

## Original article

# ASSESSMENT OF HAEMATOLOGICAL VALUES AND IRON PROFILE IN DOGS WITH IRON DEFICIENCY, IRON DEFICIENCY ANAEMIA AND ANAEMIA WITHOUT IRON DEFICIENCY

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## Summary

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Iron deficiency (ID) has important effects on both animals and humans, causing illness and nonspecific signs. The blood changes associated with ID develop as a decrease in some erythrocyte parameters and microcytic anaemia. In this study, 175 blood samples from dogs were obtained for the measurement of complete blood count (CBC), copper, and iron profiles that included serum iron, total iron binding capacity (TIBC), transferrin saturation (TS%), unbound iron binding capacity (UIBC), and canine ferritin. The cut-off values for serum iron and TS% were found using the receiver operator characteristic (ROC) curve test. The estimated cutoffs for the diagnosis of iron deficiency in the dogs were 115.74 mg/dL (serum iron) and 34.07% (TS%). The dogs with serum iron  $\leq 115.74$ mg/dL and TS%  $\leq$  34.07 were iron deficient. The dogs were divided into three groups based on the ROC curve results: iron deficiency (ID), iron deficiency anaemia (IDA), and non-iron deficiency anaemia (NIDA). The iron concentration and TS% of ID and IDA dogs were significantly lower than those of NIDA dogs, while the TIBC, UIBC, and ferritin did not differ among groups. The serum copper was not significantly lower in dogs with ID, but in the ID group (85.85±11.30 mg/dL) was less than levels in IDA and NIDA dogs (98.46±15.92 and 78.69±11.77, respectively). The study concluded that the ROC curve and area under the curve provided guidelines for the diagnostic accuracy of tests and the diagnosis of iron deficiency in dogs. The coefficient of variation of red blood cell distribution width (RDWc) was significantly higher when iron deficiency anaemia developed, as indicated by red blood cell (RBC) anisocytosis. The decreases in serum iron and TS% were considered "golden tests" for the diagnosis of IDA in dogs.

Key words: anaemia, dogs, haematological values, Iraq, iron profiles

#### INTRODUCTION

Iron deficiency (ID) has important effects on DNA synthesis, energy metabolism, and immunity. In humans, ID causes nonspecific symptoms as lethargy, and blood changes develop in later stages into microcytic hypochromic anaemia (Pantopoulos *et al.*, 2012; Houston *et al.*, 2018). The blood loss, urinary tract and gastrointestinal disorders, ecto- or endoparasites are common causes of ID in animals (McCown & Specht, 2011; Bohn, 2013). The iron supplements are important to rebalance iron status, especially in parasite infections (Al-Qayim *et al.*, 2021). Jafer *et al.* (2015) studied the relationship between hookworm infection and iron deficiency anaemia.

The body iron is heme, haemoglobin, and is stored as ferritin. The site of iron absorbed is the duodenum, as ferrous form, and it is transported through the enterocyte (Harvey, 2008). Iron is transported across enterocytes by ferroportin, then reaches the plasma and is bound to transferrin, then comes to the body organs or storage sites (Knovich et al., 2009). Ferritin concentration in serum is a good biomarker of ID in humans (Ferraro et al., 2012). However, ferritin levels can be elevated when inflammation occurs. High levels of ferritin are associated with reduced kidney function (Al-Rubaie et al., 2016). Many factors contribute to ferritin's limited diagnostic utility in iron deficiency (Ottenjann et al., 2006). The serum iron, the total iron-binding capacity (TIBC) and transferrin saturation (TS%) are used to evaluate iron disorders in various animal species (McCown & Specht, 2011; Bohn, 2013). The correlation between ferritin concentration and emaciation was estimated in sheep in Iraq (Badawi, 2014).

The iron-regulating hormone hepcidin, which acts as a regulator, regulates the level of iron absorbed from food in the intestinal lumen (Brasse-Lagnel *et al.*, 2011). As a result, when hepcidin concentrations rise, little iron can enter the enterocyte. When iron is absorbed by the intestine from the gut lumen, it can be released into the circulatory system via a unique membrane transport protein (ferroportin) on the enterocyte's basolateral membrane. The level of ferroportin released depends on hepcidin concentrations; lower hepcidin concentration levels provide more ferroportin (Collins *et al.*, 2008).

Clinical signs of iron deficiency, especially with anaemia can include exercise intolerance, lethargy, weakness, growth slowdown, weight loss, and malaise (Harvey, 2008). Pica development in IDA is distinct; evidence of bleeding, blood loss, or blood in faeces, pale mucus membrane, tachypnea, heart arrhythmia, heart murmur, or tachycardia may be found (Giger, 2005).

The mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular volume (MCV) can be used to assess ID but they are considered nonspecific indicators of ID (Vora et al., 2021). The iron status assays include serum iron, transferrin or TIBC and serum ferritin (Bohn, 2013). The parameters in IDA are expected to be low haematocrit, low MCV, low MCHC, low serum iron, normal or low TIBC, low ferritin (Naigamwalla et al., 2012). However, because of the lack of data references discussing the sensitivity, specificity, test accuracy of the iron profile and copper tests in the diagnosis and classification of dogs as irondeficient, iron-deficient with anaemia, or non iron-deficient with anaemia, this study aimed to diagnose iron deficiency in dogs and assess the haematological values in iron deficiency, iron deficiency anaemia, and in anaemia without iron deficiency.

## MATERIALS AND METHODS

## Animals and samples collection

In this study, 175 dogs were chosen at random from a private veterinary clinic and a veterinary hospital in Baghdad between November 2021 and March 2022. The dog cohort consisted of 50 police dogs (K9), 92 pet dogs, and 33 stray dogs, with a total of 106 males and 69 females aged from 2 months to 12 years. K9 dogs worked long hours for the police and were stressed, whereas stray dogs suffered from body condition loss due to negligence and the difficult environment on the streets. All 175 dogs comprised 81 dogs younger than one year, 49 dogs between one year to three years, and 45 dogs older than three years. According to the breed, K9 and pet dogs were 61 German Shepherds, 19 Malinois, 17 Terriers, 12 Huskies, 10 Kangal dogs, four Rottweilers, four Pointers, four Pitbulls, three wolfdogs, three Lolo foxes, and two Labrador retrievers, one of each Bulldog, Mastiff, and Samoyed breeds. All 33 stray dogs were crossbred. The dogs were clinically examined, and body temperature, pulse, respiratory rate, and clinical signs of babesiosis were recorded. The dogs were classified into three groups: iron-deficiency group (ID), iron-deficiency anaemia group (IDA), and non-iron deficiency anaemia group, according to haematological changes and the statistical analysis of the ROC curve and cut-off values of the iron profile.

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Blood for complete blood count was obtained from the cephalic vein, while serum was collected in plain tubes for iron profiles and copper measures. Complete blood count was analysed on Vet. Scan BIOTOO HA 300 VET haematology system. Commercial colorimetric kits were used to determine serum iron, copper, and total iron binding capacity (TIBC) (CENTRONIC GMBH, Germany). The canine ferritin (FE) ELISA kit was used to assay serum ferritin (Sunlong Biotech Co., Ltd., China). The transferrin saturation (TS%) and the unbound iron binding capacity (UIBC) were determined by equations of Weiss & Wardrop (2010).

#### Statistical analysis

SPSS software (version 20, USA) was used for statistical analysis to detect variance and significant differences using oneway ANOVA. Odds ratio was used to detect the risk factors related to the frequency of some physiological factors. ROC curve analysis was used to detect sensitivity, specificity, accuracy, and prognostic values of test cutoffs.

#### RESULTS

The receiver operating characteristic (ROC) curve as a statistical test was done to evaluate the diagnostic accuracy of tests for the diagnosis of iron deficiency in dogs (Table 1). The areas under curves (AUC) for the diagnosis of iron deficiency

Table 1. Area under curve values and test quality according to ROC curve

Tests	AUC	Test quality	Standard error	Significance	Asymptotic 95% confidence interval
Serum iron	0.979	Excellent **	0.013	0.0001	0.953-1.000
TIBC	0.462	Unsatisfactory	0.065	0.529	0.335-0.588
Ferritin	0.605	Satisfactory*	0.058	0.084	0.491-0.719
Transferrin saturation	0.920	Excellent**	0.027	0.0001	0.866-0.973

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Table 2. Detection of the prognostic values according to ROC curve

Test	Prognostic value	Sensitivity	Specificity
Serum iron	115.74	0.963	0.923
TIBC	547.275	0.308	0.873
Ferritin	2.5	0.923	0.291
Transferrin saturation	34.075	0.818	0.897

were 0.979 (excellent) for serum iron, 0.462 (unsatisfactory) for TIBC, 0.605 (satisfactory) for ferritin and 0.920 (excellent) for TS%. The study depended on the prognostic values of serum iron and TS% for detecting iron deficiency in the dogs. The results estimated sensitivity and specificity for cutoffs of serum iron (115.74 mg/dL) and a TS% (34.07%) for the diagnosis of iron deficiency in the dogs. Those dogs having serum iron equal or less than 115.74 mg/dL and a TS% equal or less than 34.07 were iron deficient (Table 2).

The ROC curve plots are presented on Fig. 1–3. These figures showed that the AUCs of serum iron and TS% were excellent and that of serum ferritin satisfactory, indicating that these tests are useful for the diagnosis of iron deficiency in dogs. The total number of dogs suffering from iron deficiency was 36 out of 175 dogs, including 19 with iron deficiency without

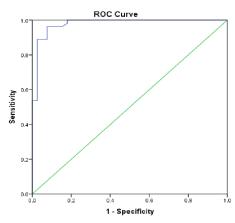
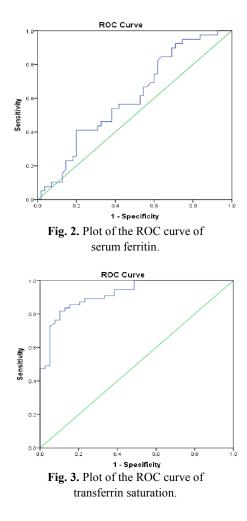


Fig. 1. Plot of the ROC curve of serum iron.

anaemia (ID) and 17 dogs with iron deficiency anaemia (IDA).

The clinical signs and haematological and biochemical values of ID, IDA, and non iron deficiency anaemia (NIDA) dogs were classified according to prognostic



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Tests	ID (n=19)	IDA (n=17)	NIDA (n=28)
RBC (×10 <sup>6</sup> /µL)	3.88–6.34	2.10-7.18	1.94–6.73
	5.37±0.15 A	3.87±0.25 B	4.08±0.20 B
HGB (g/dL)	13.10–16.20	7.30–13.0	3.80–12.50
	14.31±0.19 A	10.97±0.35 B	10.54±0.36 B
HCT (%)	27.70–67.50	15.70–36.75	11.20–38.98
	39.40±1.80 A	26.51±1.08 B	27.11±1.15 B
MCV (fL)	62.30–77.0	45.20-81.60	51.00–78.00
	70.53±0.93 A	69.12±2.13 A	68.02±1.26 A
MCH (pg)	23.60–39.90	16.70–43.60	18.30–44.50
	27.63±0.93 A	29.33±1.64 A	26.59±1.06 A
MCHC (g/dL)	27.60–51.50	33.60–54.90	31.60–59.30
	38.65±1.33 B	42.47±1.64 A	39.04±1.17 B
RDWs (fL)	25.40–52.60	21.70–64.70	20.30–375.00
	35.54±1.28 A	38.74±2.78 A	49.62±12.15 A
RDWc (%)	9.80–19.50	10.40–25.90	9.90–26.00
	12.41±0.56 C	15.89±0.97 A	15.01±0.74 AB

**Table 3.** Haemogram values of iron deficiency (ID), iron deficiency anaemia (IDA), and non-iron deficiency anaemia (NIDA) dogs (range and mean±SE).

The differences in small letters within rows refer to presence of significant value (P<0.05). RBC: red blood cells count, HGB: haemoglobin, HCT: haematocrit, MCV: mean corpuscular volume, MCH: mean corpuscular haemoglobin, MCHC: mean corpuscular haemoglobin concentration, RDWs: red blood cell distribution widths, RDWc: red blood cell distribution width coefficient of variation.

values and the quality of tests of the ROC curve. The results showed a significant decrease in the red blood cells (RBC), haemoglobin (HGB), and haematocrit (HCT) in both IDA and NIDA dogs. Mean corpuscular haemoglobin concentration (MCHC) was significantly higher in IDA dogs than in other groups. The IDA group was influenced by iron deficiency anaemia and had red blood cell distribution widths (RDWs) significantly increased when compared to iron deficient dogs (ID) dogs (Table 3).

Platelets count (PLT), plateletcrit (PCT), mean platelet volume (MPV), and platelets large cells count (PLCC) values were non-significantly lower in the iron deficiency group when compared to IDA and NIDA; however, white blood cells count (WBC) was significantly higher in

NIDA dogs when compared to ID and IDA groups (Table 4). The iron concentration and TS% of the values of iron-deficient and iron-deficient anaemic dogs were significantly lower than those of non iron-deficient anaemic dogs, while the TIBC, UIBC, and ferritin did not differ among groups. On the other hand, the copper concentration was not significantly lower in dogs with iron deficiency, but in the iron deficiency group (85.85±11.30 mg/dL) it was lower than in IDA and NIDA dog groups (98.46±15.92 and 78.69±11.77, respectively) (Table 5).

There were no significant differences between iron-deficient and non-irondeficient anaemic dogs, but this study found out a non-significant increase in frequency of clinical signs like abnormal Assessment of haematological values and iron profile in dogs with iron deficiency, iron deficiency ...

mass, urinary tract infection, and bloody diarrhoea (Table 6). When the relationship of physiological states with iron deficiency was investigated (Table 7), 45.5% of stray dogs had anaemia without iron deficiency (odds ratios: 7.6 and 9.5), which was significantly higher when compared to pets and K9 dogs while the K9 dogs were highly affected by ID and IDA compared to NIDA dogs. The risks of pregnancy, parturition, age, and sex between IDA and NIDA were investigated. The risk was non-significantly higher in dogs between 1 and 3 years of age. Also,

**Table 4.** Platelet parameters and leukocyte counts of iron deficiency (ID), iron deficiency anaemia (IDA), and non-iron deficiency anaemia (NIDA) dogs (range and mean±SE)

Tests	ID (n=19)	IDA (n=17)	NIDA (n=28)
PLT (×10 <sup>3</sup> /μL)	45.00–582.00	42.00–1075.00	33.0–853.00
	277.94±41.93 A	375.50±71.98 A	389.17±47.91 A
PCT (%)	0.03-0.96	0.04–1.47	0.02–1.13
	0.31±0.06 A	0.47±0.10 A	0.45±0.06 A
MPV (fl)	6.10–14.80	5.50–18.10	5.70-15.80
	9.62±0.60 A	11.33±0.87 A	10.81±0.54 A
PLCC (×10 <sup>3</sup> / $\mu$ L)	12.00–250.00	11.0–397.00	2.50–317.00
	84.94±15.60 A	137.22±29.54 A	131.76±19.25 A
WBC (×10 <sup>3</sup> /µL)	1.50–13.10	0.81–21.40	0.87-44.43
	7.75±0.77 B	8.68±1.28 B	13.38±2.16 A

The differences in small letters within rows refer to presence of significant value (P<0.05). PLT: platelets count, PCT: plateletcrit, MPV: mean platelet volume, PLCC: platelets large cells count, WBC: white blood cells count.

**Table 5.** Iron profile values and copper concentration of iron deficiency (ID), iron deficiency anaemia (IDA), and non-iron deficiency anaemia (NIDA) dogs (range and mean±SE).

Tests	ID (n=19)	IDA (n=17)	NIDA (n=28)
Iron (µg/dL)	18.22–106.78	0.83–113.9	121.61–291.67
	59.43±6.71 C	49.65±9.94 C	188.72±7.11 B
TIBC (µg/dL)	153.85–1258.97	105.13–1258.97	146.15–617.95
	427.53±56.26 A	446.47±66.25 A	421.73±21.96 A
UIBC (µg/dL)	116.93–1181.43	103.46–1233.97	24.54–460.32
	368.09±55.90 A	396.82±63.77 A	233.01±22.69 A
TS (%)	3.33–40.78	0.37–25.34	25.51–86.78
	16.78±2.28 B	11.26±2.07 B	48.73±3.32 A
Ferritin (ng/dL)	1.90–39.73	1.57–40.94	1.20–16.17
	7.55±1.87 A	8.57±2.04 A	6.64±0.68 A
Copper (µg/dL)	13.53–223.53	15.29–242.35	18.82–317.06
	85.85±11.30 A	98.46±15.92 A	78.69±11.77 A

The differences in small letters within rows refer to presence of significant value (P<0.05). TIBC: total iron binding capacity, UIBC: unbound iron binding capacity, TS: transferrin saturation.

Clinical signs	ID (n=19)	IDA (n=17)	IDA and ID (n=37)	NIDA (n=28)
Ticks' manifestation	0	3	3 (8.1 %)	2 (7.1 %)
Bleeding	3	3	6 (16.2 %)	6 (21.4 %)
Abnormal mass	0	3	3 (8.1 %) *	0 (0%)
Pale mucus membrane	14	16	31 (83.7%)	23 (82.1 %)
Mild pale mucus membrane	5	1	6 (16.2%)	5 (17.8%)
Skin lesion	0	1	1 (2.7 %)	2 (7.1 %)
Fever	12	5	17 (45.9%)	12 (42.8%)
Bloody diarrhoea	3	0	3 (8.1%) **	1 (3.5 %)
Anuria and urinary tract infection	2	1	3 (8.1%) *	0 (0%)
Otitis	1	0	1 (2.7 %)	0 (0%)
Respiratory signs	0	0	0 (0%)	2 (7.1%)
Metritis	0	0	0 (0%)	1 (3.5%)

Table 6. Clinical signs in iron deficiency (ID), iron deficiency anaemia (IDA), and non-iron deficiency anaemia (NIDA) dogs

\* Odds Ratio (OR): 1.08 (CI: 0.98-1.19); \*\* OR: 2.3 (CI: 0.23-24.2).

**Table 7.** Physiological states related to iron deficiency (ID), iron deficiency anaemia (IDA), and non-iron deficiency anaemia (NIDA) dogs

Physiological states	ID (n=19)	IDA (n=17)	IDA and ID (n=36)	NIDA (n=28)	
Pregnancy or parturition (n=7)	1	1	2 (28.5%)	2 (28.5%)	
Pet dogs (n=92)	11	6	17 (18.4%)	9 (9.7%)	
Stray dogs (n=33)	3	5	8 (24.2 %)*	15 (45.4%)**	
K9 dogs (n=50)	5	6	11 (22%)	4 (8%)	
*OR = 1.41(CI: 0.50	)–3.6); **OR 7	7.6 (CI : 2.9–20	.2), 9.5 (CI : 2.8–3	32)	
Less than 1 year (n=81)	9	6	15 (18.5%)	14 (17.2%)**	
1 to 3 years (n=49)	9	4	13 (26.5%) *	9 (18.3%)**	
More than 3 years $(n=45)$	1	6	7 (15.5%)	5 (11.1%)	
*OR =1.96 (CI: 0.70–5.4); **OR= 1.07 (CI: 0.47–2.7)					
Males (n=106)	12	10	22 (20.7%)*	17 (16%)**	
Females (n=69)	7	7	14 (20.2%)	11 (15.9%)	
*OR=1.02 (CI:0.48–2.1); **OR= 1.08 (CI: 0.55–5.8)					

the results demonstrated that the iron deficiency was not significantly influenced by pregnancy, parturition, or sex.

#### DISCUSSION

The results indicated the ability of the ROC curve to identify the tests with high sensitivity, specificity, and tests accuracy to diagnose iron deficiency in dogs. The serum iron and TS% had a high area under the ROC curve, indicating they are very effective in the diagnosis of iron deficiency. The dogs with serum iron of less than 115.74 mg/dL and a TS% of less than 34.074%, were considered iron deficient. Out of 175 dogs in this study, the total percentage of iron deficient dogs was

20.57%, including 9.71% of dogs suffering from iron deficiency anaemia. Some studies determined the iron deficiency in dogs by means of ROC curve analysis (Betting *et al.*, 2022) and determined cutoff values of some haematological values in dogs (Paltrinieri *et al.*, 2010).

Naigamwalla et al. (2012) affirmed that in iron deficiency anaemia in dogs, the serum iron and ferritin were significantly decreased but the TIBC may be within the normal range, while this investigation did not report the role of the TS% in the iron deficiency. Bohan (2013) found that the expected serum iron, TS, and ferritin values in anaemic dogs and cats with iron deficiency were lower; these findings were consistent with our findings, which determined serum iron and TS% to be excellent tests and ferritin to be a satisfactory test in the diagnosis of iron deficiency in dogs. Bhamarasuta et al. (2021) found a mean serum iron concentration of 103.4 µg/dL in anaemic dogs. Marchetti et al. (2010) reported the normal reference ranges in dogs for serum iron: 80-220 µg/dL and for TS%: 25-52%. Although the authors did not study the cut-off values, they indicated that when serum iron and TS% were elevated to 184 µg/dL and 49.3%, respectively, clinical signs of iron deficiency disappeared. This confirms the importance of investigating the cut-off values and testing accuracy and their role in clinical pathology.

The results did not record any significant changes in the copper concentration among groups of dogs. Copper is the element associated with anaemia, especially iron deficiency anaemia (Zentek & Meyer, 1991). The primary iron deficiency is not related to copper deficiency, but the latter can produce iron deficiency because of the importance of copper containing enzymes such as ceruloplamin and hephaestin in iron transport in the enterocyte (Harvey, 2008).

The results of the haematological values suggested that red blood cells, haemoglobin, and haematocrit were decreased in both IDA and non-IDA dogs. Bohn (2013) reviewed the blood values in IDA dogs and cats and also detected low or normal MCHC, as well as low MCV. The decreases in the MCV and MCHC were demonstrated in IDA (Chikazawa & Dunning, 2016). The present study noted high MCHC and RDWs in IDA dogs. The MCHC is the average of the haemoglobin concentration by the percentage of the RBC volume and the blood volume, calculated in g/dL (Ware, 2020). MCHC elevations were noticed due to RBC agglutination, optical interference of the samples, RBC disease and other causes such as RBC haemolysis, icterus, and lipaemia (Berda-Haddad et al., 2017). Another study also reported decreased serum iron and TIBC in anaemic dogs with decreased MCV and MCHC (Chikazawa et al., 2013). RDW is the parameter that reflects the degree of variation in RBC size (anisocytosis). RDW was elevated in iron deficiency anaemia because many cells in the population of erythrocytes decreased in size (microcytosis) in iron deficiency (Bermejo & García-López, 2009).

The increased total white blood cell count was recorded in aneamic dogs without iron deficiency. They occurred due to hypoxia and ischaemia in anaemia that may affect the leukocyte count (Singh, 2010). Also, the neutrophils release cytotoxic materials and hydrolytic enzymes that increase ischaemic injuries (Chia *et al.*, 2009).

According to the clinical findings, the most common signs associated with the IDA and ID groups were abnormal masses, urinary tract infections, and bloody diarrhoea. Iron deficiency anaemia is caused by a reduction in diet, iron absorption, chronic bleeding, and neoplasia (McCown & Specht, 2011). Other studies showed decreases ferroprotein, serum iron, and ferritin in tumour patients because of iron importance to cells that are rapidly growing and differentiating (Andrews, 2008; Alkhateeb *et al.*, 2013).

The frequency of ID, IDA and NIDA was increased in stray dogs, while ID and IDA affected K9 dogs more than NIDA. The starvation of stray dogs is considered the main cause of low blood values (Khan et al., 2011). The stray dogs are not provided with any veterinary care or antiparasitic treatment in Iraq. Anaemia develops in dogs without anti-parasitic drugs as some external or internal parasites can ingest more than 0.5 mL of blood per day (Epe, 2009). Some nematodes can cause bleeding or ulcer in the intestinal mucosa (Campos et al., 2017). Intestinal nematodes are considered a factor that can cause anaemia to develop (Christodoulou et al., 2010). Many studies found that stray dogs had a high parasite prevalence: 98% in Mexico (Alvarado-Esquivel et al., 2015), 66% in Iran (Beiromvand et al., 2013), and 75% in Serbia (Sommer et al., 2017). The stray dogs are exposed to various parasites or bacterial infections that may lead to haematological disorders such as iron deficiency anaemia. The K9 dogs in Iraq are exposed to severe stress during work. This stress increases the inflammatory disorders and corticoid production (Neel et al., 2012). The glucocorticoids are commonly administered to K9 dogs in Iraq to treat different conditions. Glucocorticoid administration is associated with haematological changes (Hammer, 1991).

The results demonstrated that the occurrence of anaemia was not affected by age or sex factors. Khan *et al.* (2011) detected non-significant decrease in haemoglobin in juvenile dogs when compared to adults. Some studies had reported significantly higher RBC, HCT, and HGB in male dogs, while others had documented non-significant differences between females and males (Neel *et al.*, 2012).

#### CONCLUSION

The study concluded that the receiver operating characteristics (ROC) curve analysis and area under the curve provided guidelines for the diagnostic accuracy of tests and the diagnosis of iron deficiency in dogs. The study detected that serum iron and transferrin saturation were better tests for diagnosing iron deficiency in dogs than ferritin, UIBC, and TIBC. The cut-offs for serum iron less than 115.74 mg/dL and a TS% less than 34.074% were considered to indicate iron deficiency. The total leukocyte count was significantly lower in iron deficiency anaemia than in non-iron deficiency anaemia, whereas RDWc was significantly higher in developing iron deficiency anaemia and points on RBC anisocytosis.

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