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## Studies on the effect of blood transfusion on haematological alterations and reticulocyte count in anaemic dogs

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### Abstract

The present research focuses on etiopathology, haematological alterations and reticulocytic changes in Twenty anaemic dogs having proven etiology as pyometra, CKD and *B canis* infection and presented to Dept of Vet. Medicine, Mumbai Veterinary College. The aim of study was to evaluate efficacy of transfusion of blood and blood components at 24, 48, 72 hrs and on the 7th day. The assessment of anaemic cases was based on clinical and physical examination and laboratory evaluation of blood. The highest prevalence of anaemia in present study was noted in Labrador (45%), mixed breeds (35%), and Non-Descript (20%) dogs. CKD and *B canis*-originated anaemia cases revealed the highest prevalence of anaemia in males (74.99%) as compared to females (25%). There were highly significant differences observed between the Hb and RBC values before treatment and 24 hours and between before-treatment values and 7 days post-transfusion ( $p<0.01$ ). 40% cases were recorded as microcytic hypochromic anaemia and normocytic and normochromic each, whereas macrocytic hypochromic anaemia cases were 20%. There was a significant increase ( $p<0.01$ ) in the leucocyte count after 24 hours when compared to pre-transfusion values.

**Keywords:** Canine, blood transfusion, erythrocyte, pyometra, CKD, *B. canis*

### Introduction

Anaemia in CKD dogs is mainly related to the decreased production of erythropoietin as well as other mechanisms, such as inflammation, iron deficiency or inappropriate utilization, decreased red blood cell (RBC) survival due to metabolic or mechanical injuries, and the antiproliferative effects of uremic toxins<sup>[1]</sup>. The anaemia present in pyometra occurs due to the migration of RBCs into the uterus by diapedesis in a chronic process or due to bone marrow suppression as a result of endotoxins from systemic bacterial proliferation, which also causes leukocytosis. Babesia parasites infect and destroy red blood cells, babesiosis can cause a special type of anaemia called hemolytic anemia. Blood transfusion is indicated in all conditions in which there is acute, subacute or chronic loss of blood and is a supportive measure while the underlying cause of the anaemia is receiving specific treatment<sup>[2]</sup>. The timely performance of blood transfusion is a life-saving measure. Early classification of anaemia as regenerative versus non-regenerative is an imperative diagnostic step guiding further clinical evaluation and treatment of anaemic dogs<sup>[3]</sup>.

### Materials and Methods

The present study was carried out on dogs referred to the Department of Veterinary Clinical Medicine, Ethics and Jurisprudence, Mumbai Veterinary College, Parel and the dogs presented at the medical ward of Bai Sakarabai Dinshaw Petit Hospital for Animals (BSDPHA) affiliated to Mumbai Veterinary College, Parel, Mumbai- 400012 from March 2021 to September 2023. The study included 20 (Twenty) anaemic dogs having confirmed pyometra, Chronic Kidney Disease (CKD) and *B canis* (Table 1).

**Table 1:** Breed, Age, Sex, Disease Diagnosis and type of anaemia in the anaemic dogs used in the study (n=20)

Sr. No	Breed	Sex	Age (Years)	Disease	Type of anaemia
1	Labrador	Female	8	Pyometra	Microcytic Hypochromic Anaemia
2	Labrador	Female	5	CKD	Normocytic normochromic Anaemia
3	Goden Retriever	Male	2	<i>B canis</i>	Macrocytic Hypochromic Anaemia
4	Labrador	Female	7	Pyometra	Microcytic Hypochromic Anaemia
5	Labrador	Male	5	CKD	Normocytic normochromic Anaemia
6	Pug	Female	8	Pyometra	Microcytic Hypochromic Anaemia
7	German Shepherd	Female	7	Pyometra	Microcytic Hypochromic Anaemia
8	Labrador	Male	6	CKD	Normocytic normochromic Anaemia
9	Sptiz	Female	8	Pyometra	Microcytic Hypochromic Anaemia
10	Labrador	Male	5	CKD	Normocytic normochromic Anaemia
11	Labrador	Female	9	Pyometra	Microcytic Hypochromic Anaemia
12	German Shepherd	Male	6	CKD	Normocytic normochromic Anaemia
13	Non Descript	Male	7	CKD	Normocytic normochromic Anaemia
14	Non Descript	Female	7	CKD	Normocytic normochromic Anaemia
15	Non Descript	Male	2	<i>B canis</i>	Macrocytic Hypochromic Anaemia
16	Pomeranian	Female	9	Pyometra	Microcytic Hypochromic Anaemia
17	Doberman	Male	6	CKD	Normocytic normochromic Anaemia
18	Labrador	Male	1	<i>B canis</i>	Macrocytic Hypochromic Anaemia
19	Non Descript	Female	8	Pyometra	Microcytic Hypochromic Anaemia
20	Labrador	Female	1	<i>B canis</i>	Macrocytic Hypochromic Anaemia

Treatment for the basic etiological cause was done to the respective anaemic dog, however, blood transfusion was made to each and every dog under study. Although the definitive criteria for diagnosing anaemia is variable with workups, for the purpose of blood transfusion only those dogs which had haemoglobin <5gm/dl and hematocrit < 20 per cent were selected considering that the degree of anaemia being critical, replacement transfusion would be more beneficial. The blood and serum samples were collected before and after transfusion at 24, 48, 72 hours and on the 7th day. Clinical inspection findings included examination of the mucus membrane, heart rate (HR, bpm), respiration rate (RR, rpm), temperature ( $^{\circ}$ F), hydration status etc. The haematological examination included CBC and reticulocyte count and was done on the fully automated Mindray BC-3000 haemoanalyzer. Major and minor cross-match along with donor and recipient control were performed as follows:

1. Major Crossmatch: 0.5 ml of donor RBC suspension plus 0.5ml of recipient serum in a glass test tube.
2. Minor Cross match:- 0.5ml of recipient RBC plus 0.5 ml of donor serum in a glass test tube.
3. Control: Donor and recipient controls were analyzed.

On confirmation of major and minor blood compatibilities blood transfusion was undertaken as per the procedure given [4].

## Results and Discussion

Twenty (20) clinical cases of anaemia were studied with respect to clinical examination haematologic values and reticulocyte count. The clinical signs observed in 20 cases were pallor of mucus membrane (100%), loss of appetite (100%), lethargy (100%), tachycardia (100%), polypnoea (100%), pyrexia (19.22%), altered gait (92.47%), dehydration (100%), vomition (19.17%), melana (33.25%), exercise intolerance (100%), splenomegaly (25.47%), palpable lymphonodes enlargement (66.32%) and dependent part oedema (32.14%). The capillary refill time (CRT) was also prolonged more than four seconds. These changes were relieved soon after the completion of the transfusion. The mucus membrane turned pinkish and was more appreciable at 24 hours post-transfusion. Similarly, capillary refill time also showed appreciable improvement. These changes could be

attributed to the favorable effect of blood transfusion. Similar results were obtained by O'Rourke (1983) [5] who observed that the full effect of transfusion is realized within 12 to 24 hours after the fluid has been distributed well. The body temperature, heart rate, pulse rate and respiratory rate before transfusion of whole blood and immediately after transfusion were recorded. The mean rectal temperature ( $^{\circ}$ F), heart rate (per min), pulse rate (per min), and respiration rate (per min) before and after whole blood transfusion was 101.8 $\pm$ 0.4, 107.8 $\pm$ 5.3, 99.3 $\pm$ 5.6, 31.0 $\pm$ 4.2 and 101.46 $\pm$ 03, 105.4 $\pm$ 5.5, 101.4 $\pm$ 5.9, 29.2 $\pm$ 3.4 respectively. Though there is a slightly lowered heart rate, respiration rate and slightly increased pulse rate insignificant ( $p$ <0.05) difference was recorded between them. There is no alteration in the mean temperature on before and after blood transfusion.

The highest prevalence of anaemia in the present study was noted in Labrador (45%) followed by mixed breeds (35%), and Non-Descript (ND) (20%) (Table 1). Our study was in accordance with Tandel *et al.* (2019) who revealed a high prevalence of anaemia in Labrador (35.29%) followed by non-descript (ND) (19.60%) German Shepherd and Doberman (13.73%). Lynch *et al.* (2016) [6] also recorded a more scattered distribution of anaemia in mixed breeds (n=132) followed by Labrador retrievers (n=74). The highest overall prevalence of anaemia was recorded in females (55%) as compared to males (45%) owing to the inclusion of pyometra as one of the causative agents of anaemia. However, if only CKD and *B canis*-originated anaemia cases were studied the highest prevalence of anaemia was recorded in males (74.99%) as compared to females (25%). All the anaemic cases of pyometra, CKD and *B canis* origin were recorded in geriatric dogs aged between 7-9 years, 5 to 7 years old and 1 to 2 years old respectively. Bhat (2016) [7] reported the same vulnerability of anaemia in pyometra and tick-borne disease and postulated the lowered innate immunity favoring the attack of ticks.

For the purpose of therapeutic decision, it is prudent to classify the type of anaemia considering the haematological indices. When the data of clinical cases of anaemia was analysed 40% (8/20) cases were recorded as microcytic hypochromic anaemia and normocytic and normochromic each, whereas macrocytic hypochromic anaemia cases were 20% (4/20) (Table 1). The hemoglobin concentration, red

blood cell count, and packed cell volume ranged between 3.48 to 8.92 g/dl, 2.56 to 4.54 ( $\times 10^6$  and 11.03 to 29.17 per cent with a mean value of 6.20 g/dl,  $3.55 \times 10^6/\mu\text{l}$ ., 20.1 per cent (Table 2). Tasker (1966) [8] stated that the degree of anaemia has to be evaluated by determining the packed cell volume, haemoglobin and total erythrocyte count. In his opinion, the packed cell volume is the test of choice that can be easily performed and more reliable. There were highly significant differences observed between the haemoglobin and erythrocyte values before treatment and 24 hours and between before-treatment values and 7 days post-transfusion ( $p < 0.01$ ). Also, there were significant differences between the values before treatment and 48 hours, 72 hours and 7 days post-transfusion ( $p < 0.05$ ). Similar observations were made by Keskar *et al.* (1985) [9] and Gupta (1992) [10]. Though the values were not within the normal range for dogs (12-15%) as per Jain (2022) [11], but are still adequate for oxygenation. The transfusion of blood in these cases has served the purpose of tiding over the critical period. The PCV values were highly significant ( $p < 0.01$ ) when the before-treatment and after-treatment values were compared to each other. There was a significant difference in MCH ( $p < 0.05$ ) after blood transfusion whereas the remaining two indices did not differ significantly. The association of non-regenerative anaemia with normochromic normocytic red cell indices is well established. Since the incidence of non-regenerative anaemia was recorded in the present study a normocytic normochromic pattern was expected. The reference range for normal dogs as suggested by Cowgill *et al.*, (2022) [12] for reticulocyte percentage is 0-1.5%. In the present study, reticulocyte count was less in before-treatment values. However, on

administration of blood, the reticulocyte count moved towards normalcy indicating positive effects of the ready source of red blood cells. Compensated and chronic anaemias are well tolerated in dogs as the stimulus (hypoxia) may not be strong enough to promote the continued synthesis of erythropoietin and thus initiate reticulocytosis [13]. Similarly, anaemia may appear nonregenerative late in the course of the disease (10-14 days after onset) even though the bone marrow is releasing increased numbers of mature erythrocytes.

A slight leucocytosis was observed which however started decreasing after 48 hours and further decreased when observed after 7 days (Table 3). There was a significant increase ( $p < 0.01$ ) in the leucocyte count after 24 hours when compared to pre-transfusion values. The values were found to be returning to pre-transfusion values after 7 days. Differential leucocyte count of the anaemic dogs revealed a highly significant difference between the values of total leucocyte count after transfusion of blood ( $p < 0.01$ ). The values of neutrophils and lymphocytes also showed highly significant differences ( $p < 0.01$ ). Schappap *et al.*, (1987) [14] and Hinton and Jones (1977) [15] have also reported similar results of constant leucocytosis. This could be attributed to lowered resistance and concurrent bacterial infection. In the present study, all the cases that were given blood transfusions were also simultaneously treated with antibiotics for three days as a precautionary measure. The leucocytosis that was evident initially showed a gradual decrease and by the 7th day post-transfusion, the mean TLC values showed a tendency to return to normal levels. This could be attributed to the antibiotic therapy.

**Table 2:** Mean  $\pm$  S.E of Haematological values of Anaemic dogs (n=20) before and after transfusion of Whole blood

Group	Hb (g/dl)	RBC's ( $\times 10^6/\mu\text{l}$ )	PCV (%)	MCV (fl)	MCH (fl)	MCHC (%)	Reticulocyte Count (%)	Absolute Reticulocyte count (millions/cmm)	Reticulocyte Production Index (RPI)
Before Blood Transfusion	3.48 $\pm$ 0.4	2.56 $\pm$ 0.4	11.03 $\pm$ 0.8	50.49 $\pm$ 5.7	14.3 $\pm$ 1.4	28.86 $\pm$ 2.0	0.6 $\pm$ 0.57	11.34 $\pm$ 1.02	0.09 $\pm$ 1.14
24 Hours	8.0 $\pm$ 0.7	3.79 $\pm$ 0.5	23.5 $\pm$ 1.4	66.55 $\pm$ 7.8	22.0 $\pm$ 1.7	34.06 $\pm$ 2.3	0.8 $\pm$ 0.05	30.32 $\pm$ 0.99	0.11 $\pm$ 1.21
48 Hours	8.57 $\pm$ 0.7	4.35 $\pm$ 0.5	27.5 $\pm$ 2.6	69.98 $\pm$ 10.6	20.86 $\pm$ 2.6	31.73 $\pm$ 2.2	1 $\pm$ 1.2	43.50 $\pm$ 1.23	0.28 $\pm$ 1.05
72 Hours	8.92 $\pm$ 0.5	4.62 $\pm$ 0.4	29.0 $\pm$ 1.9	65.37 $\pm$ 6.5	19.88 $\pm$ 1.3	30.96 $\pm$ 1.7	1.4 $\pm$ 2.1	64.68 $\pm$ 1.17	0.46 $\pm$ 1.14
7 Days	8.92 $\pm$ 0.3	4.54 $\pm$ 0.3	29.17 $\pm$ 1.8	65.43 $\pm$ 5.1	20.11 $\pm$ 1.5	31.10 $\pm$ 1.0	1.5 $\pm$ 0.25	133.80 $\pm$ 1.33	0.52 $\pm$ 1.13

**Table 3:** Mean  $\pm$  S.E of Leucogram in Anaemic dogs (n=20) before and after transfusion of Whole blood

Group	WBC ( $\times 10^3/\mu\text{l}$ )	Neutrophils (%)	Lymphocytes (%)	Monocytes (%)	Basophils (%)	Eosinophil (%)
Before Blood Transfusion	9.13 $\pm$ 1.03	80.33 $\pm$ 2.0	0.66 $\pm$ 0.3	18.33 $\pm$ 2.1	0.0	0.66 $\pm$ 0
24 Hours	13.85 $\pm$ 7.84	87.00 $\pm$ 1.8	0.83 $\pm$ 0.2	12.0 $\pm$ 1.6	0.0	0.17 $\pm$ 0.0
48 Hours	11.25 $\pm$ 8.74	76.0 $\pm$ 3.2	1.83 $\pm$ 0.0	21.17 $\pm$ 3.0	0.0	1.0 $\pm$ 0.0
72 Hours	9.73 $\pm$ 6.73	73.33 $\pm$ 2.4	0.67 $\pm$ 0.2	23.33 $\pm$ 1.7	0.0	2.67 $\pm$ 0
7 Days	9.25 $\pm$ 5.77	77.0 $\pm$ 1.0	2.5 $\pm$ 1.1	19.83 $\pm$ 1.2	0.0	0.67 $\pm$ 1.0

## Conclusion

From the present study, it can be concluded that whole blood transfusion is effective in anaemic dogs having etiology of CKD, Pyometra and *B. canis* infection when volume replacement is required. Blood can safely be given to needy anaemic patients after appropriate crossmatching only. Similar observations were made by Srinivasan (2000) [16] and Leisewitz (1996) [17].

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