



Original Research

Influence of Exercise, Age, Body weight, and Growth on the Development of Tarsal Osteoarthritis in Young Mangalarga Marchador Horses



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ABSTRACT

In this study, the effect of early exercise, age, body weight (BW), and growth on the articular cartilage and subchondral bones of the tarsocrural joints was evaluated in 40 young Mangalarga Marchador horses allowed free choice exercise in pasture. Twenty of the horses had additional controlled exercise 3 days per week from a mean age of 30 ± 20 days until 36 months. The training program consisted of an increasing number of 15-minute gallop sprints in an oval paddock with a concrete floor covered by a thick layer of sand. BW, withers height (WH), and neck circumference were measured and body condition scores, cresty neck scores, and obesity index were rated. For each tarsus and foal, 5 standard radiographic projections were evaluated. All evaluations were performed at time point 1 (18 months of age) and time point 2 (36 months of age). Radiographic changes suggestive of tarsal osteoarthritis were observed in two male foals of the trained group at time point 2 (10% of 20). No horses from the untrained group developed OA. Training of the foals did not result in alterations of the morphometric parameters evaluated. However, significant differences were found between time point 1 and 2 in trained and untrained animals. At time point 2, the animals presented greater weights and WHs than at time point 1. We conclude that specific levels of physical activity during initial development do not increase the prevalence of osteoarthritic injury in Mangalarga Marchador foals.

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1. Introduction

Tarsal disease, specifically osteoarthritis (OA), is one of the most common causes of pain and lameness in athletic horses, which limits their performance and implies early retirement [1,2]. Osteoarthritis is a chronic degenerative joint disease characterized by progressive cartilage deterioration, accompanied by changes in the

bone and soft tissues of the joint. The end stage of OA results in a loss of joint space, pain, marginal osteophytosis, deformity, loss of motion, and decreased of joint function [3]. Cartilage damage due to trauma [1], failures of vascularization [3], joint instability [4,5], synovitis-capsulitis [6,7], nutritional factors [8,9], and genetic predisposition [10] can lead to changes in the composition, structure, and material properties of the tissue [11]. The articular cartilage is an avascular tissue that acts as a shock absorber for bone and provides a frictionless surface bathed in synovial fluid. Articular cartilage is composed of chondrocytes. The extracellular matrix (ECM) provides cartilage with its compressive strength and is composed of type II collagen and proteoglycans (PGs). Collagen forms a fibrous network, giving cartilage its tensile strength. Large aggregating PGs, composed of a protein core with several glycosaminoglycan (GAG) side chains attached, hydrate the collagen network and provide the tissue with viscoelastic properties and the ability to resist mechanical compression [12]. When the joint

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capsule is disrupted, proteolytic enzymes are secreted into the synovial fluid and can facilitate PG and collagen degradation [13]. In an attempt to repair structural changes in the ECM, chondrocytes proliferate and stimulate synthesis of these components. However, over time, the metabolic activity of chondrocytes shifts toward a state where the breakdown of matrix constituents outweighs new matrix synthesis [11,14], beginning the gradual process of ECM degeneration and thus the loss of articular cartilage [15].

Subchondral bone can also be affected by OA. Mechanical stimulation of that bone often leads to microdamage, which may result in (1) normal remodeling; (2) excessive remodeling, leading to bone sclerosis; or (3) accumulation of microdamage, leading to gross fracture [16]. Subchondral bone thickening is a normal response in exercising horses. However, an increase in the degree of subchondral bone sclerosis corresponds to greater degrees of generalized OA in joints [17]. In this context, moderate exercise has a positive effect on collagen network properties of subchondral bone [18,19]. However, forced exercise during the first months after birth appears to have a negative effect on collagen [20] and GAG levels [21]. Progressive degenerative changes in articular cartilage and subchondral bone, including fibrillation, loss of cartilage PG, striking focal resorptive lesions, have been identified in horses subjected to repetitive and strenuous exercise [22,23].

The Mangalarga Marchador (MM) breed is the most numerous and representative horse breed in Brazil and has been experiencing national and international expansion. The versatility of the MM horses together with their conformation and gait (Marcha Picada or Marcha Batida) are some of the attractive characteristics of these animals [24]. But recent studies have revealed alarming data on the incidence of tarsal OA in MM horses. High radiographic incidence of tarsal OA (83.3%, 25/30) was described in a group of MM horses aged between 12 and 36 months evaluated in training centers in the State of Espírito Santo, Brazil [25]. The authors suggested that excessive exercise in immature horses could be responsible for these findings. Another study carried out during the 33rd National Mangalarga Marchador Show in 2014 revealed the presence of tarsal disease in 65.38% of horses aged between 18 and 34 months, whereas 75% of the ridden horses aged between 41 and 58 months presented tarsal disease [26].

To our knowledge, the effects of controlled exercise in early life as well as the effect of age, body weight (BW), and growth, have not been investigated in a controlled manner in MM foals. The aim of this study was to investigate the influence of early exercise, age, BW and growth on the distribution and manifestation of tarsal OA lesions in a large group of foals at two stages in their development. The foals were raised under controlled conditions and subjected to two different exercise regimens. We hypothesized that in young foals, a conditioning exercise regimen combined with free pasture exercise would positively influence the development of tarsal OA.

2. Materials and Methods

2.1. Horses and Timing of Evaluation

All procedures were approved by the ethics committee of North Fluminense State University's ethics committee under protocol 212/2013. Forty newborn MM foals born within a two-month period were matched for sex and divided into two groups. The horses were managed identically from birth in pastures of approximately 3 hectares and weaned at 6 months of age. From a mean age of 30 ± 20 days to 36 months of age, the exercised group underwent a program of carefully regulated extra exercise combined with their normal pasture activity; the second group's horses had no imposed exercise but were free to move in the pasture. All horses were observed daily for clinical abnormalities, weighed, and

condition scored every two weeks and underwent clinical examination by a veterinarian every month. Feeding was based on grass (*Brachiaria mutica*) and Tifton hay (*Cynodon* spp), water, and mineral salt *ad libitum*. Foals of the trained and untrained groups were evaluated clinically and radiographically at time point 1 (18 months of age) and time point 2 (36 months of age).

2.2. Exercise Program

Horses in the trained group were exercised three days per week in an oval paddock with a concrete floor covered by a thick layer of sand. The horses were encouraged to trot, canter, and gallop in both a clockwise and counterclockwise direction with equal frequency. Horses were initially exercised with their mares at a constant base speed of 4.20 m/second. This was increased to 5.56 m/second at 6–18 months of age, and at 36 months of age, the base speed was increased to 6.6 m/second. Each exercise session lasted 15 minutes.

2.3. Radiographic Examination

For each tarsus and foal, five standard radiographic projections were evaluated (lateromedial, dorsoplantar, dorsolateral-plantaromedial oblique, dorsomedial-plantarolateral oblique, and mediolateral) [26]. Images were acquired using a portable X-ray machine (Quantus) and analog radiology system. Exposure parameters were set between 60 and 65 kv and 4 mAs. Two experienced clinicians reviewed the radiographs independently on separate occasions, without knowledge of the sex, age, or group of the horses. A foal was considered affected by OA if at least one radiographic finding for OA, fissures, and erosion of articular cartilage combined with osteophytosis, sclerosis, or cysts in the subchondral bone was identified [1,22,27]. Radiographic findings for OA in each limb were graded on a scale of 0–3 (0 = none, 1 = mild, 2 = moderate, and 3 = marked).

2.4. Morphometric Evaluations

Body weight was determined with an electronic scale with accuracy of 0.5 kg and capacity of 2,000 kg. The withers height (WH) was measured in the standing horses using a hipometer stick [28]. The neck circumference was obtained by means of the neck diameter corresponding to 25, 50, and 75% of the neck [29]. Neck measurements were taken while the neck was held in a relaxed position, at approximately a 45° angle. Two evaluators graded body condition score (BCS) ranging from BSC 1 (very poor) to BSC 9 (extremely fat) [30] and neck crest adiposity as cresty neck score (CNS) ranging from CNS 0 (no palpable crest) to CNS 5 (crest is so large it permanently droops to one side) [31]. Obesity index (OI) was calculated from the division of the circumference measurement of the neck corresponding to 50% of its total by the WH (P50: height).

2.5. Statistical Analysis

Descriptive statistics were calculated from the data on radiographic abnormalities observed. Analysis of variance was applied, followed by the Tukey test for parametric data and Kruskal–Wallis test for nonparametric data. Pearson's correlation was used to verify possible differences in prevalence of OA between groups and to verify the association between OA and age, weight, height, BCS, CNS, training, and OI. The statistical analyses were performed with SAS [32]. For all analyses, the significance level was set at $P < .05$. Data are presented as means $\pm$ standard deviation.

3. Results

At time point 1, no radiographic changes indicative of OA were identified in foals of either group. At time point 2, radiographic changes indicative of moderate (degree 2) OA of the tarsal joint were detected in 10% (2 of 20) of the foals only in the trained group. The changes affected only males. A significant relationship was found between the weight ($r = 0.4$, $P < .04$), training time ($r = 0.4$, $P < .05$), and occurrence of radiographic lesions. Significant differences were found between time points 1 and 2 in trained and untrained animals. At time point 2, all the animals presented weights and WHs larger than at time point 1 (Table 1).

A positive correlation was found between the weight and WH ($r = 0.8$; $P < .0001$), BCS and CNS ($r = 0.8$; $P < .0001$), and between the BCS and OI ($r = 0.7$; $P < .0001$) of untrained foals. Similarly, a positive correlation was found between weight and WH ($r = 0.8$; $P < .0001$), BCS and CNS ($r = 0.7$; $P < .0001$), and between BSC and OI ($r = 0.9$; $P < .0001$) in trained foals. At time point 1, a positive correlation was found between weight and BCS ($r = 0.8$; $P < .0001$). At time point 2, a positive correlation was observed between weight and WH ($r = 0.8$, $P < .0001$) and between CNS and OI ($r = 0.7$; $P < .0001$).

4. Discussion

Mangalarga Marchador horses usually begin training at a very young age, and they start competing in hand at 15–18 months and ridden from 36 to 39 months of age. Sometimes they are subjected to strenuous exercise including many cross-country competitions throughout the year. However, in this study, we found low prevalence of radiographic signs of tarsal OA in foals undergoing an early training program, contrasting with our hypothesis. Similar findings were described by Dykgraaf [33], when evaluating 12 young Thoroughbred horses allowed free choice exercise in pasture. In that study, six of the horses had additional controlled exercise 5 days per week from mean age of 21 ± 20 days (range: 3–83 days) until 17.1 months of age. Confocal laser scanning microscopy was used to quantify viable and nonviable chondrocytes, PG scoring was performed, and subchondral bone mineral density was measured by computed tomography. The results showed that the number of viable chondrocytes was significantly greater in the conditioned group, and there was no difference in mean bone mineral density between groups, nor was there a relationship between subchondral bone mineral density and chondrocyte viability. In view of the findings, the authors concluded that it is advantageous to provide controlled graded exercise in addition to free choice exercise to optimize articular cartilage quality.

Table 1
Measured parameters (mean $\pm$ SD) at time points 1 and 2 of trained and untrained foals.

Parameter	Time point 1		Time point 2	
	Untrained	Trained	Untrained	Trained
Weight (kg)	264.7 $\pm$ 48.63 ^a	219.5 $\pm$ 28.46 ^b	330 $\pm$ 36.21 ^a	284 $\pm$ 51.84 ^b
Height (m)	1.346 $\pm$ 0.07 ^a	1.328 $\pm$ 0.05 ^b	1.442 $\pm$ 0.03 ^a	1.407 $\pm$ 0.06 ^b
OI	0.54 $\pm$ 0.03	0.53 $\pm$ 0.01	0.59 $\pm$ 0.04	0.57 $\pm$ 0.05
BCS	6.0 $\pm$ 0.47	5.5 $\pm$ 0.52	6.7 $\pm$ 0.48	6.4 $\pm$ 0.69
CNS	2.1 $\pm$ 0.31	2.0 $\pm$ 0.00	2.6 $\pm$ 0.50	2.8 $\pm$ 0.91

Abbreviations: BCS, body condition score; CNS, cresty neck score; Moment 1, foals with 18 months of age; moment 2, foals with 36 months of age; OI, obesity index. Statistically significant difference between trained and untrained foals at specific time point, $P < .01$.

^a Statistically significant difference between time points 1 and 2 in untrained foals, $P < .01$.

^b Statistically significant difference between time points 1 and 2 in trained foals, $P < .01$.

Another study demonstrated that moderate exercise did not influence the collagen component of the ECM of 43 foals that were subdivided into 3 groups and submitted to different exercise regimens from one week after birth until 5 months of age [34]. The same authors also revealed that the exercise elicited a greater response on subchondral bone mineral density in quantitative terms [35]. A certain level of exercise seems essential for formation of biochemical heterogeneity of articular cartilage, so withholding exercise in early life may result in a delay in the adaptation of cartilage [36]. On the other hand, in many cases, training starts prematurely during the early stages of musculoskeletal system development, placing excessive and/or prolonged mechanical loads on immature articular cartilage, when periarticular tissues are inadequately developed to support intense loads [25]. However, although early exercise is often associated with the development of OA in young horses [20,21], other factors may be involved. Genetic predisposition, body size and growth rate, nutritional excesses or imbalances may also be involved. According to Van Weeren [21], all of these may play a role, but not all of them need to be present at the same time and their relative contribution to the possible clinical manifestation of the disease can vary significantly. In this context, a recent study in young Crioulo horses revealed a positive correlation of tarsal OA and body condition score, weight, and length of time needed for preparation to conformation competitions [37]. In the present study, despite the low incidence of OA, it was positively correlated with the weight of the animals and the time of physical training.

Conformation of the horse, especially tarsal angles and excess training, was associated with OA of distal tarsal joints in a group of MM gait champions [26]. Excess weight could have been responsible for OA in a group of MM horses aged between 12 and 36 months [25]. Positive correlation of tarsal OA and body score, body weight, and length of time needed for preparation to conformation competitions was observed in young Crioulo horses [37]. In rats, leptin, a cytokine produced by white adipocytes, regulates appetite, energy expenditure, and the activity of the physes and the metabolism of bone, shown to correlate positively with the severity of OA. There are no studies regarding the role of leptin in OA in horses. However, according to Buff [9], leptin is positively correlated with a high body mass index (obesity) in horses.

In the present study, only males presented radiographic changes indicative of OA. Different findings were obtained during the 33rd National Mangalarga Marchador Show. In this competition, of the 35 horses with tarsal diseases detected by radiology, 48.57% (17 of 35) were males and 51.43% (18 of 35) were females [26]. According to the authors, the findings were related to the overtraining of males in the attempt to improve their gait. Lower prevalence of radiographic signs of OA in males compared with females was also described in Icelandic horses [38]. However, young female Thoroughbreds in training were more affected by OA than males [39]. Gender differences in bone turnover between colts and fillies have been reported in 2-year-old racehorses [40] and could help in understanding the findings. Gender difference in sport horses has already been reported by other researchers, and in the case of OA, it could have a direct implication in establishing different training protocols for males and females.

5. Conclusions

The prevalence of tarsal OA in the MM foals was low, and the influence of the training program was not significant. However, the training time and body weight of the horses were positively related with the development of OA. Other factors associated or not with early exercise may contribute to the occurrence of OA in MM horses and should be better investigated.

CRediT authorship contribution statement

Paula Alessandra Di Filippo: Conceptualization, Methodology, Writing - original draft, Writing - review & editing. **Marcos Aurélio Dias Meireles:** Conceptualization, Methodology, Writing - original draft, Writing - review & editing. **Luiza Maria Feitosa Ribeiro:** Data curation, Formal analysis, Methodology. **Saulo Tinoco de Lannes:** Data curation, Formal analysis, Methodology. **Natália Ferreira Torres Meireles:** Data curation, Formal analysis, Methodology. **Inácio Silva Viana:** Data curation, Formal analysis, Methodology. **Helena Kiyomi Hokamura:** Writing - original draft, Writing - review & editing.

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