

RESEARCH ARTICLE

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Effect of pregnancy in hematological profile of dogs

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Abstract

Pregnant impact on hematological parameters in the dogs was award in this study. Blood was collected in the cephalic vein of 32 female animals, clinically healthy, in different physiological condition. Hematological parameters that studied were RBC, WBC and platelets count, Hgb concentration, HCT, differential counting of leukocytes and erythrocyte indicators. The obtained results from the analysis showed that pregnancy can affect the values of hematological parameters. The number of erythrocytes, the level of hematocrit and hemoglobin concentration in pregnant animals resulted lower ($P < 0.05$) than in non-pregnant animals. No volatility resulted in the values of MCV, MCH and MCHC. In the pregnant animals group was found the increase of the circulating leucocytes number, of relative value of neutrophils and lymphocytes decrease. The pregnancy condition should be taken in consideration during the results interpretation of laboratory analysis.

Keywords: RBC, WBC, hematocrit, hemoglobin, pregnant, dog.

1. Introduction

Clinical problems and their monitoring and treatment effectiveness necessarily require laboratory examinations. The analyze and correct interpretation of the laboratory tests results provide adequate information for assessing of conditions of breeding, feeding, stress conditions, non-specific resistance, etc., [18]. They also serve to monitor specific therapeutic protocols and to determine prognosis. At the same time, they are the basis of experimental and scientific research. However, the basis of these parameters must be stabilized before the data can be interpreted and applied.

Blood parameters vary in different physiological stages, as well as in pathological conditions [5]. Pregnancy is one of the physiological conditions that lead significant changes in hematological and biochemical parameters of all animal species. During pregnancy some blood components are subject to significant changes, confusing the correct interpretation of blood parameters in diseases or disorders that may occur during this period [2]. In order to prevent metabolic disorders or subclinical disease during the pregnancy, it is necessary to define the physiological fluctuations of hematological and biochemical parameters of blood in healthy bitches and carried out systematic monitoring of these parameters in different stages of pregnancy [17].

2. Material and methods:

To study the influence of pregnancy in blood parameters of dogs, 32 healthy bitches were divided

into two groups. The first group was represented by 16 non-pregnant bitches and the second group of 16 bitches that were in the last stage of pregnancy. The females of the second group had similar body weight and age with them of the first group.

The samples were collected from September 2010 to February 2012. Clinically healthy status was based on these criteria: no visible signs of illness were observed, while temperature, pulse and respiratory rate were normal. All of the dogs were clinically examined and none of these dogs receive any treatment before. To determine the condition of pregnancy was based on the date of copulation and on manual methods to control it.

Blood samples from 8-h fasted dogs (early in the morning) were taken from the cephalic vein. Venous blood was gathered in tubes with anticoagulant (1.6 mg K3-EDTA/ml blood). Samples were kept at room temperature ($18 - 25^{\circ}\text{C}$) and tests were performed within 8 hours after blood was collected. The evaluation of red blood cells (RBC) and white blood cells (WBC) count for $1\mu\text{l}$ blood was carried out manually with hemocytometer method. Hematocrit (HCT) was determined by the method of microhematocrit with capillary tubes, according to Alexander & Griffiths (1993). The determination of hemoglobin was performed cyanmethemoglobin method [1] and the values were read in spectrophotometers with 540 nm wavelength. Erythrocyte indices, mean cells volume average, mean cells hemoglobin and mean cells hemoglobin concentration was calculated with the formulas described by Jain [8]. The leucogram was designed

through the smears prepared with fresh blood, after May-Grunwald-Giemsa laboratory staining method and reading by immersion objective was performed [11]. For statistical processing of data was used SPSS Inc program.16.0. For different parameters measured by the above analysis it was determined the average \pm δ , minimum and maximum values of fluctuation, the truthfulness of change averages two populations (t_D). Results were evaluated statistically using the Student test, t-test and $P < 0.05$ were considered significant.

3. Results and discussion:

From the results obtained were observed fluctuation of blood values in female animals, in the second month of pregnancy. Hematological parameters such as number of RBC and PLT, HCT and Hgb values were lower ($P < 0.05$) in pregnant animals, than the values of non-pregnant ones.

Table 1. Values of hematological parameters (mean \pm sd) in bitches

Parameters	RBC ($\times 10^6/\mu\text{l}$)	PLT ($\times 10^5/\mu\text{l}$)	HCT (%)	Hgb (g/dl)	MCV (fl)	MCHC (%)
Non-pregnant animals	6.4 \pm 0.76 ^a	3.4 \pm 0.97 ^b	45.4 \pm 3.6 ^c	15.2 \pm 1.4 ^d	69.9 \pm 4.6	33.5 \pm 1.9
Pregnant animals	5.8 \pm 0.6 ^a	2.8 \pm 1.06 ^b	41 \pm 4.9 ^c	13.9 \pm 1.6 ^d	70.7 \pm 4.5	33.9 \pm 1.5
Reference intervals for normal dogs	5.5-8.5	2-5	37-55	12-18	60-77	32-36

Statistical difference between the average values of groups ^{a,b,c,d} $P < 0:05$

Table 2. Changing the averages (t_D) of hematological indices according to physiological status

Hematological parameters	RBC	PTL	HCT	Hgb	MCV	MCHC
t_D non-pregnant/ pregnant	2.151*	2.092*	2.371*	2.126*	-0.374	0.000

$P < 0:05$ *

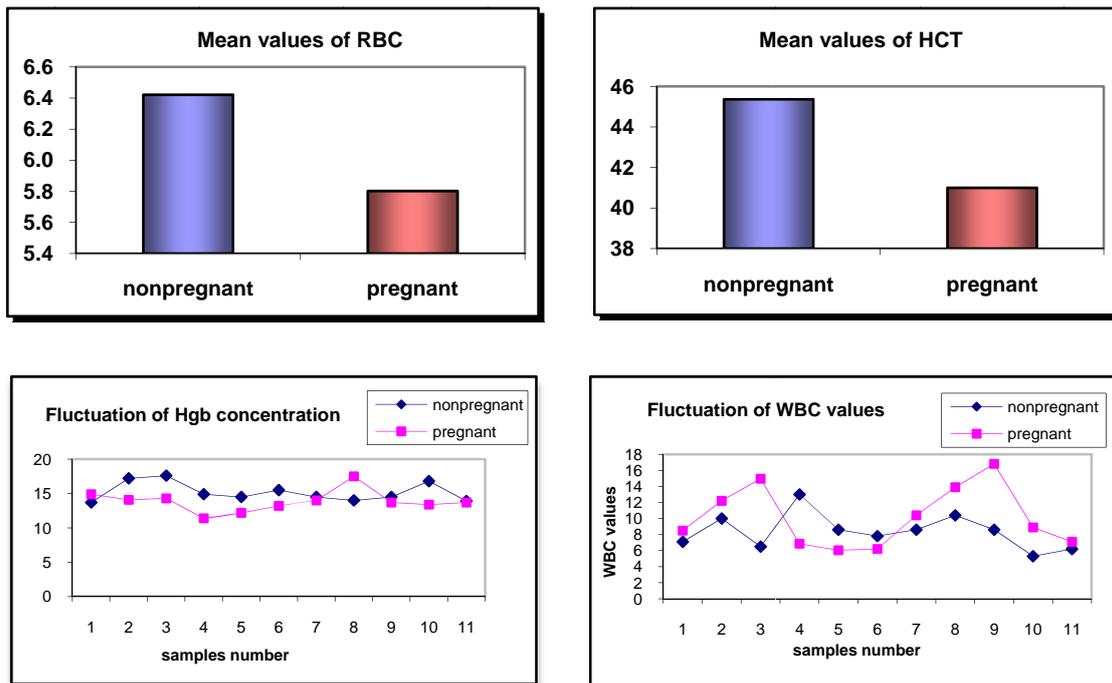


Figure. 1. Diagrams of the fluctuation of the values between the two groups

We found no significant fluctuation in the values of MCV, MCH and MCHC. As it is reflected in table 1, the average number of red blood cells (RBC) was reduced from $6.4 \times 10^6/\mu\text{l}$ in non-pregnant female dogs to $5.8 \times 10^6/\mu\text{l}$ in pregnant animals, platelets count was decreased from $3.4 \times 10^5/\mu\text{l}$ to $2.8 \times 10^5/\mu\text{l}$, hemoglobin level from 15.2 g/dl to 13.9 g/dl and

hematocrit from 45.4% to 41%. Similar findings were found by some authors [4, 8, 10 and 12].

The decrease of RBC, PLT, HCT and Hgb values in female animals in their last stage pregnancy is explained by the fact that in pregnant animals occur hemodilution and a slight anemia as a consequence of increased plasma volume. In addition, a shortening of the erythrocytes life in pregnant animals is another

cause of the reduction of red blood cells number [5, 16]. While, other authors [7, 8, 9] report increased levels of these indicators in pregnant cattle.

According to statistical processing of the data the number of circulating leucocytes in pregnant animals was high. The number of white blood cells in the average value was increased from 8.4×10^3 to $10.2 \times 10^3/\mu\text{L}$ blood, while neutrophils were increased and lymphocytes were decreased. This change is comparable to that reported by Coles (1986). According to some authors, it is expected a physiological increase of leucocytes and mostly

granulocytes neutrophil in female animals in the last stage of pregnancy [9]. This increase can be attributed to a number of reasons, such as the secretion of estrogen, a higher level of cortisol in plasma, or maternal immune system response to fetal allograph [2]. The increase of granulocytes neutrophil during the recent weeks of pregnancy may reflect the numerous neutrophils aggregation in placental blood vessels, close to the degenerative and necrotic cells [14]. On the contrary, some authors [5] have observed the reduction of leucocytes number in pregnant animals because of immunosuppression in young bitch.

Table 3. Values of leukocytes parameters (mean \pm sd) in female dogs

Leukocytes parameters	WBC		Leukograma (%)			
	($\times 10^3/\mu\text{L}$)	Lymphocytes	Monocytes	Eosinophils	Basophils	Neutrophils
Non-pregnant	8.4 \pm 2.2	20.8 \pm 4.2	5.0 \pm 1.3	4.5 \pm 1.2	0.4 \pm 0.5	69.3 \pm 3.1 ^a
Pregnant	10.2 \pm 3.8	17.4 \pm 4.1	5.1 \pm 1.9	3.9 \pm 1.8	0.2 \pm 0.4	73.5 \pm 5.8 ^a
Reference intervals for dogs	6-17	12-30	3-9	2-10	0-0.5	60-75

Statistical difference between the average values of two groups ^aP<0.05

Table 4. Changing the averages (t_D) indices according to physiological status

Leukocytes indices	WBC	L	M	E	B	N
t_D non-pregnant/ pregnant	-1.366	1.970	-0.131	0.988	0.933	-2.113*

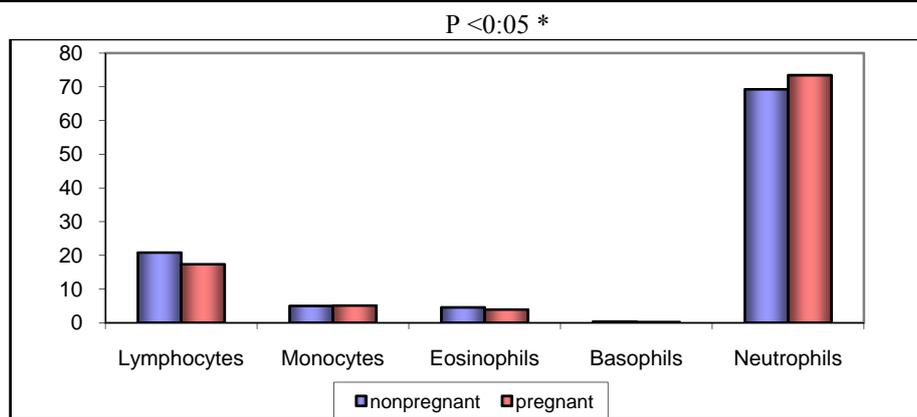


Figure 2. Diagram of differential leucocytes count in both groups of animals

Female animals in the last month of pregnancy showed different neutrophils/lymphocytes ratio, compared with non-pregnant group, because the relative values of lymphocytes decrease while neutrophils increase at this stage of pregnancy (Figure 2). In fact, the absolute number of lymphocytes remains constant, but while the number of white blood cells increases, the percentage (relative value) of lymphocytes decreases. On the other hand, the reduction of lymphocyte values during pregnancy can be attributed to the physiological stress leading to lymphopenia [3, 8]. However, this finding is different of that reported by some authors [4], who observed increased lymphocytes during the last period of pregnancy.

In conclusion, statistical tests for the difference of average values of the parameters of red blood cells between the pregnant and non-pregnant animals showed significant difference (P < 0.05) only for RBC, HCT and Hgb. Statistical tests for the change of average values of white blood cells were not significant, excepted of the relative number of neutrophils.

The female dogs at the last pregnancy stage exhibit fluctuations of hematological parameters, however, these fluctuations remain within the limit of reference values. The pregnancy condition should be taken in consideration during the results interpretation of laboratory analysis.

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