towards the middle third of the neck until the base. A third line joins the two lines described projecting along the base of the neck.



Figure 3. Position of the transducer along the neck. (A) starting the scan in the C2 region until C2-3 joint identification. (B) following the regions of the joints C3-4, C4-5 (C) and C6-7 (D).

RESULTS

From the ultrasound images obtained in the region of the articular processes of the joints between the second and third cervical vertebrae (C2-3), between the third and fourth cervical

vertebrae (C3-4), between the fourth and fifth cervical vertebrae (C4-5), between the fifth and sixth cervical vertebrae (C5-6) and between the sixth and seventh cervical vertebrae (C6-7), one of each joint was selected on donkeys and mules, and are shown in Fig. 4 to 7.

Ultrasonography of the...



Figure 4. Cervical joints in MULES. A, between the second and third vertebrae (C2-3). B, between the third and fourth vertebrae (C3-4). C; between the fourth and fifth vertebrae (C4-5). D, between the fifth and sixth vertebrae (C5-6). E, between the sixth and seventh cervical vertebrae (C6-7). P2 = articular process of the second cervical vertebra (C2); P3 = articular process of the third cervical vertebra (C3); P4 = articular process of the fourth cervical vertebra (C4); P5 = articular process of the fifth cervical vertebra (C5); P6 = articular process of the sixth cervical vertebra (C6); P7 = articular process of the seventh cervical vertebra (C7); mm = epaxial musculature. Technique: Esaote MyLab 30, variable frequency linear transducer 3 to 11 Mhz.



Figure 5. Cervical joints (CJ) with delimitation of articular processes in MULES. A, between the second and third vertebrae (C2-3). B, between the third and fourth vertebrae (C3-4). C; between the fourth and fifth vertebrae (C4-5). D, between the fifth and sixth vertebrae (C5-6). E, between the sixth and seventh cervical vertebrae (C6-7). P2 = articular process of the second cervical vertebra (C2); P3 = articular process of the third cervical vertebra (C3); P4 = articular process of the fourth cervical vertebra (C4); P5 = articular process of the fifth cervical vertebra (C5); P6 = articular process of the sixth cervical vertebra (C6); P7 = articular process of the seventh cervical vertebra (C6); P7 = articular process of the seventh cervical vertebra (C7); mm = epaxial musculature. Technique: Esaote MyLab 30, variable frequency linear transducer 3 to 11 Mhz.



Figure 6. Cervical joints (CJ) in DONKEYS. A, between the second and third vertebrae (C2-3). B, between the third and fourth vertebrae (C3-4). C; between the fourth and fifth vertebrae (C4-5). D, between the fifth and sixth vertebrae (C5-6). E, between the sixth and seventh cervical vertebrae (C6-7). P2 = articular process of the second cervical vertebra (C2); P3 = articular process of the third cervical vertebra (C3); P4 = articular process of the fourth cervical vertebra (C4); P5 = articular process of the fifth cervical vertebra (C5); P6 = articular process of the sixth cervical vertebra (C6); P7 = articular.



Figure 7. Cervical joints (CJ) with delimitation of articular processes in DONKEYS. A, between the second and third vertebrae (C2-3). B, between the third and fourth vertebrae (C3-4). C; between the fourth and fifth vertebrae (C4-5). D, between the fifth and sixth vertebrae (C5-6). E, between the sixth and seventh cervical vertebrae (C6-7). P2 = articular process of the second cervical vertebra (C2); P3 = articular process of the third cervical vertebra (C3); P4 = articular process of the fourth cervical vertebra (C4); P5 = articular process of the fifth cervical vertebra (C5); P6 = articular process of the sixth cervical vertebra (C6); P7 = articular process of the second cervical vertebra (C4); P5 = articular process of the second cervical vertebra (C5); P6 = articular process of the sixth cervical vertebra (C6); P7 = articular process of the second cervical vertebra (C7); mm = epaxial musculature. Technique: Esaote MyLab 70, variable frequency linear transducer 3 to 11 Mhz.

DISCUSSION

There is no detailed anatomical description of the cervical vertebrae of donkeys and mules in the consulted literature and the anatomical references are based on the horse anatomy, using such descriptions (Budras *et al.*, 2011; Clayton *et al.*, 2005; Getty, 1986; Green, 2006) as a reference to the present study.

Regarding this literary absence, it is important to acquire data for specific ultrasound technique for the examination of cervical joints and joint processes in donkeys and mules, considering the established applicability in horses (Abuja *et al.*, 2014; Aleman *et al.*, 2014; Alonso *et al.*, 2019; Audigié *et al.*, 2004; Chope, 2008; Gollob *et al.*, 2002; Johnson *et al.*, 2017; Mackenzie *et al.*, 2017; Mattoon *et al.*, 2004; Pease *et al.*, 2012) in the diagnosis of diseases and as an aid to ultrasound-guided intra-articular punctures in these joints, whether for diagnosis or therapy.

However, the present study describes this technique in detail, including anatomical references and angles of the transducer in relation to the longitudinal axis of the neck depending on the joint of interest. The techniques described in horses do not present these anatomical references, nor do they indicate the best positioning of the transducer for the location and characterization of each cervical joint between C2 and C7 (Berg *et al.*, 2003; Mattoon *et al.*, 2004).

As for the technique developed in horses, there was the use of variations in the transducer angulation, from dorsal (90° in relation to the longitudinal plane of the cervical spine), at 45°, a position called Cr-D, however, they do not describe which angulation is more adequate to each cervical joint, as well as performed the examination on anatomical parts and also used ultrasound to guide the insertion of a needle aiming at intra-articular injection.

This fact distinguishes it from the technique described in this research, in which the procedure was performed in live animals, describing that in C2-3 angulations close to 90° are more desirable for the visualization of joint processes, with a tendency positioning of the transducer in horizontal plane in relation to the longitudinal cervical plane in the subsequent joints. Initially,

in C2-3, this positioning of the transducer was between 60 to 90° to the longitudinal axis of the neck, that is, in the dorsal position. In C3-4, the transducer was positioned with the reference indicator to CrD, ranging from 45 to 60°, being close to 45° in the C4-5 and C5-6 joints, and less than 45°, tending to Cr, in the C6-7 joint.

The technique and angulations performed for the cervical spine ultrasound images acquisition were the same for both donkeys and mules, being similar to those described in horses (Aleman *et al.*, 2014; Berg *et al.*, 2003; Johnson *et al.*, 2017; Mattoon *et al.*, 2004; Nielsen *et al.*, 2003), but with the need to vary the angles of the transducer depending on the joint.

Due to the differences in the cervical musculature the lower frequency was required as the thickness of the neck increases in the caudal direction.

Berg *et al.* (2003) reported an "ex vivo" anatomical study in which the technic for imaging acquisition was described positioning the transducer in a transversal plane to the vertebral bodies. Considering that the aim of our "in vivo" study was the joints formed by cervical joint processes, the transducer angulation was performed.

The variations observed in the bone margins of the articular processes of cervical vertebrae in horses (Berg *et al.*, 2003) were not observed in donkeys and mules.

CONCLUSION

The data collected reinforce the ultrasound technique of the cervical spine in donkeys and mules, serving as a reference for this exam and assisting in the punctures of joints C2-3 to C6-7. Thus, studies using the described ultrasound technique could assist an ultrasound guided intraarticular puncture in donkeys and mules for therapeutic or diagnostic purposes.

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REFERENCES

ABUJA, G.A.; GARCÍA-LÓPEZ, J.M.; MANSO-DÍAZ, G. *et al.* The cranial nuchal bursa: anatomy, ultrasonography, magnetic resonance imaging and endoscopic approach. *Equine Vet. J.*, v.46, p.745-750, 2014.

ALEMAN, M.; DIMOCK, A.N.; WISNER, E.R. *et al.* Atlanto-axial approach for cervical myelography in a thoroughbred horse with complete fusion of the atlanto-occipital bones. *Can. Vet. J.*, v.55, p.1069-1073, 2014.

ALONSO, J.M.; ÁVILA, A.R.; BUENO, L.M.C. *et al.* Cervical vertebral osteomyelitis secondary to Streptococcus equi infection in an adult horse - case report. *Arq. Bras. Med. Vet. Zootec.*, v.71, p.2041-2048, 2019.

AUDIGIÉ, F.; TAPPREST, J.; DIDIERLAURENT, D.; DENOIX, J.M. Ultrasound-guided atlanto-occipital puncture for myelography in the horse. *Vet. Radiol. Ultrasound*, v.45, p.340-344, 2004.

BERG, L.; NIELSEN. J.; THOEFNER, M.; THOMSEN, P. Ultrasonography of the equine cervical region: a descriptive study in eight horses. *Equine Vet. J.*, v.35, p.647-655, 2003.

BUDRAS, K.D.; SACK, W.O.; SABINE, R. Axial skeleton and neck. In: *Anatomy of the horse*. 6.ed. Hannover: Schlütersche Verlagsgesellschaft mbH & Co., 2011. p.56-59.

CHOPE, K. How to perform sonographic examination and ultrasound-guided injection of the cervical vertebral facet joints in horses. *Am. Assoc. Equine Pract.*, v.54, p.186-189, 2008.

CLAYTON, H.M.; FLOOD, P.F.; ROSENSTEIN, D. *Clinical anatomy of the horse*. London: Mosby Elsevier, 2005.

GETTY, R. Sisson & Grossman: anatomia dos animais domésticos. 5.ed. Rio de Janeiro: Guanabara, 1986.

GOLLOB, E.; EDINGER, H.; STANEK, C.; WURNIG, C. Ultrasonographic investigation of the atlanto-occipital articulation in the horse. *Equine Vet. J.*, v.34, p.44-50, 2002.

GREEN, J. *Horse anatomy*. Mineola, New York: Dover Publications, Inc., 2006.

JOHNSON, J.P.; PILHA, J.D.; ROWAN, C.; HANDEL, I.; O'LEARY, J.M. Ultrasoundguided approach to the cervical articular process joints in horses: A validation of the technique in cadavers. *Vet. Comp. Orthop. Traumatol.*, v.30, p.165-171, 2017.

MACKENZIE, C.J.; HAGGETT, E.F.; PINCHBECK, G.L.; MARR, C.M. Ultrasonographic assessment of the atlantooccipital space in healthy Thoroughbred foals and Thoroughbred foals with neonatal maladjustment syndrome. *Vet. J.*, v.223, p.55-59, 2017.

MATTOON, J.S.; DROST, O.T.; GRGURIC, M.R. *et al.* Technique for equine cervical articular process joint injection. *Vet. Radiol. Ultrasound*, v.45, p.238-240, 2004.

NIELSEN, J.V.; BERG, L.C.; THOEFNER, M.B.; THOMSEN, P.D. Accuracy of ultrasound-guided intra-articular injection of cervical facet joints in horses: a cadaveric study. *Equine Vet. J.*, v.35, p.657-661, 2003.

PEASE, A.; BEHAN, A.; BOHART, G. Ultrasound-guided cervical centesis to obtain cerebrospinal fluid in the standing horse. *Vet. Radiol. Ultrasound*, v.53, p.92-95, 2012.

TORIBIO, R.E. Dear donkey and mule: you deserve more appreciation and better medicine. *Vet. Clin. North Am. Equine Pract.*, v.35, n.3, 2019.