The Haematological Values and Serum Iron Profile in Dogs with Some Pathological and Physiological Conditions

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Abstract

Iron is an essential element for animal health. Haematological measurements and iron profiles can be used to assess the pathological and physiological changes. Blood samples (N: 175) were collected from the cephalic veins of dogs in Baghdad for the period from November 2021 to March 2022 into EDTA tubes for complete blood counts, while plain tubes were used for serum collection for iron profiles measurements. The RBC count was significantly lower in groups of dogs that were exposed to accidents or bleeding, external parasites, or during pregnancy and gestation periods. HGB was significantly lower in the pregnancy or parturition group, the bleeding group, the inflammation group, the parvovirus infection group, and external parasite manifestation. A significant decrease in PLCR was noted in the infected group with parvovirus. The dogs' group with inflammation had the mean iron concentration significantly lower than the clinically healthy dogs. The study concludes that some haematological and iron profile characteristic changes were associated with pathological and physiological conditions in dogs.

Keywords: *Haematological, Iron, TIBC, Ferritin, dogs, Iraq.*

Introduction

Primary iron deficiency in dogs due to iron deficiency in the diet is uncommon, though changes in the pH of the gastrointestinal tract cause malabsorption of iron and an increase in the oxidised state of iron causes iron deficiency. Iron deficiency mostly occurs with haemorrhage and inflammatory conditions (Aird, 2000). Blood donation in dogs appears to not have iron deficiency, but transferrin saturation (TS%) is significantly decreased, especially in dogs with repeated blood (Zaldivar‐Lopez et al., 2014). Also, the marrow regenerative responses associated with blood donation can recover from blood changes within 10 days (Ferreira et al., 2014). Chronic hematoma can cause secondary iron deficiency anaemia. The pseudocyst of hematoma in the dog was reported by (Nakamizo et al., 2011).

On the other hand, ferritin is the iron storage protein in the liver. It is found in serum in small amounts when iron deficiency is present, and it is higher in some diseases, tumours, and conditions that cause inflammation (Friedrichs et al., 2010). Canine parvovirus is a target for rapidly dividing cells of lymphoid tissue, the gastrointestinal tract, and bone marrow. Parvovirus is damaged in the GIT, resulting in

haemorrhagic diarrhoea (Mazzaferro, 2020). The relationship between iron deficiency anaemia and parasitic worms like hookworm, especially with inadequate food intake, was investigated by (Jafer et al., 2015). The role of iron nanoparticles in parasite elimination and the importance of iron availability (Al-Qayim et al., 2021). The relationship is also presented between ferritin, liver iron concentration, and alanine aminotransferase (ALT) (Al-Momen et al., 2018). Hepcidin is the hormone that regulates iron absorption in the intestinal lumen; it is increased in pathophysiological processes (Al-Abedy, et al., 2021).

Serum iron, Total Iron Binding Capacity (TIBC), Unbound Iron Binding Capacity (UIBC), Transferrin Saturation (TS%), and ferritin are required tests to diagnose iron deficiency in dogs (Weiss and Wardrop, 2010). The TIBC amount represents serum transferrin. When TIBC levels rise above normal, the risk of inflammation and anaemia rises (Harvey, 2008). Serum ferritin and vitamin B12 levels must be measured in patients with some diseases, and a healthy diet must be rich in iron and vitamin B12 (Ali, 2022).

Blood is important to the health of animals. The pathological and physiological conditions of animals can be evaluated by haematological values and biochemical profiles (Khan et al., 2011). The aim of the study was to find differences in the haematological values and iron profile in some of the pathological and physiological states in dogs.

Materials and Methods

Samples collections

This study was done on 175 dogs that were chosen at random from the Baghdad veterinary hospital and a private veterinary clinic in Baghdad for the period from November 2021 to March 2022. Blood samples were collected from the cephalic vein into EDTA tubes for complete blood counts, while plain tubes were used for serum collection for iron profiles measurements.

Complete Blood Counts (CBCs)

The study was designed to detect haematological values by (Vet. Scan BIOTOO HA_300 VET haematology system): Haematocrit (HCT), Haemoglobin (HGB), Red Blood Cell Counts (RBCs), Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin Concentration (MCHC), Mean Corpuscular Haemoglobin (MCH), Red Cell Distribution Widths (RDWs), Red Cell Distribution Widths coefficient (RDWc), White Blood Cell Counts (WBCs), differential leukocyte count, Platelet count (PLT), Plateletcrit PCT, Mean Platelet Volume (MPV), Platelet Distribution Widths (PDWs), Platelet Distribution Widths Coefficient (PDWc), Platelet Large Cell Count (PLCC), Platelet Large Cell Ratio (PLCR).

Iron profile

The serum iron and Total Iron Binding Capacity (TIBC) concentrations were measured by the colorimetric method and commercial chemical kits (CENTRONIC GMBH, Germany). Ferritin concentration was estimated by Enzyme-Linked Immunosorbent Assay (Canine Ferritin (FE) ELISA Kit, Sunlong Biotech Co.Ltd, China), the Unbound Iron Binding Capacity (UIBC) and TS% were calculated according to (Weiss and Wardrop, 2010).

Detection canine parvovirus by immunochromatographic assay

Canine parvovirus was detected by commercial kit (CPV Ag, Rohi Biotechnology, China).

Statistical analysis: Statistical analysis was conducted by SPSS software (version 20, USA) for detecting variance and differences in means by one-way ANOVA at level P> 0.05.

Results and Discussion

One hundred thirty-seven dogs were chosen and divided into seven groups based on the following pathophysiological processes: 22 dogs were involved in an accident or bleeding, four dogs developed cancer masses, 17 dogs were exposed to inflammation as metritis, pneumonia, otitis, and urinary tract inflammation, 11 dogs were infected with parvovirus, seven dogs were pregnant or parturition, and 22 dogs manifested with external parasites (screw worm or ticks).

To investigate the effect of pathophysiological processes on hematological values, all test values are presented as mean + SE in (Tables 1, 2, and 3). The RBC count was significantly lower in groups of dogs that were exposed to accidents or bleeding, external parasites, or during pregnancy and gestation periods when compared with clinically healthy dogs. Furthermore, HGB was significantly lower in the pregnancy or parturition group, the bleeding group, the inflammation group, parvovirus infection group, and external parasite manifestation group than in any other clinically healthy group (Table 1). There were no significant differences in platelet parameters between dogs with a pathophysiological process and clinically healthy dogs (Table 2).

There were no significant differences in platelet parameters between dogs with a pathophysiological process and clinically healthy dogs except significant decrease PLCR of infected group with parvovirus when compared with clinically healthy (Table 2). The means of leukocyte counts (WBC) in the dogs' groups with inflammation, during parturition or pregnancy period, was significantly increased when compared to clinically healthy dogs, while only the monocyte of the inflammation group was significantly higher than that of the clinically healthy group (Table 3). Only the dogs' group that had evidence of an inflammation state had the mean iron concentration significantly lower than clinically healthy dogs. In contrast, there were no significant differences between other pathophysiological processes of dogs and healthy dogs in other values of iron profile (Table 4).

Table 1: The comparison of hemogram values between clinically healthy and in dogs' groups classified according to some pathophysiological process, Mean±SE.

Groups Test	Bleeding (22)	Abnormal mass (4)	Inflam-mation (17)	Parvo virus (11)	Pregnant or parturition (7)	External parasites (22)	Clinically Healthy (54)
RBC	4.85 ±0.25*	5.60 ±0.88	5.26 ±0.28	5.23 ±0.30	4.60 ±0.63*	4.78 ±0.29*	6.37 ±0.16
HGB	13.50 ±0.71*	16.20 ±1.66	14.25 ±0.54	14.33 ±0.56	13.91 ±0.78*	14.73 ±0.64	16.66 ±0.37
HCT	35.36 ±2.25*	39.63 ±4.95	35.89 ±1.34*	35.69 ±1.90*	32.05 ±3.29*	34.55 ±1.98*	44.26 ±1.04
MCV	69.82 ±1.39	72.12 ±3.39	69.33 ±2.10	68.66 ±1.51	72.60 ±1.52	72.84 ±1.36	70.05 ±0.78
MCH	38.61 ±10.97	28.72 ±1.18	27.35 ±1.09	28.83 ±2.63	35.22 ±4.23	31.97 ±1.10	27.09 ±0.40

MCHC	39.76 ±1.02	41.22 ±1.23	39.54 ±1.00	40.86 ±3.07	46.67 ±5.20	43.90 ±1.23	38.64 ±0.30
RDWs	36.24 ±1.68	34.87 ±3.09	33.88 ±1.29	37.14 ±0.90	35.78 ±0.86	55.67 ±15.29	36.70 ±0.87
RDWc	14.10 ±0.76	12.67 ±0.93	13.60 ±0.60	11.75 ±0.54	14.21 ±0.87	14.55 ±0.81	13.14 ±0.57

*Refer to presence of significant value at ($P \leq 0.05$) with clinically healthy. RBC ($\times 10^{6}$ / μ L), HGB (g/dl), HCT (%), MCV (fl), MCH (pg), MCHC (g/dl), RDWs (fl), RDWc (%).

Table 2: The comparison of platelet parameters between clinically healthy and in dogs' groups classified according to some pathophysiological process, Mean±SE.

	Bleeding (22)	**Abnormal mass (4)**	**Inflam-mation (17)**	**Parvo virus (11)**	**Pregnant or parturition (7)**	**External parasites (22)**	**Clinically Healthy (54)**
PLT	451.13 ±66.52	336.25 ±117.53	351.41 ±60.02	301.90 ±70.36	503.85 ±131.26	409.95 ±47.21	336.33 ±29.02
PCT	0.55 ±0.09	0.46 ±0.21	0.47 ±0.09	0.33 ±0.11	0.64 ±0.19	0.52 ±0.08	0.40 ±0.05
MPV	11.38 ±0.68	11.77 ±2.41	10.81 ±0.87	9.10 ±0.75	11.75 ±0.99	11.62 ±0.84	10.00 ±0.46
PDWs	18.25 ±1.23	20.75 ±4.35	19.14 ±1.47	19.06 ±2.31	16.85 ±1.15	16.89 ±1.17	16.52 ±0.81
PDWc	38.62 ±0.84	40.40 ±1.84	40.10 ±0.75	39.57 ±1.44	38.61 ±1.09	38.71 ±0.79	38.52 ±0.47
PLCC	161 ±26.47	142 ±61.97	126.88 ±28.33	103.18 ±31.20	176.57 ±50.67	154.63 ±23.23	114.74 ±13.43
PLCR	34.29 ±2.75	29.63 ±10.86	30.70 ±3.11	21.84 ±4.94*	36.77 ±2.98	31.12 ±3.29	29.41 ±1.73

*Refer to presence of significant value at ($P \leq 0.05$) with clinically healthy. PLT ($\times 10^{3/\mu L}$), PCT (%), MPV (fl), PDWs (fl), PDWc (%), PLCC ($\times 10^{3/\mu L}$), PLCR (%).

Table 3: The comparison of leukogram values between clinically healthy and in dogs' groups classified according to some pathophysiological process, Mean±SE.

	Bleeding (22)	**Abnormal mass (4)**	**Inflam-mation (17)**	**Parvo virus (11)**	**Pregnant or parturition (7)**	**External parasites (22)**	**Clinically Healthy (54)**
WBC	11.85 ±2.00*	7.30 ±1.65	11.18 ±2.11*	4.93 ±1.21	11.03 ±1.73*	9.53 ±1.62	8.85 ±0.49
Lymphocytes	2.76 ±0.59	2.61 ±0.88	3.92 ±0.99	1.70 ±0.54	3.20 ±0.96	2.51 ±0.52	3.05 ±0.31
Neutrophils	8.24 ±1.93	4.26 ±0.99	6.57 ±1.68	3.04 ±0.72	7.29 ±1.44	6.21 ±1.14	5.42 ±0.39

Monocytes	0.52 ±0.11	0.27 ±0.09	0.62 ±0.17*	0.13 ±0.03	0.41 ±0.13	0.36 ±0.07	0.31 ±0.03
Eosinophiles	0.32 ±0.12	0.13 ±0.08	0.07 ±0.03	0.04 ±0.03	0.11 ±0.07	0.40 ±0.24	0.05 ±0.01
Basophiles	0	0	0	0	0	0.02+0.02	0

*Refer to presence of significant value at (P≤0.05) with clinically healthy. WBC, Lymphocytes, Neutrophils, Monocytes, Eosinophiles, Basophiles ($\times 10^{3/\mu L}$).

Table 4: The comparison of iron profile values and copper concentration between clinically healthy and in dogs' groups classified according to some pathophysiological process, Mean±SE.

	Bleeding (22)	**Abnormal mass (4)**	**Inflam-mation (17)**	**Parvo virus (11)**	**Pregnant or parturition (7)**	**External parasites (22)**	**Clinically Healthy (54)**
Iron	161.06 ±15.56	245.27 ±123.92	142.53 ±16.35*	166.37 ±25.02	190.19 ±41.95	215.29 ±24.69	227.83 ±10.64
TIBC	419.64 ±28.61	408.97 ±81.10	566.06 ±85.05	459.00 ±40.78	424.90 ±58.52	582.57 ±74.75	496.31 ±47.53
UIBC	258.57 ±31.11	163.69 ±48.37	423.52 ±85.56	292.63 ±53.99	234.71 ±48.49	367.27 ±78.37	268.48 ±44.88
TS%	43.20 ±5.26	48.67 ±19.61	33.75 ±5.96	41.20 ±8.26	46.22 ±8.21	47.44 ±6.31	54.80 ±2.99
Ferritin	6.51 ±0.65	7.78 ±2.06	6.45 ±0.82	6.55 ±1.27	5.82 ±1.50	5.72 ±1.00	6.00 ±0.83
Copper	95.24 ±18.00	81.02 ±20.35	78.58 ±13.25	74.27 ±8.46	113.10 ±48.45	89.11 ±9.64	87.32 ±9.07

*Refer to presence of significant value at (P≤0.05) with clinically healthy. Iron, TIBC, UIBC (µg/dl), Ferritin (ng/ dl), Copper (mg/dl).

The haematological values and serum iron profile were documented in this study according to the pathophysiological process. A bleeding dog had a significant decrease in the RBC, HGB, and HCT and a significant increase in the WBC count. Iron deficiency can result from inadequate intake of the diet, decreased iron absorption, chronic blood loss, bleeding, neoplasia, and parasite infection (McCown and Specht, 2011), Foy et al. (2015) studied dogs that had blood donated more than six times in a 12-month period. Those dogs had HCT and TIBC significantly decreased. Other previous studies studied iron profiles in donor dogs that reported iron deficiency (Ferreira et al., 2014, Zaldivar‐Lopez et al., 2014). The results of the iron profile were not changed in the present study with bleeding, while Paltrinieri et al. (2010) showed a significant increase in serum iron and TIBC, and a significant increase in the TS% with acute haemorrhage in dogs. The TIBC was increased in animals with iron deficiency anaemia due to the role of the transferrin molecules in iron transport (Al-Rubaie et al., 2020). The haematological values

and iron profile were assessed in dogs with abnormal mass. No significant differences were detected when compared to the clinically healthy dogs. Serum ferritin and TIBC levels did not change significantly in immunoprecipitation dogs or tumour cases such as lung adenocarcinoma, haemangioma, and sarcoma (Chikazawa et al., 2013a). Also, another study observed no significant differences in the hepcidin, ferroprotein, transferrin, and ferritin of both dogs affected by benign and malignant tumours (Marques et al., 2017). While other studies reported decreased iron, ferritin (Alkhateeb et al., 2013), ferroprotein in the tumour state when compared to normal because iron is an important element for growth and cell differentiation (Andrews, 2008).

There was a significant decrease in HCT and serum iron with leucocytosis and monocities in dogs with inflammation cases. Paltrinieri et al. (2010) confirmed that TIBC in dogs with inflammation was higher than in control dogs. TIBC in inflammation may be normal, but it can rise due to inflammatory disease or secondary anaemia (Harvey, 2008). Another study also reported decreased serum iron and TIBC with anaemic dogs that decreased in MCV and MCHC (Chikazawa et al., 2013b).

When compared to clinically healthy dogs, dogs were infected with the parvovirus. There were significant decreases in HCT and PLCR only. However, a non-significantly lower in RBC, HGB, MCV, PLT, WBC, and serum iron was found in parvovirus cases compared to healthy dogs. Terzungwe (2018) found that the main haematological changes in parvovirus infection in dogs were anaemia and leukopenia. Castro et al. (2007) reported low HCT in most cases of parvo infection. In dogs, the low HCT occurred due to severe haemorrhage in the intestine following canine parvovirus infection (Terzungwe, 2018). Salem et al. (2018) investigated the significant increase in hepcidin levels in dogs infected with canine parvovirus. Hepcidin increased iron metabolism (Grimes et al., 2012) and stimulated inflammation (Nemeth et al., 2003). Canine parvovirus infected dividing cells, particularly those in the gastrointestinal tract, lymphoid tissue, and bone marrow, resulting in bloody diarrhoea (Mazzaferro, 2020).

The results revealed lower RBC, HGB, HCT, and leucocytosis in the pregnant and parturient dogs' states than in healthy dogs. The result provides information on the pregnancy or parturition status that is not affected by iron profiles. The relationship between reproductive period and haematological values in dogs was estimated by (Khan et al., 2011) who found a significant decrease in MCV and MCH. The erythrogram decreased during pregnancy due to the dilation of the circulatory system and the shorter life span of RBC (Cavill, 1995). Leucocytosis can occur in the post-partum stage, but this is not a reliable indicator of bacterial infection (Dior et al., 2014). The study suggested that iron deficiency has no predilection for pregnancy or parturition period in dogs. The study also suggested that the period of pregnancy in dogs is shorter than in humans or other animals. For this reason, iron deficiency is uncommon in pregnant dogs.

External parasites were manifested in 22 dogs. There was a lower rate of RBC and HCT in these dogs. The results did not observe any abnormalities in other haematological or biochemical values. A similar observation was documented by (Kebbi et al., 2020), who revealed that anaemia resulted in dogs due to hematophagous ticks. The ticks and external parasites had haemolytic effects because of their role in the protozoan parasites transmitted. The inflammation group included dogs with

otitis, pneumonia, and urinary tract infections. Increased levels of ferritin were reported when kidney function decreased (Al-Rubaie et al., 2016). The normal ranges of haematological values in dogs in Iraq were reported in the previous study (Badawi and Yousi, 2020), but ferritin values detected in other animal species (Badawi, 2014). The present investigation was the first documented observation of changes in the serum iron profile with blood values in dogs in Iraq.

Conclusion

The study concludes that some haematological and iron profile characteristic changes were associated with pathological and physiological conditions in dogs. This evidence was documented for the first time in Iraq.

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