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Risk factors and metabolic alterations in healthy obese companion dogs in India

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Abstract
The main objective of this study was to determine potential risk factors associated with obesity and to evaluate haemato-biochemical and lipid alterations in overweight and obese dogs. A total of 95 healthy adult dogs (33 overweight, 46 obese and 16 control dogs) presented for routine health checkup or vaccination were selected for study. Results- Risk factors associated with obesity were feeding several times a day, table scraps, treats/snacks, obese and aged owners and small living areas. Hemoglobin (Hb) and Packed Cell Volume (PCV) values were significantly higher in overweight and obese dogs at $P \leq 0.10$ and $P \leq 0.05$ respectively. Total Erythrocyte Count (TEC) and Mean Corpuscular Volume (MCV) values were significantly ($P<0.05$) higher in both overweight and obese dogs. Albumin level was significantly ($P \leq 0.05$) higher in obese dogs while serum calcium level was significantly higher in overweight and obese dogs at $P \leq 0.05$ and $P \leq 0.10$ respectively. Serum lipid levels (Cholesterol, triglycerides, High Density Lipoprotein Cholesterol, Low Density Lipoprotein Cholesterol and Very Low Density Lipoprotein Cholesterol) were significantly ($P<0.05$) higher in overweight and obese dogs as compared to normal weight dogs. Hence, these haemato-biochemical parameters along with risk factor can be used for early diagnosis of obesity in companion dogs.

Keywords: Obesity, haemato-biochemical alterations, risk factors, dogs

1. Introduction
Obesity is defined as a body weight 15-20 percent above ideal weight and is usually the result of either excessive dietary intake or inadequate energy utilization, which causes a state of positive energy balance (German, 2006) [8]. This positive energy balance leads to adipose tissue hypertrophy, recruitment of macrophage and also changes in the adipocytes, their supporting tissue, blood supply and immunological milieu (Revelo *et al*., 2014) [27]. In many developing countries, particularly India, this is an emerging problem and Punjab is considered to be an obesogenic state in India, with highest prevalence (29.55%) of obesity in overweight/obese adults (Girdhar *et al*., 2016) [9]. During the last 50 years, the fast developing economy of Punjab with an agricultural base transformed the whole India from a food deficient to grain surplus nation and simultaneously, the Punjab witnessed a White revolution. This resulted in higher per capita availability of milk and milk products. Due to improved economic conditions and availability of nutritious food products, the living conditions and nutritional status of Punjabi population experienced a tremendous upward transformation (Kaur, 2016) [13]. In this prevailing transitional situation of the state, the increased overweight and obesity prevalence may be expected in companion animals as a possible link between obesity and the human-animal bond (Shearer *et al*., 2010) [33] and owners of overweight and obese dogs are of the opinion that food is an acceptable form of communication and interaction with their pets.
Keeping in view the above facts and paucity of literature pertaining to obesity in companion population in Punjab, this the first study to evaluate the metabolic alterations associated with obesity and to find possible risk factors leading to obesity in companion dogs in Punjab.

2. Material Methods
In healthy adult dogs presented for routine vaccination and general checkup, a complete physical examination was done at the beginning of the study. Body Condition Score (BCS) of each dog was evaluated using 5-points score (Mawby *et al*., 2004) [19] (1: thin, 2: underweight, 3: ideal, 4: overweight, 5: obese). Dogs with scale 4 and 5 were selected for further study.

Detailed questionnaire-based information was collected from the owners of these overweight (n=33) and obese dogs (n=46). Parameters recorded were feeding type, feeding frequency, table scraps, treats, exercise status and duration of exercise, owner health and age, house type and area of house.

Complete hematological, biochemical and lipid profiling of overweight group (n=33) and obese (n=46) of different breeds was carried out and results were compared with control group (n= 16).Whole blood sample (2ml) collected in sodium EDTA coated vials was used for complete blood count and Fully Automatic Laser Based Hematology Analyser (ADVIA ® 2120 Hematology system, Siemens Healthcare diagnostics Inc, USA) was used for determination of Hemoglobin (Hb, g/dl), Total Erythrocyte Count (TEC, $x10^6 \mu L^{-1}$), Packed Cell Volume (PCV, %), Platelet Count ($x10^3 \mu L^{-1}$), Total Leukocyte Count (TLC, μL^{-1}), Mean Corpuscular Volume (MCV, fL), Mean Corpuscular Hemoglobin (MCH, pg), Mean Corpuscular Hemoglobin Concentration (MCHC, g/dL). Differential Leukocyte Count (DLC) was done manually using Giemsa stain.

Metabolic analysis was carried out on serum samples that were stored at -20˚C before use. Vitros DT 350 Chemistry system (Ortho Clinical Diagnostics, Johnson & Johnson Company) by using Virtos DT slides was used for estimation of following parameters Renal function test: Blood Urea Nitrogen (BUN) (mg/dl) and Creatinine (Cr, mg/d1), Liver function test: Alkaline Phosphatase (ALP, U/L), Alanine Aminotransferase (ALT, U/L), Total Bilirubin (Tb, mg/d1), Total Protein (TP, g/dl), and Albumin (Alb, g/dl), Mineral estimation: Calcium (Ca, mg/dl) and Phosphorous (P, mg/dl). Diabetic markers- Glucose (Glu, mg/d1), Lipid profile: Cholesterol (Chol, mg/d1), Triglycerides (Tg, mg/dl), High Density Lipoprotein Cholesterol (HDL-C mg/dl).

Low Density Lipoprotein Cholesterol (LDL-C, mg/dl) and Very Low Density Lipoprotein Cholesterol (VLDL-C mg/dl) were calculated by Friedewald formula

VLDL-C = Triglyceride /5

LDL-C = Total cholesterol – (HDL-C + VLDL-C)

2.1 Statistical analysis

Data were recorded as average (Mean±S.E) and comparison between groups was done using one-way ANOVA with Tukey-Kramer Adjustment for hematological and biochemical parameters. Statistical analyses were performed with computer software (The SAS System, GLM Procedure).

3. Results

3.1 Risk Factors Associated with Obesity

Majority of overweight and obese dogs were being fed table scraps, snacks/treats and feed was provided several times a day (Table 1). In most of the overweight and obese dogs, movement was restricted inside house only and in majority of dogs which were taken for walk or exercise outside, the duration was less than half an hour per day. Owner's health and age were also found to be proportional to obesity (Table 1). Higher proportion of dogs living in apartment building or small area houses were found to be overweight and obese as compared to those living in villa or houses with >200 yard areas (Table 1).

Table 1: Analysis of Risk Factors Predisposing to Obesity in Companion Dogs

Parameters	Variables	Overweight (n=33)	Obese (n=46)	Total (n=79)
Feeding Records	Snacks/treats	84.8%	91.3%	88.61%
	Table scraps	66.6%	72.7%	69.62%
	Several times	84.8%	82.6%	83.54%
Exercise	Exercise Status	45.45%	43.47%	44.3%
	Duration of exercise 0.5-1H	6.66%	20%	14.28%
Owners health	Obese	57.58%	54.35%	55.96
	Normal	42.42%	45.65	44.30%
Owners age	<20Yr	3.03%	0	1.26%
	20-40Yr	24.24%	23.91%	25%
	>40Yr	72.72%	76.09%	75%
Home Demographics	Apartments	66.66%	63.04%	64.56%
	Single story	21.21%	19.56%	20.25%
	Villa	12.12%	17.39%	15.19%
Housing Area	<100y	78.79%	58.70%	67.09%
	100-200y	18.18%	28.26%	24.05%
	>200y	3.03%	13.04%	8.86%

Where H stands for Hour, Yr stands for year and y stands for yards

3.2 Haematology

The hemoglobin and PCV of overweight and obese dogs were significantly ($P \leq 0.10$ and $P \leq 0.05$) higher than the control group, respectively (Table 2). The TEC values were significantly higher ($P \leq 0.05$) in overweight and obese groups as compared to control group (Table 2). There was no significant difference in the platelet count in all the three groups at $P \leq 0.05$ but at $P \leq 0.10$, the platelet count was significant higher in obese dogs as compared to overweight group of dogs. The mean values of TLC of overweight and obese group did not differ significantly ($P \leq 0.05$) from control group (Table 2). The mean relative and absolute neutrophils, lymphocytes, eosinophils count of control, overweight and obese dogs are depicted in table 2 and counts did not differ from their respective control values. The mean MCV values of overweight and obese group were significantly higher

Table 2: Hematological Parameters (Mean ± S.E.) of Overweight and Obese Dogs

Parameters		Control (n=16)		Overweight dogs(n=33)		Obese dogs(n=46)		Ref. Range
		Mean ± S.E.	Range (min-max)	Mean ± S.E.	Range (min-max)	Mean ± S.E.	Range (min-max)	
Hb (gm %)		12.34±0.55[b]	9-15.1	13.88±0.38[b]	10-17.6	13.93±0.32[a]	10.5-18	12-18
PCV (%)		33.74±1.38[b]	26.9-39.5	37.30±0.96[b]	28.8-48.2	38.61±0.81[a]	30.4-52.5	37-55
TEC($x10^6\mu L^{-1}$)		5.70±0.25[b]	3.71-8.17	6.65±0.17[a]	4.87-8.4	6.61±0.15[a]	4.58-9.32	5.5-8.5
Platelets ($x10^3\mu L^{-1}$)		371.87±48.28	122-729	338.66±33.61	106-786	438.09±28.47	109-965	200-800
TLC (μL^{-1})		10695.63±564.88	7120-13790	11093.33±393.33	7280-15060	11570.04±333.14	6801-16650	6000-17000
N	(%)	81.81±2.22	68-96	84.42±1.54	54-98	84.72±1.31	58-98	58-85
	(μL^{-1})	8735.84±392.41	5553-11032	9449.87±433.20	4506-14156	9867.76±373.71	5376-15817	3000-11500
L	(%)	17.06±1.89	4-32	13.76±1.31	2-40	12.83 ±1.11	2-32	8-21
	(μL^{-1})	1847.9±241.83	354-3411	1449.37±134.20	254-3366	1441.46±106.10	202-3384	1000-4800
E	(%)	1.12±0.81	0-8	1.76±0.56	0-8	2.37±0.48	0-22	0-9
	(μL^{-1})	111.89±48.85	0-632	185.30±45.81	0-870	251.24±57.48	0-2039	100-1250
MCV (fL)		52.04±1.73[b]	41.1-63.7	57.77±1.20[a]	33-50	57±1.02[a]	39.6-77.2	60-77
MCH (pg)		21.11±0.45	14-24.7	21.21±0.31	18.4-23.9	21.45±0.26	15.9-25	19-24
MCHC(g/dL)		40.62±1.14	29.5-47.9	38.36±0.79	31.1-44.3	38.72±0.67	29.6-49.8	30-36

Values with different upper case letter in each row differ significantly at ($P \leq 0.05$) with control
Values in reference row are taken from (Rizzi *et al*., 2010) [28].

Table 3: Serum Metabolic Parameters (Mean ± SE) of Obese and Overweight Dogs

Parameters		Control (n=16)		Overweight Dogs (n=33)		Obese Dogs (n=46)		Reference range
		Mean ± S.E.	Range (min-max)	Mean ± S.E.	Range (min-max)	Mean ± S.E.	Range (min-max)	
Hepatic markers	Tb (mg/dL)	0.27±0.03	0.1-0.6	0.23±0.02	0.1-0.7	0.22±0.02	0.1-0.6	0.1-0.6**
	ALT (U/L)	46.31±7.46	29-74	58.69±5.19	22-174	59.78±4.40	29-154	8.2-49**
	ALP (U/L)	110.3±16.62	25-278	113.27±11.57	27-246	115.02±9.80	11-297	10.2-101**
	TP (g/dL)	6.86±0.29	4.6-8.9	7.30±0.20	5.5-10.8	7.43±0.17	6-12	5.5-7.5**
	Alb (g/dL)	3.02±0.14[b]	1.2-4.5	3.35±0.09[b]	1.9-4.5	3.49±0.08[a]	2.6-4.8	2.6-4**
Renal Markers	BUN (mg/dL)	13.56±1.59	6-25	13.78±1.10	6-25	14.02±0.93	7-29	8.8-25.9**
	Cr (mg/dL)	0.94±0.07	0.6-1.9	0.94±0.05	0.4-1.7	0.96±0.04	0.5-1.8	0.5-1.6**
Electrolytes	Ca (mg/dL)	9.2±0.27[b]	6.4-11.4	10.01±0.18[a]	7.1-12.6	9.94±0.16[b]	7.9-11.7	8.7-11.8**
	P (mg/dL)	3.81±0.33	1.9-5.5	4.26±0.22	2.4-10.7	3.85±0.19	1.3-8	2.9-6.2**
Diabetic markers	Glu (mg/dL)	74.31±7.71	15-153	89.45±5.37	19-138	82.71±4.54	11.00-153	62-108**
Lipid Profile	Chol (mg/dL)	157.12±18.41[b]	45-250	256.30±12.82[a]	116-424	286.19±10.86[a]	158-580	135-270**
	Tg(mg/dL)	55.37±11.94[b]	31-147	141.57±8.31[a]	65-220	153.65±7.04[a]	67-220	22-112**
	HDL-C(mg/dL)	88.43±6.37[b]	33-143	117.66±4.44[a]	49-170	130.04±3.76[a]	51-172	76.5*
	LDL-C(mg/dL)	57.61±16.58[b]	1-131.8	110.32±11.54[a]	-16.2-242	125.42±9.78[a]	15.6-436.2	111.5*
	VLDL-C(mg/dL)	11.07±2.39[b]	6.2-29.4	28.31±1.66[a]	13-44	30.73±1.41[a]	13.4-70.4	12.3 *

Values with different upper case letter in each row differ significantly at ($P \leq 0.05$) with control.
Values with *mark in upper case in reference row are taken from Osorio, (2009) [24]
Values with **mark in upper case in reference row are taken from Kaneko *et al*., (2008) [12]

($P \leq 0.05$) than control group (Table 2). The mean MCH and MCHC values of overweight and obese group did not differed from control (Table 2).

3.3 Biochemical Evaluation
Serum samples were analyzed for different biochemical test i.e; hepatic, renal, diabetic markers, electrolyte/mineral and lipid profile (Table-3)

3.3.1 Hepatic Markers
There was no significant difference ($P \leq 0.05$) in levels of Tb, ALT, ALP, total protein and albumin between control and overweight dogs, however, there was non-significant increase in levels of ALT, ALP, TP and Alb in overweight group in comparison to control group.
Similarly, there was no significant ($P \leq 0.05$) difference in levels of Tb, ALT, TP between control and obese group but obese group had significantly higher levels of albumin as compared to control group. In obese dogs, a non-significant increase in levels of ALT, ALP and TP were recorded as compared to control group.

3.3.2 Renal markers
There was no significant difference ($P \leq 0.05$) in levels of BUN and creatinine of obese and overweight dogs from control dogs.

3.3.3 Mineral estimation
The calcium levels of overweight and obese dogs were significantly higher ($P \leq 0.05$ and $P \leq 0.10$) than control group, respectively. However, there was no significant difference in the phosphorous levels of overweight, obese and control group dogs (Table 3).

3.3.4 Diabetic markers
There was no significant difference in mean value of serum glucose of obese and overweight dogs from control group.

3.3.5 Lipid Profiles
The mean values of cholesterol, triglyceride, HDL-C, LDL-C and VLDL-C of control, over weight and obese groups are depicted in Table 3. These values were significantly ($P \leq 0.05$) higher in overweight and obese dogs as compared to control

group. Though, there was no significant difference at $P \leq 0.05$ in the mean values between overweight and obese dogs, but all these parameters were non-significantly higher in obese group as compared to overweight group.

4. Discussion

Feeding several times, a day with reduced activity leads to positive energy balance. This positive energy is stored in the form of adipose tissue or fat leading to obesity (Mao *et al*., 2013) [18]. According to the owner's perception, the most handy and agreeable mode of communication and interaction with dogs is feeding which ultimately results in obesity (Kienzle *et al*., 1998) [15]. Prevalence of obesity is highest in dogs living in cages followed by those living in apartment buildings and single storied buildings as observed in developing country China obesity (Mao *et al*., 2013) [18].

The possible reason for increased hemoglobin values in overweight and obese dogs may be due to positive correlation between hemoglobin and Body Mass Index (BMI) and negative correlation between hemoglobin and adiponectin. Patients with high BMI have low circulating adiponectin and reduced insulin sensitivity and thus subsequently have poor glucose metabolic regulation that leads to increase in glycosylated hemoglobin level. This glycosylated hemoglobin has elevated affinity towards oxygen and high level of glycosylated hemoglobin subsequently leads to tissue hypoxia, which in turn results in an increase in red cell count and hemoglobin level that eventuate from normal physiologic feedback (Low *et al*., 2008) [17]. Increase in hemoglobin and RBC concentrations can result from elevated insulin and glucose levels in the blood of overweight and obese dogs as both insulin and glucose levels are believed to be increased in obese patients (Bersch *et al*., 1982; Kurtz *et al*., 1983; Aoki *et al*., 1994) [1, 16, 4]. Another possible reason for this increase could be setting up of inflammation leading to increased erythropoiesis in patients with obesity and metabolic syndrome. There is also increase in insulinsecretion in patients with obesity and this insulin and insulin-like-growth factors are believed to enhance erythropoiesis (Vuong *et al*., 2014) [37].

The increase in PCV could be due to increased sympathetic nerve stimulation leading to splenic contractions which forces the RBCs to move out into the blood circulation (Oystein *et al*., 2003) [25]. Another reason for increased PCV might be increase in the leptin levels in obesity which also causes sympathetic stimulation of nervous system (Nijima, 1998) [23]. The possible reason for this could be that with setting of acute or chronic inflammation; there is also increase in platelet count which may be influenced by a similar cytokine milieu as implicated in increased WBC count (Shah *et al*., 2012) [32].

A non-significant increase in WBC count in overweight and obese dogs could be due to higher neutrophilic count which is assumed to be higher due to stress or low grade inflammation (Radakovich *et al*., 2017) [26] but in the present study neutrophilic count in all the groups was within the normal range which might be the possible reason for difference in the results. Prolonged inflammation increases activation of WBC and endothelial cells which in turn leads to platelet and thrombus formation (Ross, 1999) [30]. The accompanying reductions in lymphocytes are explained by a failure of these cell types to respond to the inflammatory process to the same extent as neutrophils. Reduction in the lymphocyte numbers is common in the setting of acute inflammation-perhaps reflecting a response to physiological increases in corticosteroids, truly chronic inflammatory response, or in the recovery phase of an acute inflammatory response (Roytblat *et al*., 2000; Eder *et al*., 2009; Kaur, 2014) [31, 7, 14]. Thus, the effects seen with increasing waist circumference, as a result of obesity associated inflammation are most pronounced with neutrophils and results in a relative reduction of the effect seen on lymphocytes and other cells.

The values of MCV were slightly less than the normal values and the values of MCHC were slightly higher than the normal values (Rizzi *et al*., 2010) [28] and the possible reason for this could be due to variation of breeds (Hough *et al*., 2002) [10]. In present study, no relation of obesity with anemia was found, while there is risk of development of anemia in obese individuals (Tungtrongchitr *et al*., 2000) [35]. A negative correlation between increase in body weight and MCV values and decreased iron levels in obese individuals lead to decreased levels of MCV (Vuong *et al*., 2014) [37].

Higher level of ALT is assumed to be due to hepatic enzymes associated with higher prevalence of fatty liver as observed in obese humans (Choi, 2003; Tribuddharatana *et al*., 2011) [6, 34]. Non-significant higher serum total protein in the overweight/obese dogs may be attributed to decreased serum water fraction, antigenic stimulation or increased protein catabolism associated with a larger body mass (Radakovich *et al*., 2017) [26]. Increased albumin is typically accredited to dehydration and this increased production can also be stimulated by insulin, corticosteroids, sex hormones, thyroxine and growth hormone (Nicholson *et al*., 2000) [22].

The higher total calcium values are likely to be associated with the higher albumin concentrations in obese dogs. As the majority of calcium found in blood is bound to albumin, changes in total calcium often reflect changes in albumin. Measuring ionized calcium would be valuable to confirm this interpretation (Radakovich *et al*., 2017) [26].

Significantly higher levels of cholesterol, triglycerides, HDL-C, LDL-C and VLDL-C were found in overweight and obese dogs as compared to normal weight dogs. Increased feed intake along with sedentary lifestyle leads to increased energy intake which results in increased levels of triglycerides and cholesterol or alteration in lipid profile (Mori *et al*., 2011; Nandini *et al*., 2012) [20-21]. Increase in BMI is associated with increased body fat which is a strong predictor for the expression of a more atherogenic lipoprotein phenotype characterized by increased small, dense LDL-C particles with reduced surface lipids (Bray, 1985) [5].

The increase in level of triglycerides is also due to increase in the production of triglyceride rich lipoprotein and decrease catabolism of these proteins. Abnormal insulin activity results in increased lipolysis in adipose tissue leading to release of increased fatty acids which repack back into triglycerides in liver (Bailhache *et al*., 2003) [2]. Triglyceridemia and cholestrolemia is due to increased lipid concentration in all lipoprotein fractions (Isabelle *et al*., 2005) [31]. Increased amount of VLDL-triglycerides also causes increase in VLDL-cholesterol concentration. In addition, hepatic lipase activity can also lead to high HDL concentrations in blood (Barrie *et al*., 1993) [3]. Increase in obesity markers could be due to cumulative effect of daily feeding of high-fat diets which increases production of chylomicrons by intestinal epithelium resulting in obesity and abnormal insulin resistance (Johnson, 2005; Rocchini *et al*., 1987) [11, 29]. Increased VLDL-C and the presence of small-dense LDL-C particles are due to increased fibrinogen and PAI-1 (plasminogen-activator inhibitor) (Valle *et al*., 2001) [36].

5. Conclusion

From the present study, it is concluded that potential risk factors associated with obesity are feeding several times a day, feeding table scraps, treats, old aged owners, obese owners, dogs living in apartments or houses with less than 100 yard area.

Overweight and obese dogs have increased levels of Hb, TEC, PCV, MCV, Alb, Ca along with Chol, Tg, HDL-C, LDL-C and VLDL-C. Hence these parameters can be useful for early diagnosis of obesity in companion dogs.

It is necessary to replicate such researches to validate it further.

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7. References

1. Aoki I, Homori M, Ishikawa K, Taniyama M, Toyama K. Stimulatory effect of human insulin on erythroid progenitors (cfu-e and bfu-e) in human cd34+ separated bone marrow cells and the relationship between insulin and erythropoietin. Stem cells. 1994; 12(3):329-338.
2. Bailhache E, Ouguerram K, Gayet C, Krempf M, Siliart B, Magot T *et al*. An insulin-resistant hypertriglyceridaemic normotensive obese dog model: assessment of insulin resistance by the euglycaemic hyperinsulinaemic clamp in combination with the stable isotope technique. Journal of Animal Physiology and Animal Nutrition. 2003; 87(3-4):86-95.
3. Barrie J, Watson TD, Stear MJ, Nash AS. Plasma cholesterol and lipoprotein concentrations in the dog: The effects of age, breed, gender and endocrine disease. Journal of Small Animal Practice. 1993; 34(10):507-512.
4. Bersch N, Groopman JE, Golde DW. Natural and biosynthetic insulin stimulates the growth of human erythroid progenitors in vitro. The Journal of Clinical Endocrinology & Metabolism. 1982; 55(6):1209-1211.
5. Bray GA. Complications of obesity. Annals of Internal Medicine. 1985; 103(6):1052-1062.
6. Choi JW. Association between elevated serum hepatic enzyme activity and total body fat in obese humans. Annals of Clinical and Laboratory Science. 2003; 33(3):257-264.
7. Eder K, Baffy N, Falus A, Fulop AK. The major inflammatory mediator interleukin-6 and obesity. Inflammation Research. 2009; 58(11):727.
8. German AJ. The Growing Problem of Obesity in Dogs and Cats. Journal of Nutrition. 2006; 136(7):1940S-1946S.
9. Girdhar S, Sharma S, Chaudhary A, Bansal P, Satija M. An epidemiological study of overweight and obesity among women in an urban area of north India. Indian Journal of Community Medicine. 2016; 41(2):154.
10. Hough C, Kamisue S, Cameron C, Notley C, Tinlin S, Giles A *et al*. Aberrant splicing and premature termination of transcription of the FVIII gene as a cause of severe canine hemophilia A: similarities with the intron 22 inversion mutation in human hemophilia. thrombosis and haemostasis. 2002; 87(04):659-665.
11. Johnson MC. Hyperlipidemia disorders in dogs. Compendium on continuing Education for the Practising Veterinarian. 2005; 27:361-370.
12. Kaneko JJ, Harvey JW, Brus ML. Clinical Biochemistry of domestic animals, 6thedn. Academic Press, London, 2008.
13. Kaur A. Survey of obesity among various age group girls of Punjab. International Journal of Physical Educucation, Sports and Health. 2016; 3(2):296-299.
14. Kaur JA. Comprehensive review on metabolic syndrome. Cardiology Research and Practice, 2014.
15. Kienzle E, Bergler R, Mandernach A. A comparison of the feeding behavior and the human–animal relationship in owners of normal and obese dogs. The Journal of Nutrition. 1998; 128(12):2779S-82S.
16. Kurtz A, Jelkmann W, Bauer C. Insulin stimulates erythroid colony formation independently of erythropoietin. British Journal of Haematology. 1983; 53(2):311-316.
17. Low C, Tohit E, Chong P, Idris F. Adiponectin and hemoglobin levels in overweight and obese pregnant mothers in early pregnancy. The Internet Journal of Gynecology and Obstetrics. 2009; 11(2):1-6.
18. Mao J, Xia Z, Chen J, Yu J. Prevalence and risk factors for canine obesity surveyed in veterinary practices in Beijing, China. Preventive Veterinary Medicine. 2013; 112(3-4):438-442.
19. Mawby DI, Bartges JW, d'Avignon A, Laflamme DP, Moyers TD, Cottrell T. Comparison of various methods for estimating body fat in dogs. Journal of the American Animal Hospital Association. 2004; 40(2):109-114.
20. Mori N, Lee P, Kondo K, Kido T, Saito T, Arai T. Potential use of cholesterol lipoprotein profile to confirm obesity status in dogs. Veterinary research Communications. 2011; 35(4):223-235.
21. Nandini MK, Kamran CA, Yathiraj S, Upendra HA, Kumar VG, Rao S. Hematological, biochemical and lipid profile changes associated with obesity in dogs. Indian Veterinary Journal. 2012; 89(9):79-81.
22. Nicholson JP, Wolmarans MR, Park GR. The role of albumin in critical illness. British Journal of Anaesthesia. 2000; 85(4):599-610.
23. Nijima A. Afferent signals from leptin sensors in the white adipose tissue of the epididymis, and their reflex effect in the rat. Journal of the Autonomic Nervous System. 1998; 73(1):19-25.
24. Osorio JH. The variability in the canine lipid profile values and its possible relationship with the measurement method used. Vet Zootec. 2009; 3:70-77.
25. Oystein US, Knut H, Olav S. Blood and its function. In: Physiology of domestic animals.Edn. 1st. Scandinavian Vet. Press, Oslo, 2003, 281-303.
26. Radakovich LB, Truelove MP, Pannone SC, Olver CS, Santangelo KS. Clinically healthy overweight and obese dogs differ from lean controls in select CBC and serum biochemistry values. Veterinary Clinical Pathology. 2017; 46(2):221-226.
27. Revelo XS, Luck H, Winer S, Winer DA. Morphological and inflammatory changes in visceral adipose tissue during obesity. Endocrine Pathology. 2014; 25(1):93-101.
28. Rizzi TE, Meinkoth JH, Clinkenbeard KD. Normal

hematology of the dog. Schalm's veterinary hematology. 2010; 6:799-810.
29. Rocchini AP, Moorehead C, Wentz E, Deremer S. Obesity-induced hypertension in the dog. Hypertension. 1987; 9(6):364-368.
30. Ross R. Atherosclerosis—an inflammatory disease. New England Journal of Medicine. 1999; 340(2):115-126.
31. Roytblat L, Rachinsky M, Fisher A, Greemberg L, Shapira Y, Douvdevani A *et al*. Raised interleukin-6 levels in obese patients. Obesity Research. 2000; 8(9):673-675.
32. Shah B, Sha D, Xie D, Mohler ER, Berger JS. The relationship between diabetes, metabolic syndrome, and platelet activity as measured by mean platelet volume: the National Health and Nutrition Examination Survey, 1999-2004. Diabetes Care. 2012; 35(5):1074-1078.
33. Shearer P, Mus C, Mgt GC. Literature review: canine, feline and human overweight and obesity. Banfield Applied Research and Knowledge Team, 2010.
34. Tribuddharatana T, Kongpiromchean Y, Sribhen K, Sribhen C. Biochemical alterations and their relationships with the metabolic syndrome components in canine obesity. Kasetsart Journal (Natural Science). 2011; 45:622-628.
35. Tungtrongchitr R, Pongpaew P, Phonrat B, Tribunyatkul S, Viroonudomphol D, Supawan V *et al*. Leptin concentration in relation to body mass index (BMI) and hematological measurements in Thai obese and overweight subjects. Southeast Asian Journal of Tropical Medicine and Public Health. 2000; 31(4):787-794.
36. Valle M, Gascon F, Martos R, Ruiz F, R-ios R, Cañete R. Apo A-I decrease and tryglicerides increase serum levels in hyperinsulinemic children. Clinical Chemistry and Laboratory Medicine. 2001; 39:247.
37. Vuong J, Qiu Y, La M, Clarke G, Swinkels DW, Cembrowski G. Reference intervals of complete blood count constituents are highly correlated to waist circumference: should obese patients have their own normal values? American Journal of Hematology. 2014; 89(7):671-677.