












## Study of digital palmar and plantar arteries of horses and mules by B-mode and Doppler ultrasonography

[*Estudo das artérias digitais palmares e plantares de equinos e muares por ultrassonografia modo-B e Doppler*]

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### ABSTRACT

The present study aimed to evaluate and compare the palmar and plantar digital arteries of 10 horses and 10 mules through B-mode and spectral Doppler ultrasound. Likewise, compare the plantar arteries of the pelvic limbs with the palmar arteries of the thoracic limbs, in addition to verifying the differences of the digital arteries in the contralateral limbs (right and left) in horses and mules using B-mode and spectral Doppler ultrasonography. The evaluated parameters were the diameter and the intima-media thickness (IMT), resistivity index (RI), pulsatility index (PI), peak of systolic velocity (pSV), final diastolic velocity (fDV) and mean velocity (MV) of the lateral and medial palmar and plantar arteries digital in healthy horses and mules. The horses presented higher diameters values, IMT, pSV, fDV and MV in comparison to the mules. The variables RI and PI also presented differences between horses and mules. Also, both species showed higher values in the variables of B-mode and Doppler ultrasonography in the thoracic limbs. Differences were also detected in the comparison between the limb sides (right and left) in the two modalities. The B-Mode and spectral Doppler ultrasound techniques are viable tools to identify differences in the palmar and plantar digital arteries in healthy horses and mules, highlight the differences between the thoracic and pelvic limbs and their respective sides (right and left) as well.

Keywords: locomotor limbs, equines, ultrasound, Doppler, and B-mode

### RESUMO

*O presente estudo teve como objetivo avaliar e comparar as artérias digitais palmares e plantares de 10 equinos e 10 muares, por meio de ultrassom modo B e Doppler espectral. Da mesma forma, buscou-se comparar as artérias plantares dos membros pélvicos com as artérias palmares dos membros torácicos, além de se verificarem as diferenças das artérias digitais nos membros contralaterais (direito e esquerdo) em equinos e mulas, por meio de ultrassonografia modo B e Doppler espectral. Os parâmetros avaliados foram o diâmetro e a espessura médio-intimal (EMI), o índice de resistividade (IR), o índice de pulsatilidade (IP), o pico de velocidade sistólica (pVS), a velocidade diastólica final (VDf) e a velocidade média (VM) das artérias palmares e plantares lateral e medial digitais em equinos e muares sadios. Os equinos apresentaram maiores valores de diâmetros, EMI, pVS, VDf e VM em comparação aos muares. As variáveis IR e IP também apresentaram diferenças entre equinos e muares. Cavalos e mulas apresentaram maiores valores nas variáveis de ultrassonografia modo B e Doppler nos membros torácicos. Também foram detectadas diferenças na comparação entre os lados dos membros (direito e esquerdo) nas duas modalidades. As técnicas de ultrassom modo-B e Doppler espectral são ferramentas viáveis para identificar diferenças nas artérias digitais palmares e plantares em equinos e muares saudáveis, bem como para destacar as diferenças entre os membros torácicos e pélvicos e seus respectivos lados (direito e esquerdo).*

*Palavras-chave: membros locomotores, equinos, ultrassom, Doppler e modo B*

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Submitted: May 2, 2022. Accepted: February 28, 2023.

## INTRODUCTION

The lateral and medial digital palmar arteries originate from the bifurcation of the medial artery and cross abaxially to the proximal sesamoid bones (fetlock region). The lateral and medial digital plantar arteries originate from the bifurcation of the metatarsal artery III and cross abaxially to the proximal sesamoid bones (fetlock region) (Burg *et al.*, 2007; Parks, 2017; Fails, 2020).

Many disorders in horses result in changes in peripheral blood flow (Cochar *et al.*, 2000), and the Doppler ultrasonography is a tool coupled with conventional ultrasonography (B-Mode) that allows vascular and hemodynamic evaluation of the vessels to be studied, enabling the diagnosis of diseases (Yanik, 2002; Souza *et al.*, 2022). In the case of vascular alterations of digital arteries in healthy horses or with inflammatory diseases, such as laminitis and septic pododermatitis, may be detected with the aid of Mode-B and Doppler ultrasonography (Wongaumnaykul *et al.*, 2006; Viera *et al.*, 2016).

B-Mode ultrasonography allows the assessment of the vascular architecture (intima-media thickness (IMT)) while Doppler ultrasonography provides real-time quantitative and qualitative information on the vascular patterns, allowing an assessment of the presence or absence of flow, direction, velocity, and type of blood flow, and hemodynamic patterns (Douthit, *et al.*, 2014; Viera *et al.*, 2016; Miele, 2017).

Mules are hybrid animals resulting from the cross between horses and donkeys. The mules have aroused economic interest and need subsidies for scientific work to contribute to the basic knowledge to elucidate diseases, especially those of the locomotor system. Those animals are used essentially for work on farms or in gait competition, and they may not be treated as equines, since they result from the cross between equines and donkeys (Anderson, 1939; Camac, 1997; Araújo, 2010; Salles *et al.*, 2013).

Consequently, the welfare of these animals has increased in recent years and numerous differences have been reported. Such studies were aimed at exposing possible differences and avoiding clinical, diagnostic and treatment errors

(Salles *et al.*, 2013; Miranda and Palhares, 2017; Mendoza *et al.*, 2018).

The horses' thoracic limbs receive 60% of the animal's body weight resulting in a higher rate of illnesses when compared to the pelvic limbs (Getty, 1986; Latorre *et al.*, 2001). For this reason, studies comparing the pelvic and thoracic limbs are required, also the improvement of the knowledge related to the hemodynamics of the blood flow of the and plantar and palmar digital arteries.

In humans, there may be differences in vessels when compared between the right and left sides (Sayer and Fatherrree, 1945), and in horses this variation may also exist Cochar *et al.*, 2000). Studies comparing the hemodynamics of blood flow in the palmar and plantar digital arteries of equines and mules through B-mode ultrasonography and spectral Doppler were not found in the literature consulted.

In the face of the lack of information about this subject and considering the economic and social importance of these animals and the growing demand for scientific data, the present study aimed to evaluate and compare the palmar and plantar digital arteries of 10 horses and 10 mules through B-mode and spectral Doppler ultrasound. Likewise, compare the plantar arteries of the pelvic limbs with the palmar arteries of the thoracic limbs, in addition to verifying the differences of the digital arteries in the contralateral limbs (right and left) in horses and mules using B-mode and spectral Doppler ultrasonography.

## MATERIALS AND METHODS

This research was approved by the Ethics Committee on the Use of Animals (CEUA) of the School of Veterinary Medicine and Animal Science (FMVZ/UNESP) (Protocol No. 0257/2018).

B-Mode ultrasonography and spectral Doppler were performed on the lateral and medial digital palmar and plantar arteries of 10 healthy horses (5 males and 5 females), with body weight between 336 and 390 kg and ages between 2 and 18 years. The examinations were also performed in the same anatomical regions in 10 mules (5 males and 5 females), with body weight between

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347 and 439 kilograms (kg) and age between 4 and 15 years.

The criteria for selecting the animals were based on normal laboratory (blood cell count,  $\gamma$ -glutamyl transferase, aspartate aminotransferase, alkaline phosphatase, urea, and creatinine) and physical examinations (heart rate, respiratory rate, gastrointestinal motility, and rectal temperature). The animals healthy were submitted to trichotomy in the fetlock regions.

The animals were not subjected to water or solid fasting and did not receive sedatives to avoid hemodynamic changes. For B-mode and spectral Doppler ultrasonographic examinations, the

animals were kept in an examination stall guided by halters and remained in stance limiting the movement of the locomotor limbs, with the body weight equally distributed between the limbs and later. Isopropyl alcohol in the concentration 70% was applied to the fetlock region for image acquisition.

The GE Healthcare Logiq V2 mobile ultrasound device with a high frequency linear transducer (8 to 12 MHz) was used to obtain B-mode images in the transverse and longitudinal plans (Fig. 1) to evaluate the diameters and IMT of the lateral and medial palmar and plantar digital artery walls.



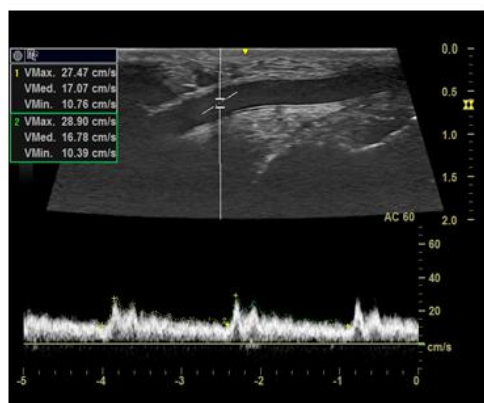
Figure 1. Position of the transducer to obtain images in the planes: transverse (A - horse) and longitudinal (C - mule). B-mode ultrasound image of the digital artery in the planes: transverse (B) and longitudinal (D).

For the analysis of blood flow (spectral Doppler), insonation angles inferior to 60 degrees (60°) were used to acquire the velocity as reliably as possible. The cursor for obtaining spectral waves was positioned in the center of the lateral and medial palmar and plantar digital arteries to minimize possible changes in blood flow velocity, as recommended by Menzies-Gow and Marr (2007).

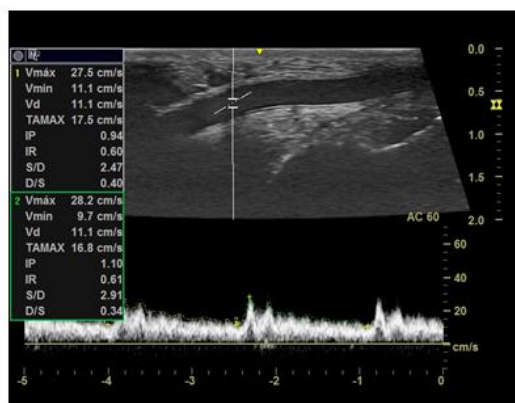
Through the spectral waves (spectral Doppler) the following variables were calculated: resistivity index (RI), pulsatility index (PI), peak systolic velocity (pSV), final diastolic velocity (fDV) and mean velocity (MV) of the lateral and medial palmar and plantar digital arteries (Fig. 2).



A



B



C

Figure 2. Position of the transducer in the longitudinal plane to perform spectral Doppler ultrasonography (A - horse). Post-processing of images with the contour of the waves to obtain the values of RI and PI (B), and pSV, fDV and MV (C).

Descriptive statistics were obtained for each group and all data were submitted to the normality test. For the statistical analysis, the Mann-Whitney test was applied to compare the two samples. Also, the Mann-Whitney test was applied to compare the measurements obtained in the pelvic and thoracic limbs, and between the contralateral limbs (right and left) in horses and mules using the SPSS V20 software.

All hypothesis tests developed in this work considered a 5% significance, with the null hypothesis being rejected when the p-value was inferior or equal to 0.05. Therefore, H0: the distributions of the two samples are equal or rejected, and H1: concludes that the two samples have significantly different distributions.

**RESULTS**

The lateral palmar digital artery of the right thoracic limb (RTL) showed no significant difference between horses and mules. However, the medial palmar digital artery of the RTL limb presented a difference in the longitudinal wall (p = 0.045), with horses presenting the highest mean (Table 1).

The lateral palmar digital artery of the left thoracic limb (LTL) presented a significant difference in the transverse diameter (p = 0.046), with horses presenting higher values than mules. There was a significant difference in the medial palmar digital artery of the LTL in the longitudinal diameter (p = 0.045), with the horses presenting higher values in comparison to mules (Tab. 1).

Table 1. Mean, median, standard deviation, Q1, Q3 and confidence interval of the variables in B-mode ultrasonography of the lateral and medial digital arteries of the thoracic limbs, for comparison between horses and mules

Thoracic limb	Face	B-Mode	Animals	Mean	Median	Standard deviation	Q1	Q3	P-value
Right	Lateral	Longitudinal Diameter	Horses	0.527	0.540	0.067	0.450	0.580	0.969
			Mules	0.538	0.540	0.090	0.490	0.580	
		Transverse Diameter	Horses	0.532	0.550	0.049	0.500	0.560	0.190
			Mules	0.520	0.530	0.083	0.460	0.560	
	Longitudinal Wall	Horses	0.113	0.110	0.025	0.100	0.130	0.168	
		Mules	0.103	0.100	0.027	0.080	0.120		
	Transverse Wall	Horses	0.116	0.120	0.028	0.093	0.138	0.380	
		Mules	0.110	0.110	0.032	0.080	0.130		
	Medial	Longitudinal Diameter	Horses	0.545	0.535	0.055	0.503	0.565	0.268
			Mules	0.490	0.520	0.129	0.500	0.530	
		Transverse Diameter	Horses	0.574	0.595	0.079	0.493	0.610	0.299
			Mules	0.550	0.530	0.083	0.500	0.600	
	Longitudinal Wall	Horses	0.119	0.115	0.026	0.100	0.138	<b>0.045*</b>	
		Mules	0.098	0.100	0.034	0.088	0.130		
Transverse Wall	Horses	0.114	0.110	0.030	0.093	0.138	0.585		
	Mules	0.109	0.100	0.031	0.090	0.130			
Left	Lateral	Longitudinal Diameter	Horses	0.541	0.520	0.073	0.510	0.555	0.052
			Mules	0.552	0.570	0.052	0.510	0.580	
		Transverse Diameter	Horses	0.610	0.600	0.093	0.545	0.640	<b>0.046*</b>
			Mules	0.531	0.540	0.183	0.500	0.600	
	Longitudinal Wall	Horses	0.120	0.120	0.032	0.110	0.130	0.869	
		Mules	0.119	0.120	0.026	0.100	0.140		
	Transverse Wall	Horses	0.142	0.140	0.037	0.110	0.150	0.257	
		Mules	0.126	0.130	0.055	0.090	0.160		
	Medial	Longitudinal Diameter	Horses	0.578	0.570	0.060	0.530	0.610	<b>0.045*</b>
			Mules	0.550	0.540	0.060	0.490	0.560	
		Transverse Diameter	Horses	0.545	0.570	0.161	0.520	0.650	0.656
			Mules	0.567	0.590	0.150	0.510	0.650	
	Longitudinal Wall	Horses	0.120	0.120	0.025	0.100	0.140	0.264	
		Mules	0.114	0.100	0.024	0.100	0.130		
Transverse Wall	Horses	0.128	0.130	0.049	0.100	0.150	0.477		
	Mules	0.141	0.130	0.052	0.100	0.185			

\* = significant values (in bold)

Table 2 shows a significant difference in the variables longitudinal (p <0.0001) and transverse (p = 0.004) diameter, and in longitudinal (p <0.0001) wall in the lateral plantar digital artery of the pelvic limb (RPL) with the mules showing higher values than horses. However, no significant differences were observed in the medial plantar digital artery of the same limb (RPL).

The lateral digital plantar artery of the left pelvic limb (LPL) presented a significant difference in the longitudinal diameter (p = 0.01), horses with higher means than mules. The medial digital plantar artery of the LPL presented a significant difference in the longitudinal (p = 0.006) and transverse (p = 0.043) variables, with the longitudinal diameter higher in horses while the transversal diameter was higher in the mules (Table 2).

Table 2. Mean, median, standard deviation, Q1 and Q3 of the variables in B-Mode ultrasonography of the lateral and medial digital arteries of the pelvic limbs, for comparison between horses and mules

Pelvic limb	Face	B-Mode	Animals	Mean	Median	Standard deviation	Q1	Q3	P-value
Right	Lateral	Longitudinal Diameter	Horses	0.445	0.455	0.076	0.395	0.490	<0.001*
			Mules	0.552	0.550	0.106	0.490	0.630	
		Transverse Diameter	Horses	0.410	0.400	0.066	0.345	0.460	0.004*
			Mules	0.498	0.480	0.114	0.400	0.590	
		Longitudinal Wall	Horses	0.084	0.080	0.023	0.070	0.100	0.001*
			Mules	0.112	0.110	0.032	0.095	0.130	
		Transverse Wall	Horses	0.083	0.090	0.024	0.070	0.100	0.052
			Mules	0.101	0.100	0.036	0.070	0.120	
	Medial	Longitudinal Diameter	Horses	0.495	0.500	0.083	0.435	0.535	0.337
			Mules	0.458	0.440	0.118	0.390	0.550	
		Transverse Diameter	Horses	0.411	0.410	0.097	0.320	0.440	0.148
			Mules	0.450	0.430	0.126	0.330	0.590	
		Longitudinal Wall	Horses	0.095	0.090	0.025	0.070	0.115	0.148
			Mules	0.084	0.080	0.026	0.070	0.100	
		Transverse Wall	Horses	0.085	0.090	0.026	0.065	0.105	0.225
			Mules	0.094	0.090	0.026	0.080	0.110	
Left	Lateral	Longitudinal Diameter	Horses	0.428	0.430	0.050	0.395	0.450	0.01*
			Mules	0.384	0.360	0.093	0.320	0.440	
		Transverse Diameter	Horses	0.411	0.390	0.061	0.370	0.455	0.383
			Mules	0.402	0.370	0.066	0.350	0.460	
		Longitudinal Wall	Horses	0.090	0.090	0.032	0.068	0.103	0.907
			Mules	0.088	0.080	0.023	0.070	0.100	
		Transverse Wall	Horses	0.083	0.075	0.027	0.068	0.100	0.93
			Mules	0.084	0.080	0.029	0.060	0.100	
	Medial	Longitudinal Diameter	Horses	0.444	0.460	0.063	0.408	0.490	0.006*
			Mules	0.401	0.400	0.076	0.350	0.450	
		Transverse Diameter	Horses	0.367	0.355	0.136	0.300	0.475	0.043*
			Mules	0.419	0.420	0.058	0.380	0.440	
		Longitudinal Wall	Horses	0.094	0.090	0.021	0.080	0.110	0.339
			Mules	0.089	0.090	0.022	0.070	0.100	
		Transverse Wall	Horses	0.076	0.075	0.035	0.050	0.100	0.082
			Mules	0.091	0.090	0.028	0.070	0.100	

\* = significant values (in bold)

Table 3 shows a significant difference between horses and mules in the lateral palmar digital artery of the right thoracic limb (RTL) in the variables pSV ( $p < 0.001$ ), fDV ( $p < 0.001$ ) and MV ( $p < 0.001$ ), with horses showing the highest values. The medial palmar digital artery of the RTL limb presented a significant difference in the variables RI ( $p < 0.001$ ), PI ( $p < 0.001$ ), pSV ( $p < 0.001$ ), fDV ( $p < 0.001$ ) and MV ( $p < 0.001$ ), with higher values observed in horses in comparison with mules.

The lateral palmar digital artery of the left thoracic limb (LTL) presented a significant difference in the variables RI ( $p = 0.003$ ) and PI ( $p = 0.029$ ), mules with the highest means. However, for the variables pSV ( $p < 0.001$ ), fDV ( $p < 0.001$ ) and MV ( $p < 0.001$ ), horses presented

higher values than mules. In the medial palmar digital artery of the LTL, there was a significant difference in the variables pSV ( $p = 0.005$ ) and MV ( $p = 0.038$ ), with the highest values observed in horses.

Table 4 shows a significant difference in the lateral digital plantar artery of the right pelvic limb (RPL) in the variables pSV ( $p < 0.001$ ), fDV ( $p = 0.005$ ) and MV ( $p < 0.001$ ), with horses showing the highest values. In the RPL medial plantar digital artery there was a difference in the variables PI ( $p = 0.004$ ), pSV ( $p < 0.001$ ), fDV ( $p = 0.006$ ) and MV ( $p = 0.002$ ), with higher values observed in horses than mules.

The lateral and medial digital plantar artery of the left pelvic limb (LPL) presented a significant

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difference in the variables pSV (p <0.001), fDV (p <0.001) and MV (p <0.001), with horses showing the highest mean.

Performing the B-mode ultrasonography in horses (Table 5) there were significant differences between the pelvic and thoracic limbs in the lateral digital artery in the longitudinal (p<0.001) and transverse (p<0.001) diameters, longitudinal (p<0.001) and transversal (p<0.001)

walls, whereby the thoracic limbs showed the highest values in the right and left limbs.

In the medial digital artery, there was a significant difference in the variables longitudinal (p<0.001) and transverse (p<0.001) diameters, longitudinal (p= 0.006; p<0.001) and transverse (p= 0.004; p<0.001) walls, with the thoracic limbs showing the highest means in both limbs.

Table 3. Mean, median, standard deviation, Q1 and Q3 of the variables of spectral Doppler ultrasonography of the lateral and medial digital arteries of the thoracic limbs, for comparison between horses and mules

Thoracic limb	Face	Doppler	Animals	Mean	Median	Standard deviation	Q1	Q3	P-value
Right	Lateral	RI	Horses	0.564	0.570	0.053	0.525	0.600	0.213
			Mules	0.576	0.580	0.077	0.510	0.640	
		PI	Horses	0.986	0.935	0.189	0.870	1.130	0.329
			Mules	0.942	0.950	0.210	0.770	1.110	
		pSV	Horses	30.98	26.96	11.95	22.62	34.54	<0.001*
			Mules	19.76	20.11	4.02	16.66	23.24	
	fDV	Horses	12.15	10.16	6.25	9.20	12.10	<0.001*	
		Mules	8.40	8.37	2.03	6.96	9.61		
	Medial	MV	Horses	19.38	16.18	8.76	14.77	20.55	<0.001*
			Mules	12.63	12.88	2.60	10.14	14.47	
		RI	Horses	0.597	0.620	0.072	0.570	0.650	<0.001*
			Mules	0.565	0.565	0.071	0.523	0.610	
		PI	Horses	1.037	1.090	0.174	0.965	1.155	<0.001*
			Mules	0.894	0.880	0.179	0.770	0.985	
	pSV	Horses	30.05	31.65	12.61	16.23	39.80	<0.001*	
		Mules	19.22	18.11	6.41	13.39	22.02		
	fDV	Horses	12.57	12.39	7.21	5.76	15.10	<0.001*	
		Mules	8.16	7.72	2.24	6.68	8.98		
MV	Horses	18.07	20.01	7.44	9.21	23.27	<0.001*		
	Mules	12.54	12.13	3.41	9.46	15.85			
Left	Lateral	RI	Horses	0.551	0.565	0.095	0.453	0.648	0.003*
			Mules	0.614	0.600	0.092	0.555	0.670	
		PI	Horses	0.941	0.915	0.218	0.735	1.125	0.029*
			Mules	1.077	0.980	0.328	0.830	1.200	
		pSV	Horses	34.12	36.47	10.98	22.25	40.89	<0.001*
			Mules	15.20	16.35	3.75	11.82	18.48	
	fDV	Horses	14.19	12.95	5.81	8.34	19.69	<0.001*	
		Mules	5.80	5.23	2.31	3.57	8.07		
	Medial	MV	Horses	21.59	22.44	7.29	15.82	26.82	<0.001*
			Mules	9.18	8.14	2.80	6.83	11.89	
		RI	Horses	0.593	0.600	0.087	0.520	0.660	0.563
			Mules	0.606	0.590	0.097	0.545	0.680	
		PI	Horses	1.082	1.100	0.260	0.920	1.270	0.542
			Mules	1.100	0.980	0.375	0.840	1.320	
	pSV	Horses	23.26	26.87	12.21	9.29	31.03	0.005*	
		Mules	17.21	16.03	5.09	13.19	22.48		
	fDV	Horses	9.25	8.55	6.37	3.24	11.82	0.099	
		Mules	6.52	5.65	2.66	4.38	9.27		
MV	Horses	14.65	14.20	8.86	5.63	18.46	0.038*		
	Mules	10.52	9.30	3.71	7.43	13.51			

RI = resistivity index; PI = pulsatility index; pSV = peak systolic velocity; fDV = final diastolic velocity; MV = mean velocity; \* = significant values (in bold)

Table 4. Mean, median, standard deviation, Q1 and Q3 of the variables of the spectral Doppler ultrasonography of the lateral and medial digital arteries of the pelvic limbs, for comparison between horses and mules

Pelvic limb	Face	Doppler	Animals	Mean	Median	Standard deviation	Q1	Q3	P-value
Right	Lateral	RI	Horses	0.613	0.650	0.082	0.540	0.680	0.834
			Mules	0.608	0.600	0.075	0.553	0.680	
		PI	Horses	1.165	1.100	0.312	0.890	1.410	0.308
			Mules	1.098	1.045	0.258	0.890	1.338	
		pSV	Horses	29.15	23.35	14.90	15.35	44.32	<0.001*
			Mules	16.22	16.35	4.13	13.77	17.53	
	fDV	Horses	10.92	8.55	6.80	4.89	16.71	0.005*	
		Mules	6.19	5.91	1.82	4.80	7.29		
	MV	Horses	16.87	13.92	9.52	8.32	27.12	<0.001*	
		Mules	9.19	8.51	2.74	7.72	10.67		
	Medial	RI	Horses	0.650	0.675	0.088	0.590	0.720	0.07
			Mules	0.626	0.600	0.078	0.560	0.680	
		PI	Horses	1.281	1.355	0.272	1.023	1.510	0.004*
			Mules	1.122	0.990	0.293	0.900	1.310	
		pSV	Horses	30.07	32.64	13.20	16.19	43.23	<0.001*
			Mules	18.53	18.61	4.84	15.04	21.93	
	fDV	Horses	10.04	9.18	5.78	4.45	13.56	0.006*	
		Mules	6.69	6.63	2.50	4.63	8.97		
MV	Horses	16.42	16.04	8.17	8.11	22.33	0.002*		
	Mules	10.91	11.14	3.27	8.21	13.61			
Left	Lateral	RI	Horses	0.626	0.640	0.074	0.570	0.680	0.434
			Mules	0.636	0.655	0.094	0.543	0.708	
		PI	Horses	1.163	1.205	0.326	0.980	1.260	0.438
			Mules	1.198	1.230	0.314	0.923	1.438	
		pSV	Horses	33.47	38.72	14.07	20.05	44.11	<0.001*
			Mules	15.78	15.41	4.89	11.39	20.92	
	fDV	Horses	12.45	12.72	6.99	6.55	14.66	<0.001*	
		Mules	5.61	5.24	2.55	3.17	7.81		
	MV	Horses	18.91	21.01	9.31	9.80	23.34	<0.001*	
		Mules	9.04	8.45	3.22	6.08	11.19		
	Medial	RI	Horses	0.620	0.610	0.075	0.560	0.670	0.369
			Mules	0.646	0.610	0.107	0.560	0.730	
		PI	Horses	1.150	1.090	0.286	0.970	1.210	0.904
			Mules	1.274	1.090	0.492	0.900	1.568	
		pSV	Horses	37.45	24.45	20.10	19.28	55.23	<0.001*
			Mules	17.89	18.09	3.55	14.88	21.15	
	fDV	Horses	13.26	9.61	6.76	7.94	19.64	<0.001*	
		Mules	6.19	6.15	2.24	4.58	8.22		
MV	Horses	21.31	14.58	11.24	11.97	31.96	<0.001*		
	Mules	10.06	9.14	2.44	8.62	12.18			

RI = resistivity index; PI = pulsatility index; pSV = peak systolic velocity; fDV = final diastolic velocity; MV = mean velocity; \* = significant values (in bold)



Table 5. Mean, median, standard deviation, Q1 and Q3 of the horses' limbs variables by B-mode ultrasonography, followed by the p-value to compare the digital arteries between the pelvic and thoracic limbs

Region	B-mode	Side	Limb	Mean	Median	Standard deviation	Q1	Q3	P-value
Lateral	Longitudinal diameter	Right	Pelvic	0.445	0.455	0.076	0.395	0.490	<0.001*
			Thoracic	0.527	0.540	0.067	0.450	0.580	
		Left	Pelvic	0.428	0.430	0.050	0.395	0.450	<0.001*
			Thoracic	0.541	0.520	0.073	0.510	0.555	
	Transversal diameter	Right	Pelvic	0.410	0.400	0.066	0.345	0.460	<0.001*
			Thoracic	0.532	0.550	0.049	0.500	0.560	
		Left	Pelvic	0.411	0.390	0.061	0.370	0.455	<0.001*
			Thoracic	0.610	0.600	0.093	0.545	0.640	
	Longitudinal wall	Right	Pelvic	0.084	0.080	0.023	0.070	0.100	<0.001*
			Thoracic	0.113	0.110	0.025	0.100	0.130	
		Left	Pelvic	0.090	0.090	0.032	0.068	0.103	<0.001*
			Thoracic	0.120	0.120	0.032	0.110	0.130	
Transversal wall	Right	Pelvic	0.083	0.090	0.024	0.070	0.100	<0.001*	
		Thoracic	0.116	0.120	0.028	0.093	0.138		
	Left	Pelvic	0.083	0.075	0.027	0.068	0.100	<0.001*	
		Thoracic	0.142	0.140	0.037	0.110	0.150		
Medial	Longitudinal diameter	Right	Pelvic	0.495	0.500	0.083	0.435	0.535	0.057
			Thoracic	0.545	0.535	0.055	0.503	0.565	
		Left	Pelvic	0.444	0.460	0.063	0.408	0.490	<0.001*
			Thoracic	0.578	0.570	0.060	0.530	0.610	
	Transversal diameter	Right	Pelvic	0.411	0.410	0.097	0.320	0.440	<0.001*
			Thoracic	0.574	0.595	0.079	0.493	0.610	
		Left	Pelvic	0.367	0.355	0.136	0.300	0.475	<0.001*
			Thoracic	0.545	0.570	0.161	0.520	0.650	
	Longitudinal wall	Right	Pelvic	0.095	0.090	0.025	0.070	0.115	0.006*
			Thoracic	0.119	0.115	0.026	0.100	0.138	
		Left	Pelvic	0.094	0.090	0.021	0.080	0.110	<0.001*
			Thoracic	0.120	0.120	0.025	0.100	0.140	
Transversal wall	Right	Pelvic	0.085	0.090	0.026	0.065	0.105	0.004*	
		Thoracic	0.114	0.110	0.030	0.093	0.138		
	Left	Pelvic	0.076	0.075	0.035	0.050	0.100	<0.001*	
		Thoracic	0.128	0.130	0.049	0.100	0.150		

\* P<0.05.

Performing B-mode ultrasonography in mules (Table 6), the lateral digital artery showed a significant difference between the pelvic and thoracic limbs by means of the variables: longitudinal (p<0.001) and transverse (p<0.001) diameters, longitudinal (p<0.001) and transversal (p<0.001) walls, with the left thoracic limb (LTL) presented higher value when compared to the left pelvic limb (LPL).

The medial digital artery presented difference between the pelvic and thoracic limbs in the longitudinal (p<0.001) and transverse (p<0.001) diameters, longitudinal (p= 0.007; p<0.001) and

transverse (p= 0.042; p<0.001) walls, with the right and left thoracic members presenting the highest values.

Performing spectral Doppler ultrasonography in equines (Table 7), the lateral digital artery showed a significant difference between the pelvic and thoracic limbs, in the RI (p= 0.004; p<0.001), PI (p= 0.006; p<0.001) and VM (p= 0.039; p= 0.012) measures, with RI and PI showing the highest values for the right and left pelvic limbs, while VM for the right and left thoracic limbs.

Table 6. Mean, median, standard deviation, Q1 and Q3 of the mules' limbs variables by B-mode ultrasonography, followed by the p-value to compare the digital arteries between the pelvic and thoracic limbs

Region	B-mode	Side	Limb	Mean	Median	Standard deviation	Q1	Q3	P-value	
Lateral	Longitudinal diameter	Right	Pelvic	0.552	0.550	0.106	0.490	0.630	0.552	
			Thoracic	0.538	0.540	0.090	0.490	0.580		
		Left	Pelvic	0.384	0.360	0.093	0.320	0.440		<0.001*
			Thoracic	0.552	0.570	0.052	0.510	0.580		
	Transversal diameter	Right	Pelvic	0.498	0.480	0.114	0.400	0.590	0.561	
			Thoracic	0.520	0.530	0.083	0.460	0.560		
		Left	Pelvic	0.402	0.370	0.066	0.350	0.460		<0.001*
			Thoracic	0.531	0.540	0.183	0.500	0.600		
	Longitudinal wall	Right	Pelvic	0.112	0.110	0.032	0.095	0.130	0.212	
			Thoracic	0.103	0.100	0.027	0.080	0.120		
		Left	Pelvic	0.088	0.080	0.023	0.070	0.100		<0.001*
			Thoracic	0.119	0.120	0.026	0.100	0.140		
Transversal wall	Right	Pelvic	0.101	0.100	0.036	0.070	0.120	0.173		
		Thoracic	0.110	0.110	0.032	0.080	0.130			
	Left	Pelvic	0.084	0.080	0.029	0.060	0.100		<0.001*	
		Thoracic	0.126	0.130	0.055	0.090	0.160			
Medial	Longitudinal diameter	Right	Pelvic	0.458	0.440	0.118	0.390	0.550	0.155	
			Thoracic	0.490	0.520	0.129	0.500	0.530		
		Left	Pelvic	0.401	0.400	0.076	0.350	0.450		<0.001*
			Thoracic	0.550	0.540	0.060	0.490	0.560		
	Longitudinal diameter	Right	Pelvic	0.450	0.430	0.126	0.330	0.590	<0.001*	
			Thoracic	0.550	0.530	0.083	0.500	0.600		
		Left	Pelvic	0.419	0.420	0.058	0.380	0.440		<0.001*
			Thoracic	0.567	0.590	0.150	0.510	0.650		
	Longitudinal wall	Right	Pelvic	0.084	0.080	0.026	0.070	0.100	0.007*	
			Thoracic	0.098	0.100	0.034	0.088	0.130		
		Left	Pelvic	0.089	0.090	0.022	0.070	0.100		<0.001*
			Thoracic	0.114	0.100	0.024	0.100	0.130		
Transversal wall	Right	Pelvic	0.094	0.090	0.026	0.080	0.110	0.042*		
		Thoracic	0.109	0.100	0.031	0.090	0.130			
	Left	Pelvic	0.091	0.090	0.028	0.070	0.100		<0.001*	
		Thoracic	0.141	0.130	0.052	0.100	0.185			

\* P&lt;0.05.

The medial digital artery showed a significant difference to RI ( $p<0.001$ ) and PI ( $p<0.001$ ) between the right pelvic limb (RPL) and the right thoracic limb (RTL), with RPL showing greater values.

The medial digital artery showed a significant difference to pSV ( $p=0.006$ ) and VM ( $p=0.010$ ) between the LPL and the LTL, in which the LTL presented the highest values. As for the fDV variable, there was a difference in the pelvic and thoracic limbs to medial digital artery, with the right ( $p=0.015$ ) and left ( $p=0.002$ ) thoracic limbs showing higher values than pelvic limbs.

There were differences in RI ( $p=0.027$  right) and PI ( $p<0.001$  right and  $p=0.011$  left) between the

pelvic and thoracic mules' limbs in the lateral digital artery assessed by spectral Doppler ultrasonography (Table 8), in which the pelvic limbs showed higher values than thoracic limbs. Regarding to pSV ( $p<0.001$ ), fDV ( $p<0.001$ ) and VM ( $p<0.001$ ), there were differences between the RPL and RTL, whereby the thoracic limb presenting the highest mean. In the medial digital artery, there were difference in RI ( $p<0.001$ ;  $p=0.039$ ) and PI ( $p<0.001$ ;  $p=0.029$ ), in which right and left pelvic limbs presented the highest means. Related to fDV ( $p=0.003$ ) and VM ( $p=0.017$ ) there was a significant difference between the RPL and the RTL, with the thoracic limbs showing higher values in comparison with pelvic limbs (Table 8).

Table 7. Mean, median, standard deviation, Q1 and Q3 of the horses' limbs variables by spectral Doppler ultrasonography, followed by the p-value to compare the digital arteries between the pelvic and thoracic limbs

Region	Doppler	Side	Limb	Mean	Median	Standard deviation	Q1	Q3	P-value	
Lateral	RI	Right	Pelvic	0.613	0.650	0.082	0.540	0.680	<b>0.004*</b>	
			Thoracic	0.564	0.570	0.053	0.525	0.600		
		Left	Pelvic	0.626	0.640	0.074	0.570	0.680		<b>&lt;0.001*</b>
			Thoracic	0.551	0.565	0.095	0.453	0.648		
	PI	Right	Pelvic	1.165	1.100	0.312	0.890	1.410	<b>0.006*</b>	
			Thoracic	0.986	0.935	0.189	0.870	1.130		
		Left	Pelvic	1.163	1.205	0.326	0.980	1.260		<b>&lt;0.001*</b>
			Thoracic	0.941	0.915	0.218	0.735	1.125		
	pSV	Right	Pelvic	29.15	23.35	14.90	15.35	44.32	0.178	
			Thoracic	30.98	26.96	11.95	22.62	34.54		
		Left	Pelvic	33.47	38.72	14.07	20.05	44.11		0.951
			Thoracic	34.12	36.47	10.98	22.25	40.89		
	fDV	Right	Pelvic	10.92	8.55	6.80	4.89	16.71	0.07	
			Thoracic	12.15	10.16	6.25	9.20	12.10		
		Left	Pelvic	12.45	12.72	6.99	6.55	14.66		0.059
			Thoracic	14.19	12.95	5.81	8.34	19.69		
	VM	Right	Pelvic	16.87	13.92	9.52	8.32	27.12	<b>0.039*</b>	
			Thoracic	19.38	16.18	8.76	14.77	20.55		
		Left	Pelvic	18.91	21.01	9.31	9.80	23.34		<b>0.012*</b>
			Thoracic	21.59	22.44	7.29	15.82	26.82		
Medial	RI	Right	Pelvic	0.650	0.675	0.088	0.590	0.720	<b>&lt;0.001</b>	
			Thoracic	0.597	0.620	0.072	0.570	0.650		
		Left	Pelvic	0.620	0.610	0.075	0.560	0.670		0.15
			Thoracic	0.593	0.600	0.087	0.520	0.660		
	PI	Right	Pelvic	1.281	1.355	0.272	1.023	1.510	<b>&lt;0.001*</b>	
			Thoracic	1.037	1.090	0.174	0.965	1.155		
		Left	Pelvic	1.150	1.090	0.286	0.970	1.210		0.627
			Thoracic	1.082	1.100	0.260	0.920	1.270		
	pSV	Right	Pelvic	30.07	32.64	13.20	16.19	43.23	0.695	
			Thoracic	30.05	31.65	12.61	16.23	39.80		
		Left	Pelvic	23.26	24.45	20.10	19.28	55.23		<b>0.006*</b>
			Thoracic	37,45	26.87	12.21	9.29	31.03		
	fDV	Right	Pelvic	10.04	9.18	5.78	4.45	13.56	<b>0.015*</b>	
			Thoracic	12.57	12.39	7.21	5.76	15.10		
		Left	Pelvic	13.26	9.61	6.76	7.94	19.64		<b>0.002*</b>
			Thoracic	9.25	8.55	6.37	3.24	11.82		
	VM	Right	Pelvic	16.42	16.04	8.17	8.11	22.33	0.111	
			Thoracic	18.07	20.01	7.44	9.21	23.27		
		Left	Pelvic	14.65	14.20	8.86	5.63	18.46		<b>0.010*</b>
			Thoracic	21.31	14.58	11.24	11.97	31.96		

RI = Resistivity index; PI = Pulsatility index; pSV = Systolic velocity; fDV = Final diastolic velocity; VM = mean velocity

\* P<0.05.

Table 8. Mean, median, standard deviation, Q1 and Q3 of the mules' limbs variables by spectral Doppler ultrasonography, followed by the p-value to compare the digital arteries between the pelvic and thoracic limbs

Region	Doppler	Side	Limb	Mean	Median	Standard deviation	Q1	Q3	P-value
Lateral	RI	Right	Pelvic	0.608	0.600	0.075	0.553	0.680	<b>0.027*</b>
			Thoracic	0.576	0.580	0.077	0.510	0.640	
		Left	Pelvic	0.636	0.655	0.094	0.543	0.708	0.13
			Thoracic	0.614	0.600	0.092	0.555	0.670	
	PI	Right	Pelvic	1.098	1.045	0.258	0.890	1.338	<b>&lt;0.001*</b>
			Thoracic	0.942	0.950	0.210	0.770	1.110	
		Left	Pelvic	1.198	1.230	0.314	0.923	1.438	<b>0.011*</b>
			Thoracic	1.077	0.980	0.328	0.830	1.200	
	pSV	Right	Pelvic	16.22	16.35	4.13	13.77	17.53	<b>&lt;0.001*</b>
			Thoracic	19.76	20.11	4.02	16.66	23.24	
		Left	Pelvic	15.78	15.41	4.89	11.39	20.92	0.65
			Thoracic	15.20	16.35	3.75	11.82	18.48	
	fDV	Right	Pelvic	6.19	5.91	1.82	4.80	7.29	<b>&lt;0.001*</b>
			Thoracic	8.40	8.37	2.03	6.96	9.61	
		Left	Pelvic	5.61	5.24	2.55	3.17	7.81	0.469
			Thoracic	5.80	5.23	2.31	3.57	8.07	
VM	Right	Pelvic	9.19	8.51	2.74	7.72	10.67	<b>&lt;0.001*</b>	
		Thoracic	12.63	12.88	2.60	10.14	14.47		
	Left	Pelvic	9.04	8.45	3.22	6.08	11.19	0.576	
		Thoracic	9.18	8.14	2.80	6.83	11.89		
Medial	RI	Right	Pelvic	0.626	0.600	0.078	0.560	0.680	<b>&lt;0.001*</b>
			Thoracic	0.565	0.565	0.071	0.523	0.610	
		Left	Pelvic	0.646	0.610	0.107	0.560	0.730	<b>0.039*</b>
			Thoracic	0.606	0.590	0.097	0.545	0.680	
	PI	Right	Pelvic	1.122	0.990	0.293	0.900	1.310	<b>&lt;0.001*</b>
			Thoracic	0.894	0.880	0.179	0.770	0.985	
		Left	Pelvic	1.274	1.090	0.492	0.900	1.568	<b>0.029*</b>
			Thoracic	1.100	0.980	0.375	0.840	1.320	
	pSV	Right	Pelvic	18.53	18.61	4.84	15.04	21.93	0.91
			Thoracic	19.22	18.11	6.41	13.39	22.02	
		Left	Pelvic	17.89	18.09	3.55	14.88	21.15	0.188
			Thoracic	17.21	16.03	5.09	13.19	22.48	
	fDV	Right	Pelvic	6.69	6.63	2.50	4.63	8.97	<b>0.003*</b>
			Thoracic	8.16	7.72	2.24	6.68	8.98	
		Left	Pelvic	6.19	6.15	2.24	4.58	8.22	0.494
			Thoracic	6.52	5.65	2.66	4.38	9.27	
VM	Right	Pelvic	10.91	11.14	3.27	8.21	13.61	<b>0.017*</b>	
		Thoracic	12.54	12.13	3.41	9.46	15.85		
	Left	Pelvic	10.06	9.14	2.44	8.62	12.18	0.881	
		Thoracic	10.52	9.30	3.71	7.43	13.51		

RI = Resistivity index; PI = Pulsatility index; pSV = Systolic velocity; fDV = Final diastolic velocity; VM = Mean velocity

\* P<0.05.

Comparing the right and left thoracic horses' limbs by B-mode ultrasonography, the lateral palmar digital artery showed a significant difference in the transverse diameter ( $p<0.001$ )

and in the transverse wall ( $p= 0.012$ ) (Table 9), with high mean to the left limb, whereas in the medial palmar digital artery of the thoracic limbs, there was a difference only in the

*Study of digital...*

longitudinal diameter ( $p= 0.046$ ), with LTL the highest mean.

There was no difference between the right and left limbs in the lateral digital plantar artery in

the pelvic horses' limbs. However, the medial plantar digital artery showed a significant difference in the longitudinal diameter ( $p=0.010$ ), with RPL showing higher mean.

Table 9. Mean, median, standard deviation, Q1 and Q3 of the horses' limbs variables by B-mode ultrasonography, followed by the p-value to compare the digital arteries between the pelvic and thoracic limbs

Limb	Region	B-mode	Side	Mean	Median	Standard deviation	Q1	Q3	P-value
Thoracic	Lateral	Longitudinal diameter	Right	0.527	0.540	0.067	0.450	0.580	0.986
			Left	0.541	0.520	0.073	0.510	0.555	
		Transversal diameter	Right	0.532	0.550	0.049	0.500	0.560	<b>&lt;0.001</b>
			Left	0.610	0.600	0.093	0.545	0.640	
		Longitudinal wall	Right	0.113	0.110	0.025	0.100	0.130	0.355
			Left	0.120	0.120	0.032	0.110	0.130	
	Transversal wall	Right	0.116	0.120	0.028	0.093	0.138	<b>0.012</b>	
		Left	0.142	0.140	0.037	0.110	0.150		
	Medial	Longitudinal diameter	Right	0.545	0.535	0.055	0.503	0.565	<b>0.046</b>
			Left	0.578	0.570	0.060	0.530	0.610	
		Transversal diameter	Right	0.574	0.595	0.079	0.493	0.610	0.739
			Left	0.545	0.570	0.161	0.520	0.650	
Longitudinal wall		Right	0.119	0.115	0.026	0.100	0.138	0.730	
		Left	0.120	0.120	0.025	0.100	0.140		
Transversal wall	Right	0.114	0.110	0.030	0.093	0.138	0.122		
	Left	0.128	0.130	0.049	0.100	0.150			
Pelvic	Lateral	Longitudinal diameter	Right	0.445	0.455	0.076	0.395	0.490	0.427
			Left	0.428	0.430	0.050	0.395	0.450	
		Transversal diameter	Right	0.410	0.400	0.066	0.345	0.460	0.894
			Left	0.411	0.390	0.061	0.370	0.455	
		Longitudinal wall	Right	0.084	0.080	0.023	0.070	0.100	0.616
			Left	0.090	0.090	0.032	0.068	0.103	
	Transversal wall	Right	0.083	0.090	0.024	0.070	0.100	0.893	
		Left	0.083	0.075	0.027	0.068	0.100		
	Medial	Longitudinal diameter	Right	0.495	0.500	0.083	0.435	0.535	<b>0.010</b>
			Left	0.444	0.460	0.063	0.408	0.490	
		Transversal diameter	Right	0.411	0.410	0.097	0.320	0.440	0.224
			Left	0.367	0.355	0.136	0.300	0.475	
Longitudinal wall		Right	0.095	0.090	0.025	0.070	0.115	0.980	
		Left	0.094	0.090	0.021	0.080	0.110		
Transversal wall	Right	0.085	0.090	0.026	0.065	0.105	0.321		
	Left	0.076	0.075	0.035	0.050	0.100			

\*  $P<0.05$ .

Comparing mules' contralateral limbs by B-mode ultrasonography (Table 10), the lateral palmar digital artery showed significant differences in the transverse diameter ( $p= 0.05$ ), longitudinal wall ( $p= 0.026$ ) and in the transverse wall ( $p = 0.049$ ), in which LTL showed the highest value. Whilst the medial palmar digital artery showed a significant difference in the longitudinal diameter ( $p= 0.047$ ) and in the transverse wall ( $p= 0.004$ ), with LTL presenting the highest mean (Table 10).

There was a significant difference in longitudinal ( $p<0.001$ ) and transverse ( $p <0.001$ ) diameters, longitudinal ( $p<0.001$ ) and transverse ( $p = 0.044$ ) walls in the lateral digital plantar artery to the pelvic limbs, with RPL showing higher values than LTL. The medial plantar digital artery there was a significant difference in diameter longitudinal ( $p= 0,034$ ), with RPL showing the highest values (Table 10).

Table 10. Mean, median, standard deviation, Q1 and Q3 of the mules' limbs variables by B-mode ultrasonography, followed by the p-value to compare the digital arteries between right and left side of pelvic and thoracic limbs

Limb	Region	B-mode	Side	Mean	Median	Standard deviation	Q1	Q3	P-value
Thoracic	Lateral	Longitudinal diameter	Right	0.538	0.540	0.090	0.490	0.580	0.296
			Left	0.552	0.570	0.052	0.510	0.580	
		Transversal diameter	Right	0.520	0.530	0.083	0.460	0.560	<b>0.05*</b>
			Left	0.531	0.540	0.183	0.500	0.600	
		Longitudinal wall	Right	0.103	0.100	0.027	0.080	0.120	<b>0.026*</b>
			Left	0.119	0.120	0.026	0.100	0.140	
		Transversal wall	Right	0.110	0.110	0.032	0.080	0.130	<b>0.049*</b>
			Left	0.126	0.130	0.055	0.090	0.160	
	Medial	Longitudinal diameter	Right	0.490	0.520	0.129	0.500	0.530	<b>0.047*</b>
			Left	0.550	0.540	0.060	0.490	0.560	
		Transversal diameter	Right	0.550	0.530	0.083	0.500	0.600	0.158
			Left	0.567	0.590	0.150	0.510	0.650	
		Longitudinal wall	Right	0.098	0.100	0.034	0.088	0.130	0.062
			Left	0.114	0.100	0.024	0.100	0.130	
		Transversal wall	Right	0.109	0.100	0.031	0.090	0.130	<b>0.004*</b>
			Left	0.141	0.130	0.052	0.100	0.185	
Pelvic	Lateral	Longitudinal diameter	Right	0.552	0.550	0.106	0.490	0.630	<b>&lt;0.001*</b>
			Left	0.384	0.360	0.093	0.320	0.440	
		Transversal diameter	Right	0.498	0.480	0.114	0.400	0.590	<b>&lt;0.001*</b>
			Left	0.402	0.370	0.066	0.350	0.460	
		Longitudinal wall	Right	0.112	0.110	0.032	0.095	0.130	<b>&lt;0.001*</b>
			Left	0.088	0.080	0.023	0.070	0.100	
		Transversal wall	Right	0.101	0.100	0.036	0.070	0.120	<b>0.044*</b>
			Left	0.084	0.080	0.029	0.060	0.100	
	Medial	Longitudinal diameter	Right	0.458	0.440	0.118	0.390	0.550	<b>0.034*</b>
			Left	0.401	0.400	0.076	0.350	0.450	
		Transversal diameter	Right	0.450	0.430	0.126	0.330	0.590	0.407
			Left	0.419	0.420	0.058	0.380	0.440	
		Longitudinal wall	Right	0.084	0.080	0.026	0.070	0.100	0.383
			Left	0.089	0.090	0.022	0.070	0.100	
		Transversal wall	Right	0.094	0.090	0.026	0.080	0.110	0.42
			Left	0.091	0.090	0.028	0.070	0.100	

\* P&lt;0.05.

The lateral digital palmar artery showed difference between the right and left, pSV ( $p = < 0.002$ ), fDV ( $p = < 0.002$ ) and VM ( $p = < 0.012$ ), with right showing higher values than left. The lateral digital plantar artery showed no difference between the right and left pelvic limbs, although the medial digital plantar artery showed a significant difference to RI ( $p = 0.039$ ), PI ( $p = 0.011$ ), pSV ( $p = 0.024$ ), fDV ( $p = 0.04$ ) and VM ( $p = 0.031$ ), with RPL values were significantly higher in RI and PI, and LPL to pSV, fDV and VM (Table 11).

Comparing the mules' contralateral limbs by spectral Doppler ultrasonography (Tab. 12), the lateral palmar digital artery showed significant differences in RI ( $p = 0.031$ ), PI ( $p = 0.039$ ), pSV ( $p = < 0.001$ ), fDV ( $p = < 0.001$ ) and VM ( $p = < 0.001$ ), which the RI and PI were higher in the LTL, and the pSV, fDV and VM to the RTL. There was a significant difference to RI ( $p = 0.022$ ), PI ( $p = < 0.001$ ), fDV ( $p < 0.001$ ) and VM ( $p = 0.002$ ) in the medial palmar digital artery, with the LTL showing higher values to RI and PI, and the RTL to fDV and VM. The mules' pelvic limbs did not presented difference between the right and left limbs.

*Study of digital...*

Table 11. Mean, median, standard deviation, Q1 and Q3 of the horses' limbs variables by spectral Doppler ultrasonography, followed by the p-value to compare the digital arteries between right and left side of pelvic and thoracic limbs

Limb	Region	Doppler	Side	Mean	Median	Standard deviation	Q1	Q3	P-value
Thoracic	Lateral	RI	Right	0.564	0.570	0.053	0.525	0.600	0.822
			Left	0.551	0.565	0.095	0.453	0.648	
		PI	Right	0.986	0.935	0.189	0.870	1.130	0.219
			Left	0.941	0.915	0.218	0.735	1.125	
		pSV	Right	30.98	26.96	11.95	22.62	34.54	0.12
			Left	34.12	36.47	10.98	22.25	40.89	
		fDV	Right	12.15	10.16	6.25	9.20	12.10	0.058
	Left		14.19	12.95	5.81	8.34	19.69		
	VM	Right	19.38	16.18	8.76	14.77	20.55	0.051	
		Left	21.59	22.44	7.29	15.82	26.82		
	Medial	RI	Right	0.597	0.620	0.072	0.570	0.650	0.701
			Left	0.593	0.600	0.087	0.520	0.660	
		PI	Right	1.037	1.090	0.174	0.965	1.155	0.381
			Left	1.082	1.100	0.260	0.920	1.270	
pSV		Right	30.05	31.65	12.61	16.23	39.80	<b>0.002*</b>	
		Left	23.26	26.87	12.21	9.29	31.03		
fDV		Right	12.57	12.39	7.21	5.76	15.10	<b>0.002*</b>	
	Left	9.25	8.55	6.37	3.24	11.82			
VM	Right	18.07	20.01	7.44	9.21	23.27	<b>0.012*</b>		
	Left	14.65	14.20	8.86	5.63	18.46			
Pelvic	Lateral	RI	Right	0.613	0.650	0.082	0.540	0.680	0.588
			Left	0.626	0.640	0.074	0.570	0.680	
		PI	Right	1.165	1.100	0.312	0.890	1.410	0.968
			Left	1.163	1.205	0.326	0.980	1.260	
		pSV	Right	29.15	23.35	14.90	15.35	44.32	0.16
			Left	33.47	38.72	14.07	20.05	44.11	
		fDV	Right	10.92	8.55	6.80	4.89	16.71	0.16
	Left		12.45	12.72	6.99	6.55	14.66		
	VM	Right	16.87	13.92	9.52	8.32	27.12	0.164	
		Left	18.91	21.01	9.31	9.80	23.34		
	Medial	RI	Right	0.650	0.675	0.088	0.590	0.720	<b>0.039*</b>
			Left	0.620	0.610	0.075	0.560	0.670	
		PI	Right	1.281	1.355	0.272	1.023	1.510	<b>0.011*</b>
			Left	1.150	1.090	0.286	0.970	1.210	
PSV		Right	30.07	32.64	13.20	16.19	43.23	<b>0.024*</b>	
		Left	37.45	24.45	20.10	19.28	55.23		
fDV		Right	10.04	9.18	5.78	4.45	13.56	<b>0.04*</b>	
	Left	13.26	9.61	6.76	7.94	19.64			
VM	Right	16.42	16.04	8.17	8.11	22.33	<b>0.031*</b>		
	Left	21.31	14.58	11.24	11.97	31.96			

RI = Resistivity index; PI = Pulsatility index; SS = Systolic velocity; fDV = Final diastolic velocity; VM = Mean velocity

\* P<0.05.

Table 12. Mean, median, standard deviation, Q1 and Q3 of the mules' limbs variables by spectral Doppler ultrasonography, followed by the p-value to compare the digital arteries between right and left side of pelvic and thoracic limbs

Limb	Region	Doppler	Side	Mean	Median	Standard deviation	Q1	Q3	P-value
Thoracic	Lateral	RI	Right	0.576	0.580	0.077	0.510	0.640	<b>0.031*</b>
			Left	0.614	0.600	0.092	0.555	0.670	
		PI	Right	0.942	0.950	0.210	0.770	1.110	<b>0.039*</b>
			Left	1.077	0.980	0.328	0.830	1.200	
		pSV	Right	19.76	20.11	4.02	16.66	23.24	<b>&lt;0.001*</b>
			Left	15.20	16.35	3.75	11.82	18.48	
		fDV	Right	8.40	8.37	2.03	6.96	9.61	<b>&lt;0.001*</b>
			Left	5.80	5.23	2.31	3.57	8.07	
	VM	Right	12.63	12.88	2.60	10.14	14.47	<b>&lt;0.001*</b>	
		Left	9.18	8.14	2.80	6.83	11.89		
	Medial	RI	Right	0.565	0.565	0.071	0.523	0.610	<b>0.022*</b>
			Left	0.606	0.590	0.097	0.545	0.680	
		PI	Right	0.894	0.880	0.179	0.770	0.985	<b>&lt;0.001*</b>
			Left	1.100	0.980	0.375	0.840	1.320	
		pSV	Right	19.22	18.11	6.41	13.39	22.02	0.082
			Left	17.21	16.03	5.09	13.19	22.48	
fDV		Right	8.16	7.72	2.24	6.68	8.98	<b>&lt;0.001*</b>	
		Left	6.52	5.65	2.66	4.38	9.27		
VM	Right	12.54	12.13	3.41	9.46	15.85	<b>0.002*</b>		
	Left	10.52	9.30	3.71	7.43	13.51			
Pelvic	Lateral	RI	Right	0.608	0.600	0.075	0.553	0.680	0.09
			Left	0.636	0.655	0.094	0.543	0.708	
		PI	Right	1.098	1.045	0.258	0.890	1.338	0.121
			Left	1.198	1.230	0.314	0.923	1.438	
		pSV	Right	16.22	16.35	4.13	13.77	17.53	0.535
			Left	15.78	15.41	4.89	11.39	20.92	
		fDV	Right	6.19	5.91	1.82	4.80	7.29	0.114
			Left	5.61	5.24	2.55	3.17	7.81	
	VM	Right	9.19	8.51	2.74	7.72	10.67	0.482	
		Left	9.04	8.45	3.22	6.08	11.19		
	Medial	RI	Right	0.626	0.600	0.078	0.560	0.680	0.348
			Left	0.646	0.610	0.107	0.560	0.730	
		PI	Right	1.122	0.990	0.293	0.900	1.310	0.214
			Left	1.274	1.090	0.492	0.900	1.568	
		pSV	Right	18.53	18.61	4.84	15.04	21.93	0.508
			Left	17.89	18.09	3.55	14.88	21.15	
fDV		Right	6.69	6.63	2.50	4.63	8.97	0.221	
		Left	6.19	6.15	2.24	4.58	8.22		
VM	Right	10.91	11.14	3.27	8.21	13.61	0.127		
	Left	10.06	9.14	2.44	8.62	12.18			

RI = Resistivity index; PI = Pulsatility index; pSV = Systolic velocity; fDV = Final diastolic velocity; VM = Mean velocity

\* P<0.05.

## DISCUSSION

According to Torres and Jardim (1997), horses have larger hooves than the mules, which suggests that the digital arteries of the thoracic

limbs are larger in horses due to the greater blood supply to the site, as confirmed ultrasound examinations B-mode and spectral Doppler in this study (Tab. 1 and 3).



We identified that the digital arteries in the pelvic limbs had higher values in the mules in some time-points (Table 2). According to Turner (2006), the horses' hooves of the pelvic limbs are smaller than those of the forelimbs, which may explain the higher values found in mules in the pelvic limbs in some measurements. Mules have intermediate characteristics between horses and donkeys (Miranda and Palhares, 2017), which may have influenced the variations in the measurements of horse and mule limbs.

Furthermore, blood vessels in bifurcating regions may present variations in diameter (Yanik, 2002) and, in addition, when evaluating B-Mode ultrasound, it is required to consider that measurements in the transverse plane may underestimate or overestimate the true values of the artery diameters and IMT (Liguori *et al.*, 2002; Broom, 2011). However, obtaining information only through the transverse plane is not totally accurate.

In the Doppler ultrasonography, there were differences between horses and mules in the values of pSV, fDV and MV of the thoracic and pelvic limbs, which were higher in horses. This also explains the fact that the vessel diameters of horses presented higher values in almost all limbs, since the increase in blood flow is related to the diameter of the vessel (Wongaumnuyaku *et al.*, 2006; Gargano, 2015). In addition, mules had a higher mean body weight (378 kg) than horses (364 kg), which cannot rule out the possibility that body weight may have influenced the Doppler measurements. According to Kanegusuku *et al.* (2020), human patients with higher body weight tend to have lower blood flow velocity, which may explain the fact that horses had higher values in the pSV, fDV and MV variables. Furthermore, some studies have shown that, despite considerable variability in measurements, the use of Doppler ultrasonography in the equine limb is sufficiently repeatable and sensitive to detect small changes in blood flow (Raisis *et al.*, 2000; Menzies-gow; Marr, 2007; Douthit *et al.*, 2014).

The variables RI and PI presented variations between horses and mules in this study. According to the literature, the mules are hybrid equines (Anderson, 1939; Camac, 1997; Araújo, 2010; Salles *et al.*, 2013) and, therefore, present differences in comparison to the

anatomophysiological studies of horses (Burnhan, 2002; Alsafy *et al.*, 2008; Smith, 2009; Miranda and Palhares, 2017) which may also explain the differences observed in this study. It is also essential to emphasize that the RI variable may be influenced by several factors, such as the compliance of the vessel wall, blood pressure and long-term effects of changing the hull geometry.

In horses and mules, the thoracic limbs are usually more affected by locomotor diseases; however, they may occur in all four limbs (Kahn, 2008). The digital palmar arteries of the thoracic limbs of horses and mules showed higher values of diameter and IMT when compared to those of the pelvic limbs. In spectral Doppler ultrasonography, horses and mules had higher values of pSV, fDV and VM in the thoracic limbs. According to Gargano (2015), blood flow is directly related to the increase in arterial diameter, which justifies the fact that horses and mules presented higher values of diameters and IMT (B-mode) for the thoracic limbs. Blood flow and vessel diameters were higher in the thoracic limbs, and naturally, the blood supply to these limbs is greater (Butler *et al.*, 2017) which may be related to the fact that those limbs support 55% to 60% of the animal's entire body weight, thus the blood flow demand for the digit must be proportional to the weight supported (Dyce *et al.*, 1997). Additionally, when a vessel undergoes increased resistance (RI), blood flow undergoes a smaller area of constant flow (Cipone *et al.*, 1997; Yanik, 2002; Varughese *et al.*, 2013), observed in the present study in horses and mules pelvic limbs.

In humans, blood vessels may present differences between the right and left limbs (Sayer and Fatherrree, 1945), also observed in the present study in both horses and mules. The arteries of the upper humans' limbs can vary from 9 to 24% (Celik *et al.*, 2001). In animals, this difference can also be observed, and this greater asymmetry is observed in the locomotor members (Wood, 1997).

The horses presented no difference in the lateral palmar digital artery and lateral plantar digital artery between the contralateral limbs by Doppler ultrasonography, similar to the observed by Menzies-Gow and Marr (2007) and Aguirre *et al.* (2013), who evaluated the lateral palmar

digital artery of both thoracic limbs of horses. However, the horses showed differences between the contralateral limbs in the medial palmar digital artery of the thoracic limbs and medial digital plantar artery of the pelvic members. Whilst in the mules, there were differences only between the thoracic limbs in the lateral and medial palmar digital arteries, in the pelvic limbs no difference was observed. According to Cochard *et al.* (2000), the comparison of blood flow in the digital palmar arteries of the horses' thoracic limbs using Doppler ultrasonography should not present significant differences between the contralateral limbs, although there may be some variability, different what was observed in the present study.

Only horses showed differences in the medial digital plantar arteries between the pelvic limbs. However, in healthy cattle Müller *et al.* (2017), observed difference between the right and left pelvic limbs in the lateral and medial digital plantar arteries by spectral Doppler ultrasonography. According to Stashak and Hill (2006), the animal's weight is not evenly distributed when resting its limb on the ground, supporting more medial lower of the hoof. The differences in changes in hoof pressure, induced by weight changes, may be responsible for the difference in blood flow between the right and left sides of the horses' locomotor limbs (Raisis *et al.*, 2000; Stashak and Hill, 2006, Müller *et al.*, 2017).

Depending on the anatomy, the lateral and medial palmar digital arteries are responsible for the digit's arterial supply, which originate from the bifurcation of the medial palmar artery and the lateral and medial digital plantar arteries originate from the bifurcation of the dorsal metatarsal artery III (Burg *et al.*, 2007; Parks, 2017; Fails, 2020).

According to Yanik (2002) changes in blood flow, vessel sizes and viscosity in regions of vessel bifurcations can occur, in addition to biological factors may change the blood flow (Cochard *et al.*, 2000; Menzies-Gow and Marr, 2007) which can justify the differences found between the contralateral in horses and mules' limbs.

The limb movement during B-mode and spectral Doppler ultrasonography exams is another factor

that can influence the digital arteries assessment, which can result in variations in blood flow velocity and the resistivity index (RI) (Menzies-Gow and Marr, 2007). Hoffmann *et al.* (2001), using spectral Doppler to analyze the animals' lateral palmar digital arteries with weight support (load) and weightlessness (no load), observed variations between 23.6 to 37.8% of the blood flow velocity in the limbs. Wongaumnuaykul *et al.* (2006), evaluated medial palmar digital arteries by spectral Doppler ultrasonography and observed variations in blood flow velocity (8.1 to 19.8%) between the control groups, the group with septic pododermatitis and laminitis. In the present study, ultrasonography examinations (B-mode and Doppler) were performed with body weight equally distributed over the four limbs to avoid hemodynamic changes and the RI, trying to maintain a pattern and minimum animals' mobility.

Despite B-mode and Doppler ultrasonography are influenced by technical and biological factors, there are studies that describe the use of this tool in diagnosis (Hunt *et al.*, 1994; Ingle-Fehr and Baxter, 1998; Cochard *et al.*, 2000; Menzies-Gow and Marr, 2007) and in the treatments monitoring, such as septic pododermatitis and laminitis in horses (Wongaumnuaykul *et al.*, 2006).

## CONCLUSION

Digital arteries had higher diameter and IMT values in horses in almost all limbs (B-Mode ultrasound). As for spectral Doppler, horses presented higher values in the variables pSV, fDV and MV, while RI and PI presented differences between horses and mules.

Horses and mules had the values of diameter, IMT, and blood flow superior to the thoracic limbs and differences between the contralateral limbs. The modalities of B-mode and spectral Doppler ultrasonography demonstrated efficacy in the evaluation of the palmar and plantar digital arteries in horses and mules and may show differences between the thoracic and pelvic limbs and their respective sides, and between animals, emphasizing the importance of performing ultrasonography in healthy animals.

## ACKNOWLEDGMENT

This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brasil (CAPES) - Finance Code 001, our sincere thanks.

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