



Using elastography (Acoustic radiation force impulse) to correlate the effects of age, weight, sex, and Norberg angle on the pectineus muscle of dysplastic dogs

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ABSTRACT: Inspired by the possibility of further understanding the role of pectineus muscle in the pathophysiology of hip dysplasia, this study evaluated the correlation between the Norberg angle, weight, sex, and age with the acoustic radiation force impulse (ARFI) elastographic findings of the pectineus muscle in dysplastic dogs through a quantitative analysis using shear wave velocity. Thirty-one dogs that had been radiographically diagnosed with hip dysplasia were selected, with a mean weight of 32.9 ± 8.8 kg and age of 4.3 ± 1.5 years. The dogs were submitted to mode B ultrasonography and ARFI elastography of the pectineus muscles, bilaterally. The mean value of the shear wave velocity was 3.5 ± 1.0 m/s, which showed a positive correlation with the degree of hip dysplasia ($P = 0.0065$; $r = 0.34$) and with age ($P = 0.0197$; $r = 0.30$). There was a negative correlation with the Norberg angle ($P = 0.0197$; $r = 0.30$), and there was no relation with body weight ($P = 0.7435$; $r = 0.04$). It was higher ($P = 0.0050$) in females (3.9 ± 1.1 m/s) compared to males (3.2 ± 0.8 m/s). The variable age contributed to the stiffness of the pectineus muscle in dysplastic dogs. The pectineus muscle was significantly stiffer in females than in males. Weight did not show any association with pectineus muscle stiffness, based on the ARFI elastography findings. The ARFI elastography exam has shown to be feasible in identifying changes in the pectineus muscle of dogs, which could be correlated with the studied variables.

Key words: dog, hip dysplasia, elastography, orthopedics.

Uso da elastografia (Impulso de força de radiação acústica) para correlacionar os efeitos de idade, peso, sexo e ângulo de Norberg no músculo pectíneo de cães displásicos

RESUMO: Inspirados na possibilidade de melhorar o entendimento sobre o papel do músculo pectíneo na fisiopatologia da displasia coxofemoral e de inclusão do exame elastográfico no rol de ferramentas diagnósticas para esta condição, objetivou-se avaliar a correlação existente entre o ângulo de Norberg, peso, sexo e idade com os achados elastográficos ARFI do músculo pectíneo de cães displásicos, por meio da análise quantitativa pela velocidade de cisalhamento. Foram selecionados 31 cães com diagnóstico radiográfico de displasia coxofemoral, com peso médio de $32,9 \pm 8,8$ kg e idade $4,3 \pm 1,5$ anos. Os cães foram submetidos à ultrassonografia modo B e à elastografia ARFI (Acoustic Radiation Force Impulse) dos músculos pectíneos, bilateralmente. O valor médio da velocidade de cisalhamento foi $3,5 \pm 1,0$ m/s, o qual apresentou correlação positiva com o grau de displasia ($P = 0,0065$; $r = 0,34$) e com a idade ($P = 0,0197$; $r = 0,30$). Houve correlação negativa com o ângulo de Norberg ($P = 0,0197$; $r = 0,30$), e não houve relação com o peso corporal ($P = 0,7435$; $r = 0,04$). Foi maior ($P = 0,0050$) em fêmeas ($3,9 \pm 1,1$ m/s) em comparação aos machos ($3,2 \pm 0,8$ m/s). As variáveis idade e grau de displasia coxofemoral contribuíram para a rigidez do músculo pectíneo em cães displásicos. O músculo pectíneo foi significativamente mais rígido em fêmeas do que em machos. O peso corporal não demonstrou associação com a rigidez do músculo pectíneo no exame de elastografia ARFI. O exame de elastografia ARFI mostra-se factível na identificação de alterações do músculo pectíneo de cães, os quais podem ser correlacionados com as variáveis estudadas.

Palavras-chave: cão, displasia coxofemoral, elastografia, ortopedia.

INTRODUCTION

Canine hip dysplasia is a common multifactorial hereditary disorder that is particularly problematic for large dogs and is highly correlated with hip laxity, osteoarthritis, chronic pain, and loss of

quality of life (LEDECKY et al., 2016; ROSSIGNOLI et al., 2020; SANTANA et al., 2021; CARNEIRO et al., 2023). Despite decades of research on this topic, the pathophysiological mechanisms of hip dysplasia have still not been completely elucidated, which highlights the relevance of studies that involve such

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mechanisms (MIKKOLA et al., 2019; CARNEIRO et al., 2023).

The pectineus muscle was extensively studied in the 1970 when the need to understand its role in the development of hip dysplasia in dogs arose (BERNARDÉ, 2010). Now, it is known that hip dysplasia promotes a limitation of limb movement due to muscle tension and rigidity of the pectineus muscle is strongly correlated with hip dysplasia in dogs. At a certain moment, it came to be considered an important part of the etiopathogenic mechanism, while others credited the changes reported to the side effects of the disease itself (ROSSIGNOLI et al., 2020). Nowadays, it is understood that theories related to secondary consequences are more plausible; however, there is still no consensus on this.

A new study, CARNEIRO et al. (2023), sought to propose an ultrasound protocol for evaluating the structures involved in hip dysplasia, the results of which increase in the pectineus muscles largely in dysplastic dogs. It was possible to identify differences between muscle specificity in young dysplastic and non-dysplastic dogs, that is, the pectineus plays a fundamental role in the disease, but it is not yet known how factors age, weight, sex and Norberg angle can behave in the face of muscle weakness.

Previous studies have shown the benefits of pectineus muscle tenotomy or myectomy in symptomatic dysplastic dogs after releasing muscle tension, which validates its role in the course of the disease; however, it does not confirm the hypothesis that it could be part of its etiology (VAUGHAN et al., 1975; BERNARDÉ, 2010). Pectineus myopathy has been identified in puppies that have become dysplastic in the future; however, no cause-and-effect relationship on the development of abnormal hip joints has been proven. Thus, pectineus muscle tenotomy or myectomy started to be used as symptomatic adjuvant therapy in the control of hip dysplasia. It was believed that pain relief would come from a combination of tension release in the joint capsule and increased abduction amplitude that occurs after surgery, which allows the femoral head to better articulate with the acetabulum. Thus, there is an improvement in the distribution of joint loads (WALLACE, 1992; EDGE-HUGHES, 2007). However, pectinectomy can worsen the severity of osteoarthritis according to some authors, which is why the procedure has become little used due to its unpredictable post-surgical nature, even allowing temporary pain relief (RAGHUVIR et al., 2013).

Pectineal myopathy is part of the set of musculoskeletal changes that accompany the

development of hip dysplasia in dogs, as it is rigid on palpation in dysplastic patients; However, to date, there is no information regarding the degree of stiffness of the pectineus muscle and its correlation with findings relating to hip dysplasia. There is a gap in knowledge regarding its role in clinical presentation or even diagnosis (CARDINET et al., 1969; KOWALESKI et al., 2013).

Elastography is an ultrasound technique that involves evaluating the degree of tissue stiffness (OPHIR et al., 2002). Additionally, it is an ultrasound-based imaging technique which provides information about the elasticity of tissue by assigning different chromatic patterns, according to different tissue elasticity responses (MCCAGHERT et al., 2020). The modality acoustic radiation force impulse (ARFI) is capable of providing quantitative and qualitative information by generating shear waves (MARONEZI et al., 2022) aiming to provide the elastic properties of a fabric (YOON et al., 2013). The method has proved to be widely useful in assessing the activity of the musculoskeletal system in humans (CASTREJÓN et al., 2013); however, it is still seldom used in veterinary medicine. In the area of small animals, only (PICCIELO et al., 2016) used elastography to evaluate patellar tendons in healthy dogs, demonstrating the importance of further studies on altered musculoskeletal tissues. The Acoustic Radiation Force Impulse (ARFI) method, conversely, qualitatively and quantitatively evaluates tissue elasticity; the quantitative evaluation consists of capturing the shear wave velocity, which expresses the degree of tissue stiffness and elasticity (GODDI et al., 2012).

Inspired by the possibility of further understanding the role of pectineus muscle in the pathophysiology of hip dysplasia and the inclusion of elastographic examinations in the list of diagnostic tools for this condition, this study evaluated the correlation between the Norberg angle, weight, sex, and age with the ARFI elastographic findings of the pectineus muscle in dysplastic dogs through a quantitative analysis using shear wave velocity.

MATERIALS AND METHODS

Experimental sampling

Thirty-one male ($n = 17$) and female ($n = 14$) dogs aged between 2 and 6 years (4.3 ± 1.5 years) with body weights of 20 to 50kg (32.9 ± 8.8 kg) were evaluated. All patients were diagnosed with some degree of hip dysplasia and were free from concomitant orthopedic conditions, based on orthopedic and radiographic examinations.

Radiographic technique

The animals were sent for hip joint evaluations using a SIEMENS X-ray generator model, Tridoros 812E, 800 milliamps and Agfa digitizer, model CR 30-X. The pre-anesthetic protocol consisted of applying 0.5mg/kg of chlorpromazine and general anesthesia through the intramuscular route with a continuous infusion of propofol at an initial rate of 0.5mg/kg/min. Mediolateral and ventrodorsal radiographic projections were used with the animals in dorsal decubitus, with the pelvic limbs fully extended, keeping the femurs parallel to each other and in relation to the spine, and the patellae internally rotated, which remained superimposed on the trochlear grooves; the same criterion is that adopted by the Colégio Brasileiro de Radiologia Veterinária (ABRV 2016). The entire pelvis, femurs, and knees were included in the radiographic image.

Norberg angle

Following the criteria of the Fédération Cynologique Internationale (FCI), based on the observed radiological characteristics, the Norberg method was adopted, which classifies the hip joint in 5 degrees: Grade A: Norberg angle greater than or equal to 105°, found in normal joints with the congruent femur head and acetabulum; Grade B: Norberg angle equal to or less than 105°, head of the femur and the acetabulum moderately incongruous; Grade C: Mild hip dysplasia reported in joints with the head of the femur and the incongruous acetabulum, osteoarthritic signs, and Norberg angle greater than 100° and less than 105°; Grade D: Moderate dysplasia with incongruity between the head of the femur and the evident acetabulum, subluxation, Norberg angle greater than 90° and less than 100°, and osteoarthritic signs; Grade E: Severe dysplasia with evident osteoarthritic signs, Norberg angle less than 90°, subluxation or dislocation (WIGGER et al., 2008).

Acoustic Radiation Force Impulse (ARFI) Elastography

ARFI (Virtual Touch Tissue Quantification) elastography was performed with a 9.0MHz linear transducer using the ACUSON S2000/SIEMENS device (Siemens, Munich, Germany) with a quantitative analysis software (FELICIANO et al., 2014). The animals were kept in dorsal decubitus, with the pelvic limbs completely flexed.

For the quantitative evaluation of the pectineus muscles of dysplastic animals, five samples of the image technique were obtained, so the shear

wave velocity was quantified in meters/second. The differences in the age, weight, and sex of the dogs were analyzed to establish the possible variations in the shear wave velocity obtained.

Statistics

The statistical analysis was performed with the software R (R Core Team 2019). Quantitative variables were compared using the Bland & Altman agreement method. A significance level of 5% ($P < 0.05$) was adopted and the data were presented as mean $\pm$ SD.

RESULTS

Thirty-one dogs with hip dysplasia were evaluated, with an average weight of 32.9 ± 8.8 kg and age of 4.3 ± 1.5 years, from 12 different breeds. In the quantitative ARFI elastography, the different regions of the pectineus muscle that had been evaluated showed similar shear wave velocity values ($P = 0.6394$) with an average difference between the regions of 0.9 ± 1.4 m/s. Thus, the mean value of the pectineus muscle shear wave velocity for the correlation study was 3.5 ± 1.0 m/s. The shear wave velocity value demonstrated a positive correlation with the classification of hip dysplasia ($P = 0.0065$; $R^2 = 0.34$). It also showed a positive correlation with age ($P = 0.0197$; $R^2 = 0.30$).

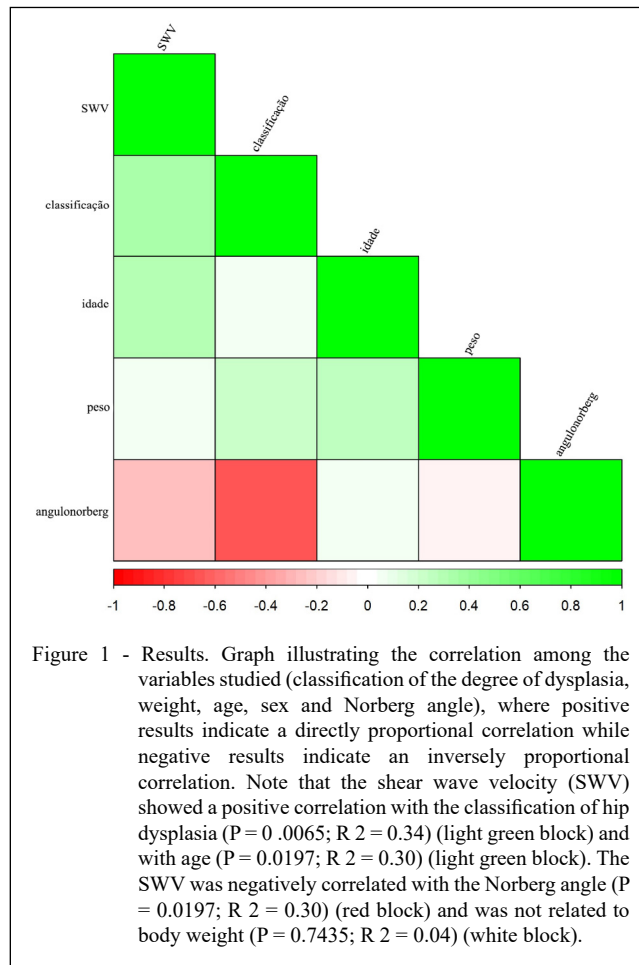
The shear wave velocity was negatively correlated with the Norberg angle ($P = 0.0494$; $r = -0.25$). There was no relationship with body weight ($P = 0.7435$; $R^2 = 0.04$). It was observed that the correlation was greater ($P = 0.0050$) in females (3.9 ± 1.1 m/s) than in males (3.2 ± 0.8 m/s) (Figure 1).

DISCUSSION

Stimulated by the need for a better understanding of the role of the pectineus muscle in the development, clinical presentation, and diagnosis of hip dysplasia, this study evaluated the use of ARFI elastography on the pectineus muscle in dysplastic dogs. The results of this study allowed an objective comparison of pectineus myopathy with variables related to patients with hip dysplasia.

In this study, the quantitative ARFI elastography showed that the stiffness of the pectineus muscle is higher in dysplastic dogs, demonstrated by the higher shear wave velocity. Clinically, there is a description of high tension upon palpation of the pectineus muscle.

However, objective or quantitative correlations had not yet been performed. Despite



the theories related to the fact that pectineus muscle tension could pull the femur against the acetabular border, and this is an important mechanism in the development of the disease (SMITH et al., 2017), there is still reason to believe that these changes are secondary. The results of this study depicted a positive correlation between the degree of dysplasia, the patient's age, and muscle tension through the shear wave velocity, which suggested that this hypothesis on secondary changes is valid. Additionally, the pectineus muscle myotomy performed on young dogs did not change the degree of dysplasia of patients in adulthood, supporting the theory that the role of the muscle is secondary (BERNARDÉ et al., 2010).

The dogs selected for experimental sampling that were 4.3 ± 1.5 years of age showed a positive correlation with the average value of shear wave velocity, which is what would have already been expected in view of what is understood about the pathophysiological mechanisms of the disease. The aging of dysplastic animals results in the progression and progressive worsening of degenerative joint

changes, and, with them, pectineus changes (VIEIRA et al., 2010). Associated the reduction in muscle volume, which was determined by the decrease in type I and II myofibrils, with advancing age (HEMELANDU et al., 1983).

The shear wave velocity value was influenced by the degree of hip dysplasia, which was demonstrated by the increased stiffness of the pectineus muscle in the higher degrees (C, D and E). If we correlate these findings with those of (MARTINEZ et al., 2016), we can verify important similarities, since histological alterations in the pectineus muscle were based on the degree of hip dysplasia. The results of this study provided, potentially, the possibility of minimally invasive evaluations on the progression and severity of degenerative joint changes related to hip dysplasia in dogs, which extends the benefits of ARFI elastography as a means of diagnosis and follow-up.

In this study, we found a negative correlation between the Norberg angle of dysplastic dogs and the elastographic findings; that is, higher values of shear wave velocity associated with the

stiffness of the pectineus muscle are correlated with a Norberg angle of less than 105°. According to the findings of (MARTINEZ et al., 2016), the degree of hip dysplasia observed from radiographic characteristics is highly correlated with the degree of morphological changes in the pectineus muscle. It was also shown that when a pectinectomy is performed before the occurrence of joint degeneration, dysplastic dogs show improvement in the Norberg angle.

The shear wave velocity was higher in females than in males, which shows greater stiffness of the pectineus muscle in dysplastic females. This finding has been mentioned previously; however, there is not enough evidence to affirm that it is a predisposition linked to sex. (VIEIRA et al., 2010) found a smaller Norberg angles, and a higher degree of hip dysplasia in females, while (ZHU et al., 2012) suggested that the discrepancy between males and females can be explained by the influence of the genes present on the sex chromosomes or by the influence of the secondary characteristics expressed by the genes. Even so, the occurrence of sex-dependent hip dysplasia remains controversial (MAKI et al., 2000; RETTENMAIER et al., 2002).

The results showed that the shear wave velocity of the pectineus muscle in dysplastic dogs did not correlate with body weight. This finding is also controversial, since it was expected that the weight gain associated with the rapid growth of large dog breeds could cause discrepancies in the development of muscle and bone tissue that could predispose them to hip dysplasia. However, based on the findings by (HELMINK et al., 2000), who reported variations in body weights due to differences in the growth curve based on breed, it can be suggested that the experimental design of this study may have led to results that differed from what had been expected.

CONCLUSION

The variables age and degree of hip dysplasia contribute to the stiffness of the pectineus muscle in dysplastic dogs, based on ARFI elastography. The pectineus muscle was significantly stiffer in females than in males. Body weight did not show any association with the stiffness of the pectineus muscle from the ARFI elastography exam. Despite its current limited utilization, elastographic analysis of the pectineus muscle constitutes a significant diagnostic adjunct in patients with hip dysplasia, warranting inclusion among complementary diagnostic modalities for informed therapeutic interventions.

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DECLARATION OF CONFLICT OF INTEREST

The authors have no conflicting interests.

AUTHORS' CONTRIBUTIONS

All authors contributed equally for the conception and writing of the manuscript. All authors critically revised the manuscript and approved of the final version.

BIOETHICS AND BIOSECURITY COMMITTEE APPROVAL

This study was evaluated by the Comitê de Ética com Uso de Animais (CEUA) of the Faculdade de Ciências Agrárias e Veterinárias (FCAV/UNESP), Jaboticabal Campus, with approval protocol number CEUA 3212/2017.

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