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Evaluation of Cortisol Levels in Female Dogs Undergoing Ovariosalpingohysterectomy after Autohemoteraphy

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Abstract **– Stressful and painful conditions may change the secretion of pituitary hormones that directly regulate functions related to animal welfare. The therapeutic effects of blood have been partially attributed to platelets, which are important for tissue repair. The use of tramadol in hospitals has been important in animals because it may reduce physiological stress. Thirty clinically healthy female dogs were randomly assigned to three groups. Rectal temperature, heart rate, respiratory rate and serum cortisol were measured at five moments. Ovariosalpingohisterectomy was adopted as the surgical technique. Serum cortisol levels were similar at all moments among the groups and did not increase. The application of tramadol and the use of blood cells in female dogs submitted to surgery did not alter the physiological variables during pre and postoperative periods. Besides, blood cells could be used as therapeutic alternative in tissue repair.**

Keywords **– Cortisol, Canine, Surgery, Ovariosalpingohisterectomy.**

I. Introduction

Stressful and painful conditions may change the secretion of pituitary hormones that directly regulate functions related to animal welfare such as reproduction, growth and immune resistance. Adrenocorticotropic (ACTH), under stress conditions, stimulates the secretion and release of glucocorticoids in blood [01], [02]. The use of tramadol in hospitals has been important in animals because it may reduce physiological stress [03].

After activation by pharmacological or physiological agents, the therapeutic effects of blood have been partially attributed to platelets, which rapidly reach the wound and release multiple growth factors (GF) and cytokines which are important in tissue repair due to mitogenic, chemotactic and neovascular actions [04].

On the other hand, the interactions of those cells with physiological variables during surgeries still require more studies. Due to their potential of differentiation and reduction of the inflammatory process, they can improve the cicatrization process, acting as coadjuvant of postoperative analgesia [05], [06].

Reversible anesthetic complications may occur during OSH, where the most common clinical signs in anesthetized dogs are: decrease in rectal temperature, increase in respiratory rate with or without acidosis, tachycardia and muscle chills. These complications can be avoided by the use of alkaline electrolytes substances and oxygen during the trans-operative and immediate post-operative period [07].

The aim of this study was to evaluate the physiological variables during pre and postoperative period of female dogs submitted to ovariosalpingohisterectomy after application of autologous spinal cord blood.

II. Materials and Methods

After approval by the Ethics Committee on Animal Experimentation of the State University of Maranhao, thirty clinically healthy female dogs were selected from the casuistry of the University Veterinary Hospital.

Animals were randomly assigned to three groups: GI (Control), GII (Conventional) and GIII (Experimental). Clinical examination, complete blood count, hemoparasites research and indirect immunofluorescence reaction (IIFR) for leishmaniasis, with rectal temperature (RT), heart rate (HR), respiratory rate (RR) and serum cortisol dosage (SC) were performed by chemiluminescence technique, in five moments: M_0 (before surgical procedures), M_1 (six hours after surgery), M_2 (48h), M_3 (120h) and M_4 (168h) according to protocol used by [02], with some modifications.

Acepromazine maleate 1% was used intravenously as preanesthetic medication at a dose of 0.1mg/kg. In GI and GIII, preemptive analgesia with tramadol hydrochloride was applied intravenously at a dose of 2mg/kg; and in GII, antibiotic chemoprophylaxis with benzathine penicillin was applied intramuscularly at a dose of 40,000 IU / kg.

Ketamine (5mg/kg) with diazepam (0.5mg/kg) were used intravenously at the anesthetic induction and then intubated and maintained with sevoflurane in 100% oxygen. In the experimental group, moments before surgery, 10 ml of medullary blood was collected from each animal from the proximal humeral metaphysis and immediately applied to biceps femoris muscle.

The animals were maintained in venoclysis with lactated Ringer's solution, 10 ml / kg / hour during the trans and postoperative period until the return of physiological parameters and anesthesia [07]. The surgical technique adopted for OSH was the traditional one [08].

The results were submitted to analysis of variance (ANOVA) followed by Student Newman Keuls test (SNK) to compare means of different observation moments

within the same group and among groups, with a significance level of 5% ($p < 0.05$).

III. Results

The anesthetic protocol used for all animals, the analgesia proposed in the preoperative period for GI, GIII (Mastrocinque and Fantoni, 2003) and antibiotic for GII were adequate. All animals recovered without complications during trans and postoperative.

RT, RC and FR (Tables 1 and 2) presented similar behavior throughout the evaluation period, with no significant differences among the groups and among moments within the groups.

Table 1 - Means and standard deviations of rectal temperature (ºC) and heart rate (beats per minute), distributed in groups and moments of female dogs submitted to OSH, means followed by different letters, upper case in the same row and lower case in the same column, do not differ from each other.

Moments	GI	GII	GIII
M0	38,72 ± 0,47Aa	38,56 ± 0,68Aa	38,57 ± 0,52Aa
	107,50 ± 24,68Aa	112,00 ± 30,39Aa	115,40 ± 20,61Aa
M1	38,78 ± 0,60Aa	38,75 ± 0,68Aa	38,35 ± 0,74Aa
	104,20 ± 17,99Aa	111,20 ± 13,70Aa	99,40 ± 11,85Aa
M2	38,63 ± 0,25Aa	38,74 ± 0,61Aa	38,76 ± 0,44Aa
	107,20 ± 16,41Aa	102,40 ± 24,08Aa	102,40 ± 8,88Aa
M3	38,75 ± 0,39Aa	38,74 ± 0,61Aa	38,75 ± 0,39Aa
	96,60 ± 11,15Aa	109,60 ± 23,94Aa	101,60 ± 10,90Aa
M4	38,71 ± 0,48Aa	38,63 ± 0,51Aa	38,52 ± 0,34Aa
	97,80 ± 97,80Aa	98,80 ± 19,23Aa	104,00 ± 11,92Aa
Overall Mean	38,69 ± 0,43	38,69 ± 0,58	38,69 ± 0,43
	102,66 ± 17,60	106,80 ± 22,62	104,56 ± 12,83

Means of SC (Table 2) showed significant differences among groups and among moments within the groups. GII showed the highest overall mean. Regarding the moments, M_1 presented the highest mean of the three groups, presenting statistical differences among them. There was an increase in values in all groups, from M_0 to M_1, in which there was a significant difference between these two moments. The GIII group, despite increasing the CS value in M1, showed no significant difference between the two moments of this group.

Table 2 - Means and standard deviations of respiratory rate (movements per minute) and serum cortisol dosage (µg / dl) of female dogs submitted to OSH. Means followed by different letters, upper case in the same row and lower case in the same column, do not differ from each other.

Moments	GI	GII	GIII
M_0	54,60 ± 12,96^{Aa}	55,40 ± 26,39^{Aa}	49,60 ± 21,76^{Aa}
	3,87 ± 1,87^{Aab}	4,03 ± 1,59^{Aa}	3,41 ± 1,66^{Aab}
M_1	35,20 ± 11,12^{Aa}	43,60 ±16,91^{Aa}	31,00 ± 8,39^{Aa}
	4,67 ±1,33^{Ab}	7,14 ± 1,93^{Bb}	5,58 ± 1,39^{ABb}
M_2	45,80 ± 16,66^{Aa}	49,20 ±16,65^{Aa}	48,00 ± 17,88^{Aa}
	2,96 ± 1,38^{Aab}	3,56 ± 2,37^{Aa}	3,88 ± 1,41^{Aab}
M_3	45,20 ± 11,47^{Aa}	44,00 ±17,48^{Aa}	46,40 ±14,00^{Aa}
	2,02 ± 0,95^{Aa}	2,94 ± 0,99^{Aa}	2,42 ± 1,30^{Aa}
M_4	40,20 ± 14,86^{Aa}	45,40 ± 12,65^{Aa}	42,80 ± 16,22^{Aa}
	2,49 ± 1,20^{Aab}	3,07 ± 1,17^{Aa}	2,17 ± 1,31^{Aa}
Overall Mean	44,2 ± 14,56	47,52 ± 18,01	43,56 ± 15,65
	3,27 ± 1,64	4,12 ± 2,22	3,48 ± 1,80

IV. Discussion

Normal rectal temperature for canine species ranges from 37.9 to 39.9ºC [09]. In the overall mean, heart rate was different among groups. These values can range from 70 to 120 beats per minute in healthy dogs [10]. In the overall mean of groups, respiratory rate was higher in GII, proving to be a more stressful procedure, whose normal values during resting are 20 to 34 respiratory movements per minute [10], respectively, had similar behavior between the groups and between the moments within the groups, demonstrating that the treatments used were the same.

Non-opioid mechanisms of tramadol may potentiate analgesia without causing alteration of RT, HR and RR [05], [06] [11] as it is observed in other opioids [12], [13], [14].

Serum cortisol levels were similar at all moments between GI and GIII. Therefore, there was no increase. On the other hand, there was a significant difference between the moments (M <sub> 1 </ sub>) within the GII group, after six hours of surgery when the effects of anesthesia ceased, where no analgesic was used and attributed to increased stress the reference values for canine species vary from 0.5-6.0 µg / dl [15]. Other studies show that female dogs submitted to OSH presented a significant increase in cortisol due to the pain arising from the surgical procedure [01], [02].

High levels of SC in pretreatment, anesthetic induction and surgery were not observed (Table 2). It may indicate that the management of animals, including physical restriction, venipuncture and surgery, did not interfere in neuroendocrine and metabolic function, as observed by other authors [01], [02].

In GIII, using blood cells, it is known that platelets arrive quickly to the wound and release multiple growth factors and cytokines [04], favoring the cicatrization process. In the present study, it was verified that the use of medullary blood associated with tramadol attenuated the increase of SC, similar to what was observed in GI, in which only tramadol was used.

V. Conclusions

The application of tramadol and blood cells in female dogs submitted to OSH did not alter the physiological variables during pre and postoperative periods. Besides, blood cells could be used as therapeutic alternative in tissue repair.

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