

Autonomic and hemodynamic effects of pre-anesthetic use of clonidine in healthy dogs

Efeitos autonômicos e hemodinâmicos do uso pré-anestésico da clonidina em cães saudáveis

João Paulo da Exaltação Pascon¹ , Fernanda Melo de Oliveira¹ , Marília Avila Valandro¹ ,
Diane Jaqueline Waschburger¹  & Roberto Thiesen¹ 

¹Veterinarian, Programa de Pós-Graduação em Ciência Animal, Universidade Federal do Pampa (UNIPAMPA), Uruguaiana, RS, Brazil.

Abstract

The aim of this study was to evaluate the effects of pre-anesthetic use of clonidine on time-domain heart rate variability (HRV) and arterial blood pressure in healthy anesthetized dogs. Six healthy adult mixed-breed dogs were administered a clonidine (clonidine group, CLG) and 30 days later, a placebo (control group, CG) preanesthetic protocol, in addition to propofol, isoflurane, and an bolus of tramadol and the continuous infusion thereafter. The total time mean values of HRV meanNN, SDNN, SDANN, SDNNI, and rMSSD were higher in the CL group, as observed in some HRV variables on tramadol bolus time (T4), tramadol continuous infusion (T8), and tracheal extubation time (T10). No significant differences in arterial blood pressure were observed, however, two dogs had a second-degree atrioventricular block (Mobitz II) at the tramadol bolus time (T4). These results led us to conclude that the clonidine anesthetic protocol resulted in sympathetic outflow block and an increase in parasympathetic tone, without significant effects on blood pressure. Notably, cardiac electrical disturbance in two dogs in the CL group. Although the pre-anesthetic use of clonidine in dogs with fear-based behavioral problems should be considered, its association with tramadol should be avoided or carried out with caution owing to the existing cardiovascular risk.

Keywords: fear-based behavioural problem, atrioventricular block, canine, alpha-2 agonist.

Resumo

Neste estudo objetivou-se avaliar os efeitos da administração pré-anestésica de clonidina na variabilidade da frequência cardíaca no domínio do tempo (VFC) e pressão arterial sanguínea de cães saudáveis anestesiados. Seis cães adultos hígidos, sem raça definida, foram submetidos a ambos protocolos anestésicos, com clonidina (grupo clonidina - GCL) e placebo (grupo controle - GC), associado ao propofol, isoflurano, *bolus* e infusão contínua de tramadol. Considerando o do tempo total de análise, os valores médios das variáveis de VFC NNmédio (GC=584.5±62.77, GCL=680.5±75.01), SDNN (GC=97.83±28.94, GCL=163.8±49.81), SDANN (GC=63.83±21.55, GCL=102.3±32.89), SDNNI (GC=60.83±28.53, GCL=110.2±42.92) e rMSSD (GC=75.83±38.91, GCL=158.0±81.20) foram maiores no protocolo anestésico com clonidina, assim como também observado em algumas variáveis de VFC durante o tempo de administração do bolus (T4) (NNmédio: GC=643,70±123,10, GCL= 819,80±78,77) e infusão contínua (T8) (NNmédio: CG=599,20±35,66, CLG=785,00±52,13) de tramadol, assim como no tempo de extubação orotraqueal (T10) (NNmédio: GC=598,70±84,75, GCL=852,50±188,60; SDNN: GC=49,83±33,49, GCL=193,80±143,40; rMSSD: GC=43,50±33,86, GCL=314,20±294,60). Nenhuma diferença significativa na pressão arterial sanguínea foi observada, porém, dois cães apresentaram bloqueio atrioventricular de segundo grau (Mobitz II) no momento de aplicação do bolus de tramadol (T4). Assim, o protocolo anestésico com uso de clonidina resultou em bloqueio eferente simpático e aumento do tônus simpático, sem efeitos significativos sobre a pressão arterial, mas com ocorrência de distúrbio elétrico de condução cardíaco em dois cães. Embora o uso pré-anestésico de clonidina em cães com problemas comportamentais baseados no medo deva ser considerado, sua associação com tramadol deve ser evitada ou realizada com cautela devido ao risco cardiovascular existente.

Palavras-chave: problema comportamental baseado no medo, bloqueio atrioventricular, canino, agonista alfa-2.




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*Correspondence

João Paulo da Exaltação Pascon
Programa de Pós-graduação em Ciência Animal, Hospital Universitário Veterinário, Universidade Federal do Pampa - UNIPAMPA BR 472, km 585
CEP 97501-970 - Uruguaiana (RS), Brasil
E-mail: joaopascon@unipampa.edu.br

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Introduction

Clonidine is a non-selective α -2 adrenergic agonist that can improve or prolong analgesia, and reduce the amount of anesthetic drugs required during surgical procedures in dogs (DeRossi et al., 2007). The drug can influence the autonomic balance of the heart, decrease catecholamine discharge in postganglionic sympathetic cardiac nerves, and decrease heart rate (Maze & Tranquilli, 1991). Therefore, clonidine is expected to interfere with cardiac autonomic balance (Cruz et al., 2011). However, some negative cardiovascular effects (autonomic and hemodynamic) limit their use in anesthetic protocols for animals (Nguyen et al., 2017; Sinclair et al., 2003).

The successful use of clonidine in the treatment of dogs with fear-based behavioral problems, by reducing fear responses, allows for new opportunity to use the drug a preanesthetic medication dog patients. These effects are related to the action exerted by the drug on presynaptic α -2 brainstem receptors, thereby decreasing sympathetic tone by inhibiting the release of norepinephrine, which is also associated with sedation, analgesia, and anesthesia (Ogata & Dodman, 2011).

In this context, heart rate variability (HRV) stands out for being an important, indirect, and non-invasive clinical tool to quantify cardiac autonomic regulation (Pattanapon et al., 2018; Vanderlei et al., 2009). The time-domain HRV is based on measuring the variation in R-R intervals of normal beats, registered by a surface electrocardiogram, which reflects the autonomic cardiac balance (Billman, 2013; Doxey & Boswood, 2004; Rasmussen et al., 2012).

Thus, to evaluate the feasibility of the use of clonidine in anesthetic protocols for dogs with fear-based behavioral problems, this study aimed to analyze the effects of the intramuscular premedication use of clonidine at 5 μ g/kg, on time-domain HRV and arterial blood pressure in anesthetized healthy dogs.

Materials and methods

A total of six mixed-breed, intact healthy dogs, including males (n=3) and females (n=3), with body weights ranging from 11.6 to 19 kg and ages ranging from 2 to 6 years, were enrolled in this study. The health status of the dogs was verified by physical and hematological evaluation (complete blood count, serum creatinine levels, urea, and liver enzymes alanine aminotransferase and alkaline phosphatase), without clinical evidence of fear-based behavioral problems.

All animals were randomly anesthetized with clonidine (CLG; clonidine group) and a placebo (CG; control group) as premedication (paired study), with a 30 day interval between each procedure. The enrolled dogs were fasted for 12 h before receiving premedication. Saline solution (NaCl 0,9%) was used in the CG, and clonidine diluted up to 1 ml with saline solution was in the CLG. Anesthetic induction was performed with propofol 15 min after premedication by cephalic vein access. The animals were intubated with an endotracheal tube of appropriate size and maintained under spontaneous ventilation. Isoflurane at 1 minimum alveolar concentration (MAC) (approximately 1,41%) was administered to maintain anesthesia. Additionally, an intravascular bolus of 2 mg/kg tramadol hydrochloride was continuously infused at a rate of 3 mg/kg/h in both groups.

To infer the hemodynamic and heart autonomic effects of clonidine, the time-domain HRV, heart rate, and arterial blood pressure were monitored. A three-channel digital Holter (Cardiolight™, manufacturing company, city, country) recorded the dog's electrocardiograms (ECG), as described by Calvert (1998), and the time-domain HRV variables were obtained by analysis and editing of the ECG records by the same experienced evaluator using a specific software (CardioManenger CS550™, manufacturing company, city, country). Premature complexes and their previous and subsequent complexes were excluded from the calculation of the variables, MeanNN (average of all regular R-R (NN) intervals), SDNN (standard deviation (SD) of all regular NN intervals), SDANN (SD of 5-minutes average NN intervals), SDNNi (Mean of the SD of all NN intervals for all 5-minute segments), rMSSD (square root of the mean of the squares of successive NN interval differences), and pNN >50 (the percentage of intervals longer than 50 ms different from the preceding interval).

Electrocardiographic recordings of all dogs were started 20 min before premedication and finalized after tracheal extubation. The analysis of HRV in the time domain was performed during the total period of electrocardiographic recording in five minute intervals and of recorded TO (before the administration of premedication- basal), T1 (after premedication, immediately before application of propofol), T2 (immediately after the beginning of induction with propofol), T3 (after

vaporization of isoflurane immediately before the bolus tramadol), T4 (beginning of application of tramadol bolus), T5 (10 min after T4), T6 (20 min after T4), T7 (30 min after T4), T8 (40 min after T4), T9 (50 min after T4), and T10 (immediately before extubation).

The invasive systolic (SAP), diastolic (DAP), and mean (MAP) arterial blood pressure were obtained by pedal artery catheterization and recorded every 10 min after tramadol infusion for 60 min in a multiparameter monitor⁴ [LW6000™, manufacturing company, city, country].

Considering the total time of evaluation, the average values of time-domain HRV (MeanNN, SDNN, SDANN, SDNNi, rMSSD, pNN^{>50}), and heart rate (maximum, average, and minimum) were compared between the CG and CLG groups using a paired t-test ($p < 0.05$). In turn, the mean variables MeanNN, SDNN, rMSSD, and pNN^{>50} of the moments (T0 to T10), and SAP, MAP, and DAP at each evaluated time (zero, 10, 20, 30, 40, 50, and 60 min) were subjected to analysis of variance (ANOVA) for repeated averages over time and compared between the CG and CLG groups within each moment using the Bonferroni test ($p < 0.05$). All analyses were performed using statistical software (GraphPad Prism v.5.04, manufacturing company, city, country).

Results

Considering the total time of evaluation, the average variables of time-domain HRV meanNN, SDNN, SDANN, SDNNi, and rMSSD were higher in the CLG ($p < 0.05$) than in the CG (Table 1). Additionally, the minimum and average heart rates were lower in the clonidine group (Table 1).

Table 1. Comparison of total time of registration of the heart rate variability, and heart rate (minimum, mean and maximum), and arterial blood pressure (SAP, DAP, MAP) values (average \pm standard deviation) between control (CG) and clonidine (CLG) groups.

Variables	CG	CLG	P'
MeanNN (ms)	584.5 \pm 62.77	680.5 \pm 75.01	0.0371
SDNN (ms)	97.83 \pm 28.94	163.8 \pm 49.81	0.0186
SDANN (ms)	63.83 \pm 21.55	102.3 \pm 32.89	0.0374
SDNNi (ms)	60.83 \pm 28.53	110.2 \pm 42.92	0.0410
rMSSD (ms)	75.83 \pm 38.91	158.0 \pm 81.20	0.0494
pNN ^{>50} (%)	21.87 \pm 15.82	39.00 \pm 15.63	0.0886
HRmin (bpm)	65.83 \pm 6.853	52.50 \pm 9.524	0.0193
HRmean (bpm)	107.8 \pm 8.542	91.83 \pm 8.542	0.0088
HRmax (bpm)	212.5 \pm 29.68	188.3 \pm 29.68	0.2605

*Paired t test p value; MeanNN - average of all regular R-R (NN) intervals; SDNN - standard deviation (SD) of all regular NN intervals; SDANN - SD of 5-minutes average NN intervals; SDNNi - Mean of the SD of all NN intervals for all 5-minute segments; rMSSD - square root of the mean of the squares of successive NN interval differences; pNN^{>50} - percentage of intervals longer than 50ms different from preceding interval; HRmin - minimal heart rate; HRmean - average heart rate; HRmax - maximum heart rate.

During the first minute of the tramadol bolus (T4), an important autonomic synergic effect with clonidine was observed in the CLG, as demonstrated by the increase in meanNN variable (Table 2, Figure 1), and the occurrence of second-degree atrioventricular block (Mobitz II) in two dogs.

The values of meanNN were also higher in the CLG at times T8 and T10 ($p < 0.05$), however, only at T10 did CLG have superiority of SDNN and rMSSD variables ($p < 0.05$), suggesting increase in parasympathetic tone and decrease in sympathetic tone at these moments, especially in T10 (Table 2, Figure 1). Considering the reduction in anesthetic drug rate and sedative effect at the extubation time (T10), it is correct to assume that the autonomic balance, with an increase in heart sympathetic and decrease in parasympathetic influence, tends to occur in a proportional manner for anesthetic recovery, which was delayed in the CLG compared to the CG at this time (T10). No difference in time to extubation was observed ($p > 0.05$) between the CG (139.2 \pm 18.96 min) and CLG (132.3 \pm 9.20 min), which probably had no influence on the observed differences between the groups.

Table 2. Time domain heart rate variability on different times of evaluation in a control (CG) and clonidine (CLG) groups of dogs.

Times	Groups	meanNN	SDNN	rMSSD	pNN>50
T0	CG	513.70±75.83	102.20±34.64	123.00±67.69	45.64±15.91
	CLG	550.20±98.28	116.30±41.38	134.00±61.99	50.81±23.98
T1	CG	546.80±113.10	112.80±49.39	125.50±81.90	45.58±19.87
	CLG	649.00±125.40	157.00±62.89	203.50±127.50	58.44±30.97
T2	CG	513.50±78.35	97.17±39.18	67.17±24.28	16.97±3.69
	CLG	593.20±113.00	86.83±37.84	57.50±42.20	16.49±15.97
T3	CG	616.70±52.19	43.50±28.50	28.00±18.53	9.81±11.32
	CLG	680.20±56.14	75.00±30.65	56.83±44.21	20.58±23.41
T4	CG	643.70±123.10*	51.83±64.37	64.50±92.59	23.63±35.86
	CLG	819.80±78.77	118.50±124.00	161.20±210.80	50.65±26.38
T5	CG	634.50±88.07	46.17±55.27	50.67±61.08	21.49±32.27
	CLG	780.50±71.96	68.17±57.46	79.83±85.56	34.17±33.30
T6	CG	640.20±74.98	44.00±38.08	52.33±58.95	21.80±33.49
	CLG	752.30±74.31	57.83±35.31	61.00±57.45	28.43±32.36
T7	CG	647.70±64.60	48.67±37.85	54.67±61.70	25.52±33.93
	CLG	762.30±34.85	73.67±44.71	79.50±59.14	33.94±29.72
T8	CG	599.20±35.66*	39.67±25.71	35.17±27.01	15.58±19.37
	CLG	785.00±52.13	87.67±61.08	112.30±93.64	48.89±33.20
T9	CG	606.30±64.30	34.00±16.75	31.67±21.59	13.28±15.13
	CLG	750.70±90.37	90.33±64.17	104.80±101.30	41.44±37.96
T10	CG	598.70±84.75*	49.83±33.49*	43.50±33.86*	16.51±17.81
	CLG	852.50±188.60	193.80±143.40	314.20±294.60	60.04±34.45

*Represent a statistical difference between groups (CG and CLG) in each time of evaluation, attested by Bonferroni test ($p < 0,05$); MeanNN - average of all regular R-R (NN) intervals; SDNN - standard deviation (SD) of all regular NN intervals; rMSSD - square root of the mean of the squares of successive NN interval differences; pNN> 50 - percentage of intervals longer than 50ms different from preceding interval.

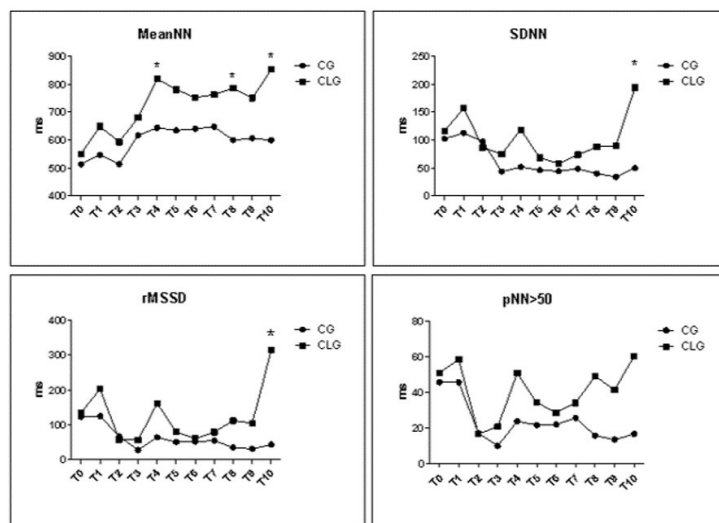


Figure 1. Time domain heart rate variability on different times of evaluation in a control (CG) and clonidine (CLG) groups of dogs. *Represent a statistical difference between groups (CG and CLG) in each time of evaluation, attested by Bonferroni test ($p < 0,05$); MeanNN - average of all regular R-R (NN) intervals; SDNN - standard deviation (SD) of all regular NN intervals; rMSSD - square root of the mean of the squares of successive NN interval differences; pNN> 50 - percentage of intervals longer than 50ms different from preceding interval.

The arterial blood pressure was also compared between groups every 10 min for 60 min after tramadol administration. However, instead of the expected reduction in the CLG, no differences in SAP, DAP, or MAP were observed at any time (Table 3 and Figure 2). Although no differences in arterial pressure were observed in the groups, at different time points, SAP, DAP, and MAP showed a tendency to increase along the evaluated times in both groups (Figure 2).

Table 3. Average and standard deviation values of systolic (SAP), diastolic (DAP), and mean (MAP) arterial blood pressure, on different times of evaluation in a control (CG) and clonidine (CLG) groups of dogs.

Times	Groups	SAP	DAP	MAP
Zero	CG	78.50±12.34	58.12±11.80	66.12±11.89
	CLG	77.62±6.41	55.50±2.33	63.75±2.81
10	CG	82.50±7.96	57.00±8.88	69.37±7.19
	CLG	81.25±12.12	55.75±6.31	65.50±7.85
20	CG	85.00±6.02	59.75±6.06	71.37±3.66
	CLG	77.87±9.32	54.75±5.33	63.25±5.36
30	CG	84.50±4.14	62.62±7.04	75.25±6.40
	CLG	79.85±13.28	57.37±10.23	66.50±9.65
40	CG	87.50±3.20	60.87±4.18	72.25±2.12
	CLG	81.12±13.98	61.50±10.98	67.50±12.45
50	CG	89.37±5.70	63.50±4.24	74.87±4.94
	CLG	80.50±13.44	62.62±8.55	69.50±8.83
60	CG	92.75±4.97	64.62±4.24	76.75±4.95
	CLG	86.00±10.21	62.25±6.62	71.12±7.14

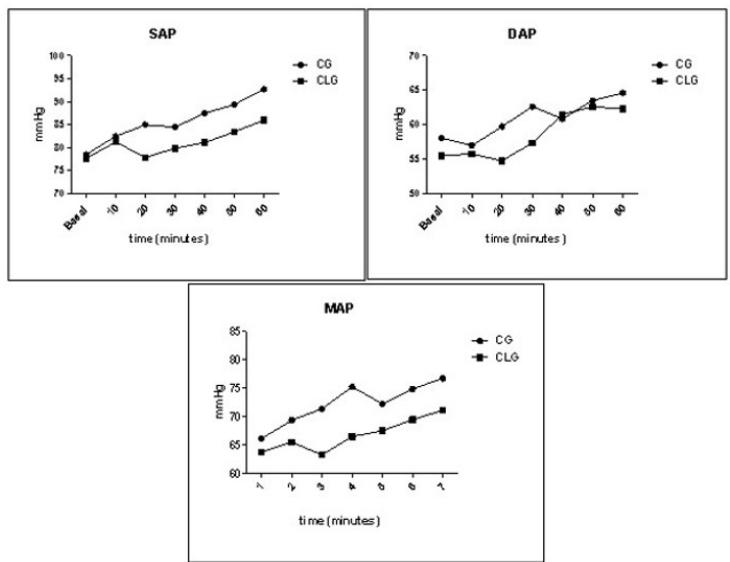


Figure 2. Average values of systolic (SAP), diastolic (DAP), and mean (MAP) arterial blood pressure obtain along times of evaluation, in a control (CG) and clonidine (CLG) groups of dogs.

Discussion

The findings of this study revealed that intramuscular clonidine premedication at a dose of 5µg/kg results in decreased sympathetic tone and increased parasympathetic tone in the heart.

Some HRV time-domain indices such as meanNN, SDNN, SDANN, and SDNNi can be influenced indistinctly by sympathetic and parasympathetic tonus (Khan et al., 1999). However, the increase in rMSSD strongly suggests an increase in the parasympathetic heart influence (Billman, 2013) by clonidine (CLG). This parasympathetic influence was already attributed to a direct central parasympathetic outflow effect of a clonidine in humans (Girgis et al., 1998).

Furthermore, the observation of an autonomic synergic effect of tramadol and clonidine in CLG can be explained by the fact that tramadol is a synthetic opioid that is widely used for analgesia in dogs by different mechanisms, including the reduction of neuronal uptake of norepinephrine in the central nervous system (McMillan et al., 2008). This is also seen as an effect of clonidine, which is responsible for the reduction of sympathetic outflow (Hysek et al., 2012; Mitchell et al., 2005; Ogata & Dodman, 2011). These complementary mechanisms of reducing neuronal epinephrine uptake by tramadol and clonidine may have resulted in an additive autonomic effect due to the potentiation of the sympathetic block.

Although this synergistic effect has been observed only in one HRV variable (meanNN), the association of clonidine and tramadol should be avoided considering the risk of patients developing cardiac conduction disturbances, as observed in the two dogs who died during the study. This kind of conductance disturbance has also been observed in dogs anesthetized with different doses of clonidine, induced by etomidate, and maintained under halothane anesthesia (DeRossi et al., 2007). However, in the referred study, clonidine was intravenously administered, and there was no description of the moment of occurrence of the electric disturbance. Nonetheless, future investigations must be conducted to clarify the synergistic mechanisms of tramadol and clonidine.

The effects of clonidine on blood pressure can be caused by multiple mechanisms, such as stimulation of alpha-2 adrenoceptors in the brain stem, attenuating sympathetic and increasing the parasympathetic outflow (Hysek et al., 2012; Mitchell et al., 2005). This is mediated by the baroreflex mechanism (Leino et al., 2020), resulting in hypotension. In contrast, vascular activation of the alpha2- adrenoceptors results in vasoconstriction and hypertension (Leino et al., 2020).

However, the spinal administration of clonidine in dogs reveals a biphasic behavior of arterial blood pressure, starting with a hypertensive status due to vasoconstriction by an alpha2- agonist action, followed by a secondary hypotension mediated by baroreflex activity, thus increasing the hearts parasympathetic outflow (Leino et al., 2020). Although there is a possibility of differences in effects related to a different route of clonidine administration, we do not believe that biphasic phenomena occurred in CLG dogs, considering that no decrease was observed in HRV variables after clonidine administration, compared to CG, as expected in the initial vascular action of clonidine.

Nonetheless, as a study limitation, the initial hypertensive phase was not registered due to the necessity of anesthetic induction to obtain an invasive arterial blood pressure in the evaluated dogs. Future studies could elucidate this hypothesis of the biphasic behavior of arterial blood pressure in dogs receiving intramuscular clonidine and its association with a baroreflex mechanism on HRV. In addition to the dual effect of clonidine on arterial blood pressure, other possible effects include an increase in vascular resistance and arterial pressure produced by tramadol (Itami et al., 2011).

Many physiological components can also influence the arterial pressure modulation of clonidine as a compensatory baroreflex mechanism (Leino et al., 2020), modulation of postganglionic sympathetic cardiac nerves (Maze & Tranquilli, 1991), and a decrease in cardiac output. The methods of administration, dose, and the possible interactive effect with other drugs should also be considered (Cruz et al., 2011; Leino et al., 2020), as observed in HRV during tramadol administration. In the present study, the sample size could also be a limitation to the expected statistical differences in arterial pressure values.

Conclusion

In conclusion, an intramuscular dose of 5µg/kg of clonidine, as a pre-anesthetic medication in healthy dogs, associated with tramadol (analgesia), propofol (induction of anesthesia), and isoflurane (maintenance) increases the time domain HRV by blocking sympathetic outflow and increasing parasympathetic tonus, with minor effects on arterial blood pressure. This was associated with the occurrence of second-degree atrioventricular block (Mobitz II), accentuated by the autonomic synergic effect of tramadol. Although the pre-anesthetic use of clonidine in

dogs with fear-based behavioral problems should be considered, its association with tramadol is discouraged due to the evidence of cardiovascular risk.

Ethics statement

This study was approved by the Ethics Committee for Animal Use of the Federal University of Pampa (CEUA/Unipampa) under the protocol 004/2013.

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None.

Conflict of interests

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

Authors' contributions

FMO, JPEP, DJW and RT - Development of methodology; preparation and writing the initial draft. JPEP - Application of statistical study data, Review and Editing manuscript. FMO, JPEP and MAV - Writing, Review and Editing manuscript.

Availability of complementary results

The study was carried out at Hospital Universitário Veterinário of Universidade Federal do Pampa - UNIPAMPA, Uruguaiana, RS, Brasil.

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