Arq. Bras. Med. Vet. Zootec., v.65, n.5, p.1306-1312, 2013

Hematologic changes in propofol-anesthetized dogs with or without tramadol administration

[*Alterações hematológicas em cães anestesiados com infusão contínua de propofol, associado ou não ao tramadol*]

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ABSTRACT

Drugs commonly used in anesthesia practice may significantly alter the oxidative state of blood cells. This mechanism could contribute to the immune suppression that occurs transiently in the early postoperative period. Thus, we assessed the effects of continuous rate infusion (CRI) of propofol associated or not with tramadol on hematologic parameters in dogs. Eight adult mongrel dogs were anesthetized on 2 occasions, 15 d apart. Two groups were formed: control group (CG) and tramadol group (GT). Propofol was used for induction (10mg kg^{-1}) followed by a CRI (0.7mg $kg^{-1}minute^{-1}$). The animals were positioned in lateral recumbency and mechanically ventilated with inspired oxygen fraction of 0.6. In TG, tramadol (2mg kg^{-1}) followed by a CRI (0.5mg $kg^{-1}minute^{-1}$) was administered in dogs. In the CG the sodium chloride (NaCl) solution at 0.9% was administered followed by its CRI, in the same volume that was used in TG. The measurement was taken before anesthesia induction (Tbasal), 30 minutes after induction (T0) and then at 30-minute intervals (T30 to T60). Red blood cells, hematocrit, hemoblogin concentration and total leukocytes count decreased from T0 in both groups. In TG, lymphocytes count at Tbasal [1.86 (0.82) $x10^3\mu l^{-1}$] was greater than at T0, T30 and T60 [0.96(0.50), 0.92(0.48) and 0.95(0.48) $x10^3\mu l^{-1}$, respectively]. No significant differences were observed for platelets neutrophil, eosinophil, basophil and monocyte count. In dogs, propofol-anesthesia associated or not with tramadol promoted decrease in blood cell count and should be used with caution in immunossupressed patients.

Keywords: blood cells, hematology, opioid, total intravenous anesthesia.

RESUMO

Fármacos comumente utilizados na prática anestésica podem alterar significativamente o estado oxidativo das células sanguíneas. Esse mecanismo pode contribuir para a supressão imunológica que ocorre transitoriamente no pós-operatório imediato. Assim, foram avaliados os efeitos da infusão contínua (CRI) de propofol associado ou não com tramadol sobre parâmetros hematológicos em cães. Oito cães adultos foram anestesiados em duas ocasiões, com 15 dias de intervalo. Dois grupos foram formados: grupo-controle (CG) e grupo tramadol (TG). O propofol foi utilizada para a indução (10mg kg-1), seguido por CRI (0,7mg kg-1 minuto-1). Os animais foram posicionados em decúbito lateral e ventilados com fração inspirada de oxigênio de 0,6. Em TG, tramadol (2mg kg-1), seguido por CRI (0,5kg de 1 minuto-1mg), foi administrado em cães. Enquanto no CG, o cloreto de sódio solução (NaCl) a 0,9% foi administrado seguido por sua CRI, no mesmo volume que foi usado no TG. As mensurações das variáveis foram realizadas antes da indução anestésica (Tbasal), 30 minutos após a indução (T0) e em intervalos de 30 minutos (T30 a T60). Hemácias, hematócrito, hemoglobina e leucócitos totais diminuíram a partir de T0 em ambos os grupos. No TG, contagem de linfócitos no Tbasal [1,86 (0,82) x103μl-1] foi maior do que em T0, T30 e T60 [0,96 (0,50), 0,92 (0,48) e 0,95 (0,48) x103μl-1, respectivamente]. Não foram observadas diferenças significativas para plaquetas, neutrófilos, eosinófilos, basófilos e monócitos. Em cães, anestesia com propofol associado ou não ao tramadol promove alterações importantes no hemograma e deve ser utilizada com cautela em pacientes imunossuprimidos.

Palavras-chave: anestesia total intravenosa, células sanguíneas, hematologia, opioide

Recebido em 5 de junho de 2012
Aceito em 16 de abril de 2013
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INTRODUCTION

Drugs commonly used in anesthesia practice could significantly affect the oxidant-antioxidant milieu of immune cells such as peripheral blood lymphocytes. The consequent oxidative stress gives rise to cellular damage, including accelerated apoptosis, which is a main contributing factor for postoperative lymphocytopenia and immunological deficit (Delogu *et al*., 2004).

Several reports have demonstrated that propofol exhibits a perturbing action on the oxidant-antioxidant state of T cells (Delogu *et al*., 2004) and has some greater potential for the reduction of oxidative stress in erythrocyte membranes than midazolam (Tsuchiya *et al*., 2001) Hence, this intravenous anesthetic, characterized by a phenolic structure similar to α-tocopherol, provides an enhancement in antioxidant efficacies and erythrocytes protection against oxidative damage (Tsuchiya *et al*., 2002; Volti *et al*., 2006).

Many published studies have described using the combination of general anesthetic drugs with opioids to promote balanced anesthesia (Coetzee *et al*., 1996; Shipton *et al*., 2003). However, the use of opioids has been reported to possess some immunosuppressive effects (Yokota *et al*., 2000). In mice, in contrast to morphine, tramadol did not suppress cellular immune functions, whereas it increased natural killer activity, lymphocyte proliferation, and interleukin-2 production (Sacerdote *et al*., 1997).

We hypothesized that propofol and tramadol association does not possess significantly immunosuppressive effects. Thus, this study was designed to establish the effects of continuous rate infusion (CRI) of propofol associated or not with tramadol on hematologic parameters in dogs.

MATERIAL AND METHODS

This study was approved by the regional Institutional Ethics and Animal Welfare Committee (Ref: 014978-07). Eight adult mongrel dogs [16.6 (2.1) kg body weight], 4 males and 4 females, were enrolled in the study. All animals were determined to be healthy based on a complete physical examination, a blood cell count, standard serum biochemistry test, chest radiograph and electrocardiogram.

The dogs were anesthetized on 2 occasions, 15 d apart, and randomly assigned to one of two groups: CG (control group) and TG (Tramadol group).

Anesthesia was induced and maintained with propofol (10mg kg^{-1}; 0.7mg $kg^{-1}min^{-1}$, respectively). After endotracheal intubation, controlled ventilation (Excel 210IF, Datex-Ohmeda, Miami, USA) with inspired oxygen fraction (FiO_2) of 0.6 (Nunes *et al*. 2008), using pressure cycled (15 cm H_2O), began immediately and the respiratory rate, amplitude and an I:E 1:2 were adjusted to achieve an end-tidal carbon dioxide partial pressure ($PE´CO_2$) of 35 to 45 mm Hg. The oxygen concentration was determined with gas analysis equipment (Model DX-2010LCD; Dixtal, Manaus, Amazonas, Brazil).

Immediately, animals in the CG received 1 ml of 0.9% sodium chloride solution IV followed by a continuous rate infusion (1ml kg^{-1} $hour^{-1}$) using an infusion pump (Bomba de seringa AS50, SAMTRONIC®, São Paulo/SP, Brazil), while the TG dogs received 2mg kg^{-1} of tramadol followed by a continuous rate infusion (0.5mg $kg^{-1}hour^{-1}$). Tramadol was diluted in NaCl at 0.9% to obtain the same volume administered in CG dogs.

The dogs were positioned in dorsal recumbency on a heating pad (T/pump Localized Heat Therapy System - Model Tpp522 Gaymar Industries, Inc., Orchard Park, NY, USA) and a teflon catheter was placed in the right tarsal artery to monitor systolic (SAP), diastolic (DAP) and mean arterial pressures (MAP) using a multiparametric monitor (Dixtal mod. DX 2010, Invasive AP module, Manaus, AM, Brazil). During the entire procedure arterial pressure and heart rate (HR) were measured using a computerized electrocardiograph (TEB, mod. ECGPC software version 1.10, São Paulo, SP, Brazil) adjusted to lead II.

The bispectral index (BIS) was computed by an Aspect A-2000 monitor. The signal was acquired with electrodes (Sensor; Aspect Medical Systems, Inc) placed as described by Guerrero and Nunes (2003). Electrodes were placed before

the induction of anesthesia. Medium BIS values were recorded.

Next, a teflon catheter was placed percutaneously in the jugular vein to collect blood (3ml), which was collected into tubes containing ethylene-diamine-tetra-acetic acid (EDTA). The blood was submitted to laboratory evaluation (Abovet da ABX, abc Pach LMGE Montpellier, São Paulo, Brazil) to assess red blood cell count (He), hematocrit (Ht), hemoblogin concentration (Hb), platelets and total leukocytes count (Le). The differential count of leukocytes was made in blood smears stained with a mixture of May-Grunwald-Giemsa to assess basophils (Bas), eosinophils (Eos), segmented neutrophil (SN), band neutrophil (BN), lymphocytes (Lym) and monocytes (Mon).

The measurement of blood cell variables was taken before anesthesia induction (Tbasal), 30 minutes after induction (T0) and then at 30-minute intervals (T30 to T60). For BIS, HR, PAS, PAD and PAM, the first measurement was taken 30min after induction (T0) and then at 15-minute intervals for another 60 minutes (T15, T30, T45 and T60, respectively). BIS was also recorded before the induction of anesthesia (Tbasal).

Data are reported as means (standard deviation). Parametric data were subjected to one-way analysis of variance (ANOVA) to determine the difference between the different time points in the same group. Two-way ANOVA was used among groups. The Bonferroni test was used for post-hoc multiple comparisons at a α level of 0.05. Analyses were performed using Prism 5 for Windows (GraphPad Software Inc, CA, USA).

RESULTS

He, Ht, Hb and Le did not differ significantly between groups (Table 1), but from T0, means decreased in both groups. Lymphocytes did not differ between groups, but in TG, the mean at Tbasal was higher than T0, T30 and T60.

Table 1. Hematologic measurements in propofol-anesthetized dogs without (CG) or with tramadol (TG) administration. Data are expressed as means (SD). He, red blood cells count; Ht, hematocrit; Hb, hemoblogin concentration; Le, total leukocytes count; Bas, basophils; Eos, eosinophils; SN, segmented neutrophil; BN, band neutrophil; Lym, lymphocytes; Mon, monocytes. Means with different lower case letters within each row differ significantly from one another

Parameters	Group	Times				P-value (Groupx Time)	P-value (Time)
		Tbasal	T0	T30	T60		
He	CG	6.48(1.12)a	5.27(0.85)b	5.19(0.86)b	5.16(0.90)b		<0.05
(x 10^6 µL^{-1})	TG	6.20(1.00)a	5.01(0.71)b	5.00(0.70)b	4.98(0.76)b		<0.05
Ht	CG	45.4(8.0)a	36.5(5.7)b	35.9(6.0)b	35.7(6.2)b		<0.05
(%)	TG	43.7(7.5)a	34.9(4.9)b	34.8(4.8)b	34.6(5.3)b		<0.05
Hb	CG	14.6(2.5)a	12.0(1.9)b	12.1(1.7)b	12.0(1.8)b		<0.05
(g%)	TG	14.3(2.4)a	11.7(1.5)b	11.8(1.5)b	11.6(1.5)b		<0.05
Le	CG	6.48(1.12)a	5.27(0.85)b	5.19(0.86)b	5.16(0.90)b		<0.05
(x 10^6 µL^{-1})	TG	6.20(1.00)a	5.01(0.71)b	5.00(0.70)b	4.98(0.76)b		<0.05
Bas	CG	0(0)	0(0)	0(0)	0(0)		
(x 10^3 µL^{-1})	TG	0(0)	0(0)	0(0)	0(0)		
Eos	CG	1.19(2.50)	0.53(0.49)	0.46(0.49)	0.48(0.57)		
(x 10^3 µL^{-1})	TG	0.53(0.44)	0.48(0.52)	0.43(0.27)	0.57(0.39)		
BN	CG	0.12(0.15)	0.05(0.08)	0.09(0.09)	0.10(0.09)		
(x 10^3 µL^{-1})	TG	0.09(0.19)	0.06(0.08)	0.05(0.07)	0.09(0.15)		
SN	CG	5.65(1.42)	4.97(1.63)	5.18(1.82)	5.12(1.88)		
(x 10^3 µL^{-1})	TG	5.86(2.47)	5.74(3.40)	5.92(3.44)	5.69(3.11)		
Lym	CG	1.41(0.62)	1.38(0.58)	1.13(0.33)	1.17(0.45)		
(x 10^3 µL^{-1})	TG	1.86(0.82)a	0.96(0.50)b	0.92(0.48)b	0.95(0.48)b		<0.05
Mon	CG	0.28(0.21)	0.21(0.10)	0.18(0.08)	0.17(0.05)		
(x 10^3 µL^{-1})	TG	0.30(0.23)	0.25(0.14)	0.27(0.22)	0.17(0.07)		
Platelets	CG	295(100)	299(116)	305(107)	265(142)		
(x 10^3 µL^{-1})	TG	267(152)	266(131)	265(132)	270(122)		

The Bas, Eos, BN, SN, Mon and platelets values did not differ significantly between groups or among measurement times (Table 1).

The BIS value recorded before induction of anesthesia (Tbasal) was 96(2) for CG and 98(1) TG. BIS value at Tbasal was higher than at other times (T0, T15, T30, T45 and T60) (Figure 1).

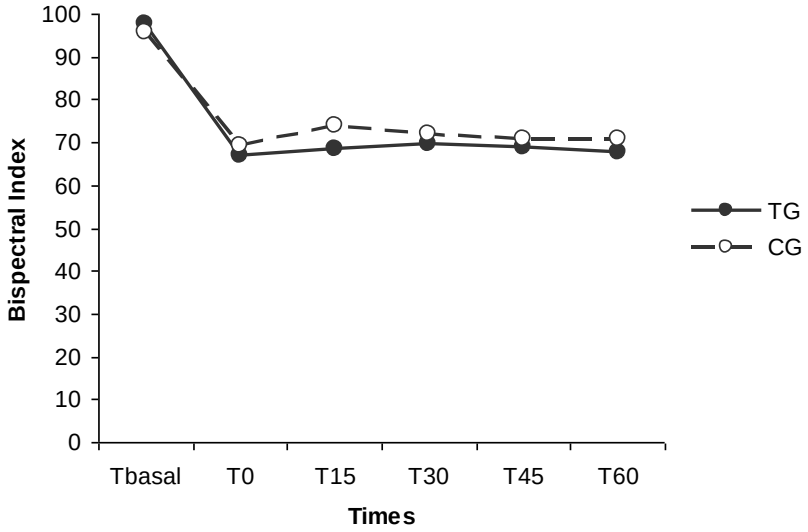

Figure 1. Bispectral index (BIS) monitoring. Black filled line and circles display the tramadol group (TG) and black dashed line and open circles represent the control group (CG). Values are presented as means (95% confidence intervals). In both groups, means at Tbasal [TG 98(1); CG 96(2)] were higher than values at T0 [TG 67(5); CG 69(7)], T15 [TG 68(6); CG 74(8)], T30[TG 70(6); CG 72(9)], T45[TG 69(7); CG 71(8)] and at T60 [TG 68(5); CG 71(9)].

HR, SAP, DAP and MAP did not differ significantly between groups (Table 2). In TG, PAS mean at T60 [126 (24) mmHg] was higher than values at T15 [109 (18) mmHg] and at T30 [112 (20) mmHg], while for DAP and MAP, values at T15 were lower than at T60 ($p<0.05$). No hypotension was observed.

Table 2. Cardiovascular measurements in propofol-anesthetized dogs without (CG) or with tramadol (TG) administration. Data are expressed as means (SD). HR, heart rate; SAP, systolic arterial pressure, DAP, diastolic arterial pressure; MAP, mean arterial pressure. Means with different lower case letters within each row differ significantly from one another

Parameters	Group	Times					P-value (Groupx Time)	P-value (Time)
		T0	T15	T30	T45	T60		
HR	CG	76(15)	72(14)	75(11)	76(16)	70(13)		
(beat minute^{-1})	TG	88(30)	88(23)	86(17)	85(21)	82(18)		
SAP	CG	110(19)	110(19)	117(23)	96(57)	127(22)		
(mmHg)	TG	119(27)	109(18)a	112(20)a	119(22)	126(24)b		<0.05
DAP	CG	60(8)	62(8)	65(11)	65(12)	68(10)		
(mmHg)	TG	66(13)	61(11)a	67(16)	67(13)	70(14)b		<0.05
MAP	CG	76(10)	77(10)	81(13)	81(15)	86(13)		
(mmHg)	TG	82(17)	76(13)a	81(16)	83(16)	88(17)b		<0.05

DISCUSSION

Regarding He, Ht and Hb, in both groups from T0, the means decreased but were within normal interval for species that present He from 5.5 to 8.5 x $10^6 \mu l^{-1}$, Hb from 12 to 18g dl^{-1} and Ht from 37 to 55% (Garcia-Navarro and Pachaly, 1994) (Table 1). As no differences between CG and TG were registered, we suggest that propofol, and not tramadol, promoted changes in these parameters. However, we believe that hemolysis did not occur because propofol provides an enhancement in antioxidant efficacies and erythrocytes protection against oxidative damage (Tsuchiya *et al*., 2002; Volti *et al*., 2006). In a study with propofol-anesthetized swine, the authors did not observe hemolysis and concluded that propofol antagonised the effects of forced peroxidation of red cells at anaesthetic and sub-anaesthetic concentrations (Ansley *et al*., 1998).

Thus, we suggested that the decrease in He, Ht and Hb occurred due to sequestration of red blood cells in nonsplenic sites. This hypothesis can be confirmed by studies in dogs which demonstrated that propofol does not cause measurable splenic enlargement (O'Brein *et al*., 2004; Wilson *et al*., 2004). Besides, Wilson *et al*. (2004) described a lack of correlation between hematocrit and spleen size following the anesthetic protocols with propofol, suggesting sequestration of red blood cells in nonsplenic sites.

In both groups, Le decreased from T0 (Table 1), but the means were within normal intervals for the species (Garcia-Navarro and Pachaly, 1994). The immune system is under elaborate control of the neuroendocrine stress response, which is affected by the anesthetic plane (Naito *et al*., 1992; Crozier *et al*., 1994; McBride *et al*., 1996), or side effects of anesthetics, such as systemic hypotension (Heesen *et al*., 1995). Thus, in both groups, the difference observed between Tbasal and other times can be justified by the animals' consciousness at Tbasal. After the beginning of continuous infusion of drugs this parameter, as well as arterial pressures and BIS were stable and maintained the stability of white blood cell counts.

Some drugs used in anesthesia induction and maintenance impaired free radical function (Naito *et al*., 1992). This reactive oxygen species (ROS) have been implicated in apoptosis (Delogu *et al*., 2004) that is an active form of cell death (Bauer *et al*., 1998). Additionally, catecholamines and glucocorticoids, released during anesthesia, impaired the immune system (Elenkov and Chrousos, 2002) because the lymphocyte production and function decreased (Inada *et al*., 2004). However, we suggested that these factors did not influence white blood cell counts because propofol provides an enhancement in antioxidant efficacies (Murphy *et al*., 1992; Demiryurek *et al*., 1998) and promotes lower release of catecholamines (Elenkov and Chrousos, 2002).

For Lym no differences were registered between groups (Table 1), but in TG Tbasal showed higher means than other times. Opioids may suppress immune system cells, such as leukocytes and lymphocytes, via an indirect mechanism operating through the central nervous system (CNS) (Chang, 1984; Molina, 2006). This CNS influence occurs by regulating the systemic concentration of humoral substances such as cortisol and epinephrine (Straub *et al*., 1998). Lymphoid organs contain a rich supply of sympathetic nerve fibers (Felten *et al*., 1987) permitting norepinephrine (NE) to influence lymphocyte activity directly (Straub *et al*., 1998). Tramadol inhibits neuronal reuptake of NE and 5-hydroxytryptamine (5-HT), and may actually facilitate 5-HT release (Lamont and Mathews, 2007). Thus, we believe that the NE concentration increased promoting the suppression of Lym.

In humans, the acute administration of 20 and 40mg kg^{-1} of racemic tramadol and of 10 and 20mg kg^{-1} of (+) tramadol induced lymphoproliferation, which was attributed to the enhancement of the serotoninergic tone (Sacerdote *et al*., 1999). The difference between Sacerdote *et al*.'s study and ours can be attributed to the use of continuous infusion of tramadol.

Regarding Bas, Eos, SN, BN, Mon and platelets, propofol associated or not with tramadol promoted the stability of these parameters.

CONCLUSION

In conclusion, in dogs the propofol-anesthesia associated or not with tramadol promoted a decrease in blood cell count and should be used with caution in immunossupressed patients.

REFERENCES

ANSLEY, D.M.; LEE, J.; GODIN, D.V. *et al*. Propofol enhances red cell antioxidant capacity in swines and humans. *Can. J. Anaesth.*, v.45, p.233-239, 1998.

BAUER, M.K.A.; VOGT, M.; LOS, M. *et al*. Role of reactive oxygen intermediates in activation-induced CD95 (Apo1-Fas) ligand expression. *J. Biol. Chem.*, v.273, p.8048-8055, 1998.

CHANG, K.J. Opioid peptides have actions on the immune system. *Trends Neurosci.*, v.7, p.234-235, 1984.

COETZEE, J.F.; MARITZ, J.S.; DU TOIT, J.C. Effect of tramadol on depth of anaesthesia. *Br. J. Anaesth.*, v.76, p.415-418, 1996.

CROZIER, T.A.; MULLER, J.E.; QUITTKAT, D. *et al*. Effect of anaesthesia on the cytokine responses to abdominal surgery. *Br. J. Anaesth.*, v.72, p.280-285,1994.

DELOGU, G.; ANTONUCCI, A.; MORETTI, S. *et al*. Oxidative stress and mitochondrial glutathione in human lymphocytes exposed to clinically relevant anesthetic drug concentrations. *J. Clin. Anesth.* v.16, p.189–194, 2004.

DEMIRYUREK, A.T.; CINEL, J.; KAHRAMAN, S. *et al*. Propofol and intralipid interact with reative oxygen spesies: a chemiluminescence study. *Br. J. Anaesth.*, v.80, p.649-654, 1998.

ELENKOV, I.J.; CHROUSOS, G.P. Stress hormones, proinflammatory and anti-inflammatory cytokines, and autoimmunity. *Annals N. Y. Acad. Sci*., v.966, p.290-303, 2002.

FELTEN, D.L.; FELTEN, S.Y.; BELLINGER, D.L. *et al.* Noradrenergic sympathetic neural interactions with the immune system: Structure and function. *Immunol. Rev.*, v.100, p.225–260, 1987.

GARCIA-NAVARRO, C.E.K.; PACHALY, J.R. *Manual de Hematologia Veterinária*, São Paulo: Livraria Varela Ltda, 1994. 169 p.

GUERRERO, P.N.H.; NUNES, N. Monitoramento do índice bispectral em cães. *Semina*, v.24, p.163-170, 2003.

HEESEN, M.; BACHMANN-MENNENGA, B.; ZEILER, D. *et al*. Increase of Interleukin-6 plasma concentration and HLA-DR positive T-lymphocytes after hypotensive anaesthesia with sodium nitroprusside. *Acta Anaesthesiol. Scand.*, v.39, p.965-969, 1995.

INADA, T.; YAMANOUCHI, Y.; JOMURA, S. *et al*. Effect of propofol and isoflurane anaesthesia on the immune response to surgery. *Anaesthesia*, v.9, p.954-959, 2004.

LAMONT, L.A.; MATHEWS, K.A. Opioids, nonsteriodal anti-inflammatories and analgesic adjuvants. In: TRANQUILLI, W.J.; THURMON, J.C.; GRIMM, K.A. eds. *Lumb & Jones' Veterinary anesthesia and analgesia.* 4.ed. Oxford: Blackwell Publising, 2007. p.241-271.

MCBRIDE, W.T.; ARMSTRONG, M.A.; MCBRIDE, S.J. Immunomodulation: An important concept in modern anaesthesia. *Anaesthesia*, v.51, p.465-473, 1996.

MOLINA, P.E. Opioids and opiates: analgesia with cardiovacular haemodynamic and immune implications en critical illness. *J. Intern. Medic.*, v.259, p.138-154, 2006.

MURPHY, P.G.; MYERS, D.S.; DAVIES, M.J. *et al*. The antioxidant potential of propofol (2,6-diisopropylphenol). *Br. J. Anaesth.* v.68, p.613-618, 1992.

NAITO, Y.; TAMAI, S.; SHINGU, K. *et al*. Responses of plasma adrenocorticotropic hormone, cortisol, and cytokines during and after upper abdominal surgery. *Anesthesiology*, v.77, p.426-431, 1992.

NUNES, N.; LOPES, P.C.F.; SANTOS, P.S.P. *et al*. Hemodyinamic effects of several inspired oxygen fractions in spontaneously breathing dogs submitted to continuous infusion of propofol. *Cien. Rural*, v.38, p.729-735, 2008.

O'BREIN, R.T.; WALLER, K.R.; OSGOOD, T.L. Sonographic features of drug-induced splenic congestion. *Vet. Radiol. Ultras.*, v.45, p.225-227, 2004.

SACERDOTE, P.; BIANCHI, M.; MANFREDI, B. *et al*. Effects of tramadol on immune responses and nociceptive thresholds in mice. *Pain*, v.72, p.325-330, 1997.

SACERDOTE, P.; BIANCHI, M.; GASPANI, L. *et al*. Effects of tramadol and its enatiomers on concavalin-A induced-proliferation and NK activity of mouse splenocytes: involvment of serotonin. *Int. J. Immunopharmacol.*, v.21, p.727-734, 1999.

SHIPTON, E.A.; ROELOFSE, J.A.; BLIGNAUT, R.J. An evaluation of analgesic efficacy and clinical acceptability of intravenous tramadol as an adjunct to propofol sedation for third molar. *Surgery Anesth. Prog.*, v.50, p.121-128, 2003.

STRAUB, R.H.; WESTERMANN, J.; SCHÖLMERICH, J. *et al*. Dialogue between the CNS and the immune system im lymphoid organs. *Immunol. Today*, v.19, p.409-413, 1989.

TSUCHIYA, M.; ASADA, A.; MAEDA, K. *et al*. Propofol versus midazolam regarding their antioxidant activities. *Am. J. Respir. Crit. Care Med.*, v.163, p.26-31, 2001.

TSUCHIYA, M.; ASADA, A.; KASAHARA, E. *et al*. Antioxidant protection of propofol and its recycling in erythrocyte membranes. *Am. J. Respir. Crit. Care Med.*, v.65, p.54-60, 2002.

VOLTI, G.L.; MURABITO, P.; ATTAGUILE, G. *et al*. Antioxidant properties of propofol when oxidative stress sleeps with patients. *Excli. J.*, v.5, p.25-32, 2006.

WILSON, D.V.; EVANS, A.T.; CARPENTER, R.E. The effect of four anesthetic protocols on splenic size in dogs. *Vet. Anaesth. Analg.*, v.31, p.102-108, 2004.

YOKOTA, T.; UEHARA, K.; NOMOTO, Y. Intrathecal morphine suppresses NK cell activity following abdominal surgery. *Can. J. Anaesth.*, v.47, p.303-308, 2000.