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Hospitalisation and Disease Severity Alter the Resting Pattern of Horses

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ABSTRACT

In humans, hospitalisation, disease type, and environmental factors evidently affect the quality of sleep, further influencing patient recovery. The objective of the present study was to report the resting and lying behaviour of hospitalised horses, and whether lying behaviours differ depending on the physiological severity of joint damage. We hypothesised that the resting and lying behaviour can change during the hospitalisation and physiological severity of joint damage affect the time of rest in horses. A descriptive observational study was performed to evaluate the effect of hospitalisation on the recumbency time of 8 adult horses with different degrees of osteoarthritis of the metacarpophalangeal joint. The horses' rest time was monitored using cameras during the first 5 days of hospitalisation. The total time of lateral recumbency and frequency of recumbency were greater after the 4^{th} day of hospitalisation (P<,05), while the total time of sternal recumbency was greater after the 3rd day (P < 0.05). Furthermore, we compared the recumbency time among animals with different degrees of osteoarthritis on the 5th day of hospitalisation. Increased recumbency time in mild osteoarthritis spared the animal's limb and reduced the overload on the affected limb; however, severe osteoarthritis decreased the frequency and time of recumbency probably due to greater difficulty during joint flexion in the transition from standing to recumbency. The severity of disease appeared to affect recumbency time, as horses with mild osteoarthritis spent more time in recumbency whilst those with severe osteoarthritis may have been partially sleepdeprived because they lay down less.

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1. Introduction

The duration of sleep has been extensively studied in humans, providing useful and applicable information for improving the well-being of people and physical performance of athletes [1]. Adequate rest is considered one of the pillars for success in therapeutic approaches and as part of training for athletes to achieve victory. Sleep can be divided into two phases, namely the rapid eye movement (REM) or paradoxical phase, where rapid eye movements occur, along with atony and loss of muscle tone, and the non-rapid eye movement (NREM) phase, where rapid eye movements do not occur, but muscle hypotonia is present and the heart and respiratory rates are decreased [2]. Complete sleep is characterised by the cyclic alternation of these phases [3].

Being a typical prey species, horses have evolved a sleep pattern divided into smaller fragments. Horses rest during approximately 30 NREM sleep periods lasting 3 –4 minutes each throughout the day [4]. However, to achieve and maintain REM or paradoxical sleep, animals must be in recumbency [5]. In horses, this phase is also short and lasts approximately 3 –5 minutes. Overall, totalling the REM and NREM phases, horses sleep for nearly 3 hours (7% to 15.5% of the 24-hour period), mainly during the night [6–10].

The recumbency time in stable horses has been characterised both during the 24-hour period and night [7–10]. While the recumbency time accounts for 8.2% to 15.5% of the 24-hour period, this percentage is higher during the night, reaching 19.9% of the time, and some animals lie down only at night [11]. Under adequate environmental conditions, most horses experience recum-







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Animal welfare/ethical statement: The present study was approved by the Ethics Committee on the Use of Animals (CEUA no. 4047060720) of FMVZ-USP. * Corresponding author at: Tiago Oliveira, FMVZ, São Paulo University, BR 05508-

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bency at least once a day [8]. However, prolonged recumbency position can result in cardiac and respiratory complications or organ and musculature compression [8].

Recumbency time has been used as an indicator of environmental adaptation, well-being, and pain [12,13], and the correlation between the animals' sleep pattern and the posture they adopt in stalls has been reported [14]. Data obtained from ethograms have shown a strong correlation of the recumbency time with the NREM and REM sleep times, finding the same sleep times from the data obtained from electroencephalograms, which indicated the kind of wave of each stage [5]. Different factors affect the recumbency time of animals, such as bedding type [15], bedding depth, presence of light in the stall [16], available lying area [17] and joint analgesia [12].

The effects of hospitalisation on rest have been described in humans [18] as well as in dogs and cats [19,20]; however, the effects of hospital environment on the physiological parameters of horses remain unknown. The type of injury is also important in the horse's choice to lie down, and some diseases, such as laminitis, can increase the recumbency time [21], while the others, such as enteroliths and back pain, can limit recumbency [22]. Osteoarthritis (OA) may affect the recumbency time, both in the hospital and at home, as it can generate different degrees of lameness and be induced as a consequence of training during the athletic career of horses [23,24]. Horses that are unable to lie down and are REM sleep-deprived may present progressive weight loss, falls, excessive sleepiness, and sores on the dorsal aspect of the fetlock and carpus joint [22,25,26]. Changes in time and type of weight overload on affected limb can damage joint cartilage [27], as a consequence of the lack of recumbency.

During routine medical care, horses exhibit numerous behavioural and physiological changes due to hospitalisation. However, the effects of joint damage severity in hospitalised horses at rest have never been studied, and whether the hospital environment affects the sleep pattern of such horses remains unknown.

To this end, the objective of the present study was to report the resting and lying behaviour of hospitalised horses, and whether lying behaviours differ depending on the physiological severity of joint damage, which may interfere with treatment and increase stress in the hospital environment. We hypothesised that the resting and lying behaviour can change during the hospitalisation and physiological severity of joint damage affect the time of rest in horses.

2. Material and Methods

2.1. Animals

Eight Lusitano mares between 5 and 8 years of age and weighing between 350 and 520 kg, that were affected by osteoarthritis (OA) of the metacarpophalangeal joint, were evaluated.

The animals were untrained and housed in $3.6 \times 4 \text{ m}^2$ stalls throughout the experimental period. Throughout the experimental period, the mares were fed coast-cross grass hay and water *ad libitum*, supplemented with commercial horse feed at a proportion of 1% of body weight and mineral vitamin compound. They were dewormed with a commercial compost based on ivermectin and were housed in stalls at the Equine Sports Medicine Laboratory (LAMEQ) in a separate building but close to the veterinary hospital. The present study was approved by the Ethics Committee on the Use of Animals (CEUA no. 4047060720) of FMVZ-USP.

The animals were subjected to lameness, radiographic, and ultrasound examinations to confirm OA and determine disease severity. Each examination was performed by a different examiner, and each examiner was unaware of the results of the other examination. Each animal was scored based on the results of lameness, radiographic [28], and ultrasound examination [29].

2.2. Experimental Design

A descriptive observational study was performed to assess the recumbency time during the first 5 days of hospitalisation, immediately after arriving at the hospital. Furthermore, an observational analytical cross-sectional study was performed to compare the recumbency time among animals with different degrees of OA [30].

The animals were brought in pairs in a randomised manner to2 monitored stables in the hospital environment, without visual or auditory contact with the animals in the stalls in which they were previously housed, but with visual and auditory contact with other hospitalised horses in the hospital's routine care. In the monitored stables at the veterinary hospital ($3.6 \times 4 \text{ m}^2$), the animals were subjected to the same food handling and stable cleaning practices performed by the same staff and were provided the same type and amount of bedding (approximately 5 cm of sawdust). The light inside the stables was turned off at night and turned on in the morning, if necessary. The animals were maintained in the monitored stables for six 6 days; the top half of a divided stable door was kept open throughout the day, allowing visual contact with other horses.

2.3. Video Image Capture and Analysis

Video images were recorded for 24 hours during the first 5 days of hospitalisation in the animal stables. Cameras that could be accessed or controlled via the internet, model Intelbras bullet VHD 1120, were installed in the stables. These cameras enable high image resolution and are infrared, allowing animal monitoring both during the day and night. This system was coupled to a DVR with HD 1Tb of four channels to record video images in HD and allow the storage of at least 15 days of recordings. The video images were backed up in person via a USB port.

After obtaining the images, the following measurements were obtained by plotting ethograms (Fig. 1):

- Wakefulness duration
- Drowsiness duration
- Right and left lateral recumbency duration
- Right and left sternal recumbency duration
- Frequency of lying down

2.4. Data Analysis

All statistical analyses were performed by 1 of the authors (PHET) in the RStudio integrated development environment (Version 4.0.2 [2020-06-22], RStudio, Inc.). The functions and packages are presented in the format "function {package}", and α was considered 5% in all analyses.

To examine the OA classificatory exams with the greatest influence on the recumbency behaviour, multiple regression ("Im {stats}") analysis was applied using the scores of radiographic, ultrasonographic, and physical examinations as the independent variables.

To investigate changes in the recumbency behaviour over the 5 evaluation days regardless of the presence of OA and according to the groups based on OA severity, generalised mixed models ("glm {stats}") were applied using the interaction of time and treatment as the fixed effect and horses as the random effect. Box charts ("ggplot {ggplot2}") were created to present the data.



Fig. 1. Images of horses showing different stages of rest (A). Wakefulness: The animal is standing, with eyes open and head above the withers. It may be eating or resting a limb. (B). Drowsiness: The animal is standing, with eyes open or closed and head below the withers. It cannot be eating and some limb may be flexed. (C). Sternal recumbency: The animal has stayed in right or left sternal recumbency, with flexed limbs, eyes open or closed, and muzzle in contact or not with the floor. (D). Lateral recumbency: The animal has stayed in right or left lateral recumbency, with limbs extended; the head, chest, and abdomen in contact with the floor; and eyes closed.

Table 1

Animal scores evaluated based on radiographic, physical (Silva et al., 2019), and ultrasound examaminations and the total of all scores (Yamada et al., 2020).

Animal	Radiographic Examination Score	Ultrasound Examination Score	Phisycal Examamination Score	Final Score
1	6	15	8	29
2	3	8	5	16
3	4	18	5	27
4	5	21	8	34
5	4	13	13	30
6	7	25	10	42
7	9	20	14	43
8	7	13	2	22

3. Results

In physical, radiographic, and ultrasound examinations to confirm OA and determine disease severity, the animals presented different degrees of OA and were allocated to two groups: those with mild damage, or discrete OA (DOA), and those with moderate-tosevere damage, or severe OA (SOA) (Table 1).

A higher physical examination score significantly reduced the right (P = .010248) and total (P = 0.00278) sternal recumbency time, whereas a higher ultrasound examination score significantly reduced the right lateral recumbency time (P = 0.02396) (Table 2). In models with other behaviours, no significant effects were observed.

Regardless of OA severity, the total lateral recumbency time and recumbency frequency were greater after the 4th day of hospitalisation, while the total sternal recumbency time was greater after the 3rd day. The left lateral recumbency time was greater on day 5 and the right lateral recumbency time was greater on day 4. The left sternal recumbency time was greater on days 3 and 5, and for the right sternal recumbency time was greater only on day 4. The total recumbency time was significantly greater after the 3rd day of hospitalisation.

Based on the effect of the interaction between time and group, the left lateral recumbency (p=0.03308) and total lateral recumbency (p=0.020552) time was longer in horses with DOA than for those with SOA (Figures 2 and 3).

4. Discussion

Regarding the time of recumbency in 24 hours, animals with both DOA (2.9%) and SOA (1.67%) rested in recumbency for a duration below the expected value for the species on the 1st day of hospitalisation. The animals with DOA reached the recumbency time close to the normal value on the 3rd day (13.92%), whereas those with SOA reached the maximum recumbency time of 9.02% on the 4th day. In previous animal studies, the average recumbency time of stable horses ranged from 11% to 15.5% [8–10]. In the present study, the reduction of recumbency time on the 1st day of hospitalisation may be explained by the inclusion of animals housed in the hospital environment, where horse handling by the medical team and the presence of sound and light near the stables can influence the resting pattern of animals, as previously reported [16].

Of note, the type and amount of bedding used were the same as those in the previous management, minimising the influence of this variable. This is important because thin [16] sawdust bedding [15] is known to shorten recumbency time. As the horses were only transferred to the hospital environment as the intervention, without changing the nutritional, housing management or han-

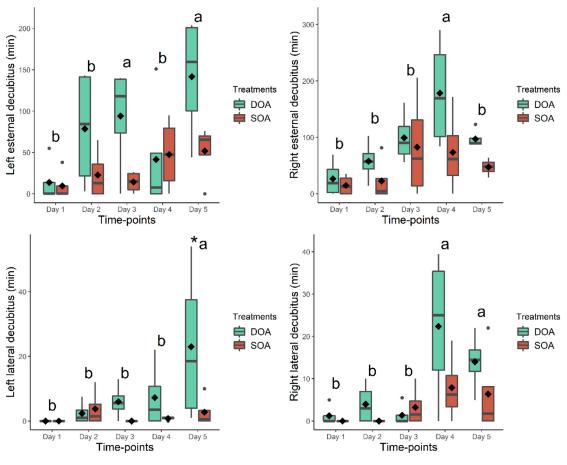


Fig. 2. Box chart showing the duration (minutes) of sternal and lateral recumbency on both sides (right and left) over 5 days (time-points) in horses with severe osteoarthritis (SOA) and discrete osteoarthritis (DOA). Different letters indicate changes over time regardless of the treatment (a > b). *Significant difference between groups taking into account the interaction of time and treatment. • indicates outliers; • indicates average.

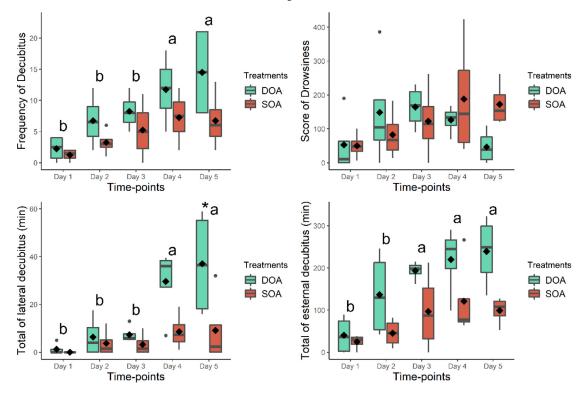


Fig. 3. Box chart showing drowsiness score, frequency of recumbency, total duration (minutes) of sternal and lateral recumbency over 5 days (time-points) in horses with severe osteoarthritis (SOA) and discrete osteoarthritis (DOA). Different letters indicate changes over time regardless of the treatment (a>b). *Significant difference between groups taking into account the interaction of time and treatment. • indicates outliers; • indicates average.

Table 2

Results of multiple regression analysis of the time of right sternal recumbency, right lateral recumbency, and total sternal recumbency.

	Right Sternal Recumbency						
Parameters	Estimate	Standart Error	t-Value	P-Value	Significance		
Intercept	137.918	36.730	3.755	.000612	***		
Radiographic examimation	6.699	6.248	1.072	.290747			
Ultrasound examimation	-2.671	2.360	-1.131	.265360			
Physical examimation	-7.526	2.777	-2.710	.010248	*		
	Right Lateral Recumbency						
Intercept	159.594	54.637	2.921	.00599	**		
Radiographic examimation	12.169	0.9293	1.309	.19867			
Ultrasound examimation	-0.8277	0.3511	-2.357	.02396	*		
Physical examimation	-0.3684	0.4132	-0.892	.37845			
	Total Sternal Recumbency						
Intercept	242.226	49.906	4.854	2.35^{-5}	***		
Radiographic examimation	2.989	8.489	0.352	.72681			
Ultrasound examimation	-2.344	3.207	-0.731	.46962			
Physical examimation	-12.121	3.774	-3.212	.00278	**		

* P<.05

** P<.01

*** P<.001

dling staff and available lying area [17], the observed partial sleep deprivation can be attributed to the changing environment and not husbandry factors within it. Further studies including hospitalised animals at other locations and with different types of management are warranted, as the impact of these factors on the sleep patterns of horses may be even greater.

Sleep is affected by several factors, including familiarity with the environment [5] and available lying area [17]. Stressing agents in the hospital environment, such as noise and light, can alter physiology and suppress immune response, cardiac function, and inflammatory response, which can cause changes in sleep patterns in humans [18,19]. Assuming that the clinical status of the horses remained stable during the 5 days of evaluation, the animals likely adapted to the hospital environment and evidently exhibited the expected normal rest behaviours after the 3rd day of evaluation. However, considering the difference in lateral recumbency between the DOA and SOA groups, the possible effects of the pain of horses during the first 5 days of hospitalisation should be considered. Overall, rest behaviours during the first 3 days of hospitalisation were strongly affected by the hospital environment.

The total recumbency time during 24 hours was longer in the horses with DOA (19.25%) and shorter in those with SOA (7.5%) than the previously reported values after the hospital adaptation period, indicating that the animals with SOA preferred to remain in the standing position rather than lying down, as opposed to those with DOA. This can be partially explained by the potential effort required for the horse to stand up after lying down. In horses, at the time of assuming recumbency, the first action is the flexion of the metacarpophalangeal and carpal joints of the forelimbs [9]. The animals evaluated here presented with OA of the metacarpophalangeal joint, however OA of the other joints may produce different effects on the recumbency time of horses. In the present study, we did not evaluate the transition from standing to recumbency, or behaviours that may have indicated pain in these animals, however significant differences in recumbent behaviours suggest that SOA influences the occurrences of recumbency.

The lateral recumbency time was longer in animals with DOA than in those with SOA. In general, the animals with DOA tended to exhibit longer time and frequency of recumbency. In a previous study, animals with LPS-induced synovitis exhibited a longer recumbency time than controls at 5 hour after induction [12]; however, the phenotype of acute illness differs from that of chronic

joint inflammation, and this difference is also important to recumbency time.

Mild OA increased recumbency time, such that the animal's limb was spared and the overload on the affected limb reduced; however, severe OA decreased the frequency and time of recumbency probably due to greater difficulty during joint flexion in the transition from standing to recumbency. The tendency of increased drowsiness in severe cases indicates sleep deprivation, since the animals recumbent less presented greater drowsiness during the rest of the day. In a previous study, no significant effect of bedding or light was identified on the drowsiness time of horses, despite a longer drowsiness time during the animals' acclimatisation to the bedding substrate [16].

Joint overload during exercise may increase the concentration of inflammatory cytokines in cases with OA [24]; however, the extent to which recumbency restriction can aggravate OA remains unknown. Our data indicate that animals with SOA spend more time standing, with more time supporting the weight on the affected limb; therefore, in these animals, OA may worsen due to the prolonged time of overload on the joint and decreased rest. In addition, our findings suggest that lateral recumbency time may serve as an additional behavioural indicator for monitoring OA severity in horses. Moreover, recumbency behaviour can be predicted based on physical and ultrasound examination scores, which may be useful for determining the severity of OA in hospital environments or animal stud farms. Such indicators may be effective to quantify the efficiency of analgesic therapy and improve prognosis.

Ethograms, as used in the present study, are easy to interpret and can effectively represent the resting patterns of horses. We noticed that with ethograms available in the literature, an animals was likely to be classified as being in REM sleep (sternal recumbency with the muzzle on the floor) although it was still responsive to the environment and had its eyes open; thus, we did not use those ethograms. We believe that the effects of the hospital environment can render the characterisation of sleep pattern difficult based solely on the position adopted in the stall. In this light, we suggest the use of our ethogram to determine the resting patterns of animals.

A limitation of the present study is the limited number of horses (n = 8) plus five time points (days 1 –5) and 2 groups (DOA and SOA), which created models with less power. Nonetheless, we observed marked differences over time and between groups, taking

into account the interaction between treatment and time. Furthermore, we did not have access to data on the sleep patterns of the included animals before they developed OA (control group); however, we could still compare our data with the recumbency times described in the literature.

The present study evaluated sleep patterns on only 5 consecutive days after the animal's introduction to the hospital environment; in other words, a longer adaptation period may allow a longer frequency and duration of resting time in horses. Of note, however, all parameters evaluated had stabilised by the 5th day of hospitalisation. Our objective was focused on short-term assessment, aiming to mimic the most frequent average length of stay in our hospital routine. The present findings may further our understanding of the resting patterns of OA affected horses in the hospital environment, representing relevant information to ensure animal safety, reduce the time of hospitalisation, and promote the well-being of the animal.

5. Conclusions

Recumbency time of horses was lower at the start of a period of hospitalisation than subsequent days. Moreover, the type and severity of injury affected the recumbency time, as horses with DOA spent more time in recumbency, whereas those with SOA spent more time in standing because they lie down less and for less time.

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