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Insights into the Prevalence of Diabetes Mellitus in Dogs in Pakistan

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ABSTRACT

Diabetes mellitus is a condition that affects dogs and is caused by inadequate insulin production or action. The disorder is influenced by genetics, the immune system, and lifestyle exposures such as obesity and medication use. Dogs typically experience Type 1 diabetes and require exogenous insulin for life. The disorder is prevalent in middle-aged to older dogs, and the incidence rate among female dogs is increasing. Dogs with diabetes require insulin treatment and regular monitoring to ensure proper dosage. Various monitoring tools are available to veterinarians for effective management. This study aims to investigate the prevalence of diabetes mellitus in dogs in District Faisalabad and Islamabad Capital Territory of Pakistan. A total of 180 samples were collected, fasting blood glucose levels were measured using a commercial glucose meter and were more in females (127.78 mg/dl) than males (96.4 mg/dl). The overall prevalence for the 43 positive dogs examined were 23.88% with 95% confidence interval (CI) limits 18.25-30.62. Female dogs were more prevalent (42.30%) than male dogs (9.80%). On the basis of age, diabetes mellitus was more in dogs with age > 2 years (29.23%) and less in dogs with age < 6 months (10.34%). Neutered female dogs were at high risk (35.93%) as compared to neutered male dogs (11.68%) while intact male dogs were at much higher risk (28.20%) than neuter male dogs but intact female dogs were considerably less at risk than neuter females. Laboratory analysis was carried out to evaluate the other markers related to diabetes which includes urinalysis, liver functioning tests, CBC and serum biochemistry. The mean values of ALT and ALP were more in females (92.95 & 264.83) than males (62.24 & 239.02). This study followed the recommendations of the Recognized Animal Ethics Committee, University of Agriculture, Faisalabad. The outcomes of this research give valuable evidence on the prevalence of diabetes in dogs in Pakistan and will help raise awareness about this disease among pet owners.

INTRODUCTION

A disorder known as diabetes mellitus which involves the endocrine system causes increased blood glucose concentration as a result of inadequate insulin production or action (Rand, 2020). Compared to cancer and cardiovascular illnesses, diabetes is predicted to be the most serious global health hazard in the coming 25 years (Hassan *et al.*, 2022). This condition is generally observed in middle-aged to old dogs (Heeley *et al.*, 2020) and is characterized by changes in carbohydrate, lipid, and protein metabolism, as well as elevated blood sugar concentration induced by a decrease in insulin action or secretion (Ahmed, 2006). The development of diabetes mellitus is influenced by various factors, including genetics (particular alleles and hereditary characteristics), immune system (such as islet cells death or inflammation of pancreas) and lifestyle exposures (such as Overweight, nutrition, and use of medications or harmful substances) (Sidhu and Randhawa, 2019). Various common clinical and pathological findings of diabetes mellitus in dogs, including fasting hyperglycemia, increased cholesterol in blood, more hepatic enzymes secretions, presence of glucose and proteins in urine, neutrophilic leukocytosis, higher specific gravity of urine (Greco, 2018). Other symptoms may include polydipsia, polyuria, weight loss, lethargy, cataract formation and hepatomegaly (Sidhu and Randhawa, 2019). Different liver functions test such as ALP and ALT are thought to be useful indicators of liver damage (Sindi *et al.*, 2023). In cases of long-standing diabetes mellitus, diabetic ketoacidosis, dehydration, vomiting, depression, and coma may also occur (Sidhu and Randhawa, 2019).

Diabetes in dogs is a disorder related to prolonged hyperglycemia because of the loss or dysfunction of insulin secretion by pancreatic beta cells, decreased insulin sensitivity in tissues, or both (Cook and Dip. ACVIM-SAIM, 2012; Wubie and Getaneh, 2015). When carbohydrates from food are broken down into glucose, insulin is responsible for regulating its absorption and delivery to diverse body tissues. Without sufficient insulin or a proper response to it, dogs face challenges in metabolizing glucose correctly, resulting in higher blood glucose levels or hyperglycemia (>180-220 mg/dl) (Nelson, 2015). In dogs, Immune-mediated death, vacuolar degradation or pancreatitis are the most common causes of beta-cell loss, which often happens quickly and gradually (Behrend *et al.*, 2018).

Type 1 diabetes results from factors like genetics, persistent hyperglycemia, pancreatic issues, or autoimmune beta cell loss (Pápa *et al.*, 2011; Stoy, 2014). Conversely, Type 2 diabetes, causing insulin resistance, is linked to environmental factors and certain illnesses (Blois *et al.*, 2011). Prolonged pancreatic inflammation, sedentary lifestyle, and obesity are common contributors (Tvarijonaviciute *et al.*, 2012). Dogs usually experience Type 1 diabetes and require lifelong insulin (Ettinger *et al.*, 2016; Kour and Chhabra, 2021). Gestational diabetes mellitus (GDM) occurs in mature female dogs during late pregnancy, linked to insulin sensitivity reduction and increased progesterone levels (Kim *et al.*, 2019). Pre-diabetic forms are seen in non-diabetic dogs with chronic pancreatitis. Breeds like Golden Retrievers, German Shepherds, and Keeshonds have higher juvenile diabetes rates (Caney, 2013).

Diabetes mellitus is a growing global health concern for both humans and dogs. Healing diabetic wounds, which are often characterized by poor blood supply and infection, is a significant challenge (Güngör *et al.*, 2022). In dogs, the prevalence of diabetes has been steadily increasing (Feldman, 2004), with a 32% rise between 2006 and 2010 based on data from the Banfield Hospital of USA (Association, 2010). Herrtage (2009) reported an increase in incidence from 0.19% in 1970 to 0.64% in 1999 in the UK. Over 120,000 dogs in the UK's first opinion practices have a 0.34% prevalence of diabetes mellitus (Mattin *et al.*, 2014). During this period, the mortality rate decreased from 37% to 5%, indicating improved management of diabetic dogs. The cause of hyperglycemia in dogs is likely complex, as noted by Nelson and Reusch (2014).

The cause of canine diabetes mellitus being related to autoimmune activity in the islets was not well established (Gilor *et al.*, 2016). Typically, middle to older-aged dogs, among 5-12 years old, are more prone to developing diabetes mellitus (Hess, 2013). Although it was previously believed that female dogs were twice as likely to be affected compared to males (Guptill *et al.*, 2003). In 2005, a survey conducted in the UK showed that only 53% of the cases were female (Davison *et al.*, 2005). This is probably because of more optional neutering procedures. A uniform rise incidence rate of 73.3% among female dogs (Das and Lodh, 2015). Intact female dogs may experience temporary or permanent diabetes because of resistant effects of insulin during the phase of diestrus. Two studies have indicated a link between excess weight and canine DM (Wejdmark *et al.*, 2011), but it's important to note that the body condition score was recorded by owners after the DM diagnosis. A study in Sweden that included 860 dogs reported a substantial rise in DM occurrence among females during spring, but no such seasonality was found for male dogs (Fall *et al.*, 2007). However, the neuter status of the dogs was not reported, although it was assumed that most females were intact (Fall *et al.*, 2007).

A serious side effect of diabetes called diabetic ketoacidosis (DKA) is brought on by an excessive generation and accumulation of keto-acids, which leads in metabolic acidosis (Nelson, 2015). Numerous researches have indicated the effectiveness and practicality of continuous glucose monitoring systems (CGMS) in the field of veterinary practice (Corradini *et al.*, 2016). A device that monitors interstitial glucose (IG) and communicates the recorded data to a transmitter is typically used in CGMS. The flash glucose monitoring system (FGMS), in contrast to other CGMS, does not require calibration, is accurate in determining IG in dogs with diabetes mellitus, and is also well-tolerated by dogs (Corradini *et al.*, 2016).

While portable glucometers designed for humans are often used in veterinary care, it's essential to validate their suitability for animal use. Glucometers designed specifically for veterinary use may provide more accurate results, and relying on human-designed ones without proper validation for animals can be unreliable (Cohen *et al.*, 2009; Domori *et al.*, 2014). The accuracy of glucometer readings can also be influenced by various factors, such as hemoglobin concentration, sample size, test kit reliability, and sample characteristics, among others (Johnson *et al.*, 2009). In the past, glucometer efficiency was assessed by comparing their results to reference methods like glucose oxidase or hexokinase (Mori *et al.*, 2016).

Objectives

Keeping in view the importance, the main goals have been set for the current research are;

- Check the prevalence of diabetes mellitus in dogs and determine the associated risk factors.
- Assess the efficacy of two transportable glucose analyzers for person use in dogs with normal hematocrit levels across three blood sugar levels (less glycemic, normo-glycemic, and more glycemic).

MATERIALS AND METHODS

Study Area

The current study was done in two different areas, District Faisalabad of province Punjab and Islamabad Capital Territory, Pakistan. Pet clinics of Islamabad and District Faisalabad were chosen to collect the samples because a large number of dogs are kept there as indoor pets. There are more than 190,200 registered dogs in Pakistan (Federation Cynologique Nationale). In large cities, a significant number of people choose to keep dogs as their pets. However, keeping pets in Pakistan is still predominantly considered a luxury of the upper-class (Kennel Club of Pakistan).

Study Animals

As of 2021, Labradors, German Shepherds, Rottweilers, and Siberian Huskies are the most popular dog breeds in Pakistan (Profit). Dogs were selected based on the history of excessive thirst, polyuria, polyphagia, and weight loss (Sidhu and Randhawa, 2019). Risk factors-based questionnaire on diabetes mellitus was include breed, age, gender, neutering status, feeding and obesity (Kour and Chhabra, 2021). Dog owners who attend the Veterinary Teaching Hospital University of Agriculture, Faisalabad, and several pet hospitals in Islamabad for check-up (routine or specific) was asked about their knowledge of canine diabetes mellitus as part of this study's concept.

Method of Study

A questionnaire with queries about the owner and the dog, was created. Owners were briefed on the study's intent prior to the sample collection and asked for their permission to collect blood samples from their dogs. Each donor dog's breed, age, gender, diet, health history, immunization history, and neutering status were recorded on the field data gathering questionnaire after obtaining consent.

Sample Collection

Seventy blood samples were collected from both healthy and ill dogs owned by clients presented at two different pet clinics in Islamabad Capital Territory: Animal Health Clinic, Bani Gala (N = 30) and Pets & Vets, F-7/4 (N = 30), along with 10 samples from CDRS Animal Welfare Benji TNVR facility. One hundred and ten blood samples were taken from dogs presented to Veterinary Teaching Hospital, University of Agriculture, Faisalabad (N = 60) and two pet clinics present in District Faisalabad, Animal Care Clinic, Sargodha Road (N = 30) and Al-Huda Pet Clinic, Canal Road Near Lyallpur Galleria, Faisalabad (N = 20). The blood was collected using alcohol swabs, sterile disposable syringes with wide-bore needles, and serum vacutainers/tubes. The blood collection procedure followed the recommendations from Washington State University's College of Veterinary Medicine's Small Animal DX & Therapeutic Techniques (Shabbir *et al.*, 2013), involving proper dog restraint, shaving the area below the elbow to access the cephalic vein, and drawing 2 to 5 ml of blood directly from the unobstructed cephalic vein into the sample tube.

Analytical Procedure

After collection of blood samples, analytical procedures were performed which includes detection of blood glucose concentration by portable glucose meters (PGM) and Liver functioning tests.

Blood Glucose Test by Portable Glucose Meter (PGM)

The analysis of blood glucose was performed using commercially available glucose meter Accu-Chek®, Easy-Gluco® and D-Check® as shown in figures 1,2,3. Two to three drops of blood were contacted to the end of strip of glucometer. Consequently, the concentration of glucose will be noted in a moment on the display screen of device. If the fasting blood glucose value is >180 mg/dl, then it considered as diabetic.

Collection of Serum from Blood Samples

The separated serum was then kept at 20°C for further usage after the collected blood samples had been centrifuged at 4000 rpm for 8 minutes. After separation, serum samples from serum vacutainers were converted into Eppendorf tubes with proper labelling on each tube. Then, to assess additional diabetes-related indicators, serum samples from all dogs were brought to the Veterinary Preventive Medicine & Public Health Laboratory, Department of Clinical Medicine and Surgery, University of Agriculture Faisalabad.

Liver Function Tests (LFT's)

Liver function tests (LFT's) diagnose and monitor liver conditions. Key enzymes in this test are ALT, AST, and ALP (Kuotsu *et al.*, 2023). The materials used include serum samples, pipettes, specimen tubes, ALT and ALP test kits from Centronic GmbH/Germany, and a URIT-880 chemistry analyzer. The procedure followed manufacturer guidelines for the test kit. Compliance with safety and quality control procedures is crucial for accurate results. Serum samples in Eppendorf tubes were refrigerated for further analysis.

Other Recommended Tests

Urinalysis (specific gravity of urine, glycosuria, ketonuria), CBC (normal to neutrophilic leukocytosis) and serum biochemistry (hyponatremia, hypokalemia, hypochloremia, hypophosphatemia) were further aid in to confirm the diabetes mellitus in such dogs.

Statistical Analysis

In this study, after categorizing the data, 95 percent confidence intervals (CI) and prevalence rates were calculated using the method discussed previously (Newcombe, 1998). The Statistical Software Statistics 8.1 for Windows® was used to conduct the statistical analysis. The degree of association between the various variables was evaluated using the chi-square test. Odd ratio with 95% confidence interval was calculated using software WinPepi. The statistical significance of the connections was established using a significance level of $P < 0.05$. The research was conducted following the guidelines provided by IBM Corporation (Route 100 Somers, New York, USA) for using their software. The study aims to establish meaningful associations and draw valid conclusions based on the obtained results.

RESULTS

Figure 1: *Accu-Chek*

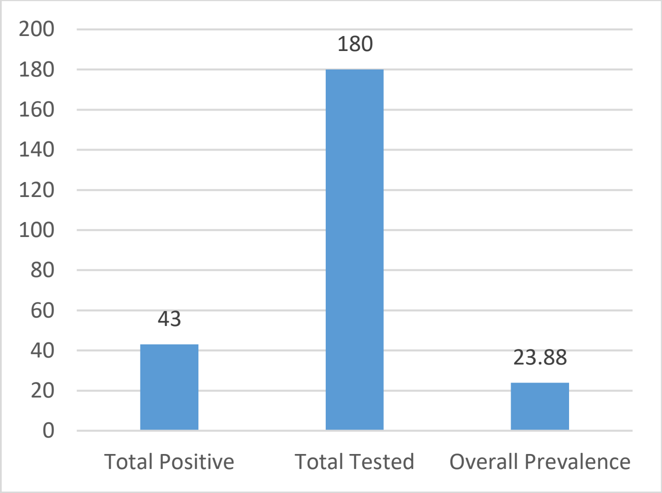
Figure 2: *Easy-Gluco*

Figure 3: *D-Check*

Overall Prevalence

Diabetes mellitus was diagnosed in 43 dogs out of 180 dogs who came to different pet clinics for their specific or general checkup. The overall prevalence for the positive cases were 23.88% (Figure 4) with 95% confidence interval (CI) limits 18.25–30.62. In univariate study, notable risk factors were weight, gender, neutered or spayed versus intact, mixed versus pure breeds, and female versus male. The multivariate model incorporated every potential risk factor.

Figure 4: *Overall prevalence of diabetes mellitus in dogs*



Sex Wise Prevalence

Sex wise prevalence of diabetes mellitus in dogs were as follows: 33/78 (42.30%) were female dogs and 10/102 (9.80%) male dogs (Table 1). Female dogs were at an increased risk compared with male dogs ($P = 0.0001$). The mean values of blood sugar, ALT and ALP were 96.4, 62.24 and 239.02 for males and 127.78, 92.95 and 264.83 for females, respectively and were more in female dogs than male dogs (Table 2, Figure 5).

Figure 5: *Sex wise prevalence of diabetes mellitus with relation to ALT and ALP*

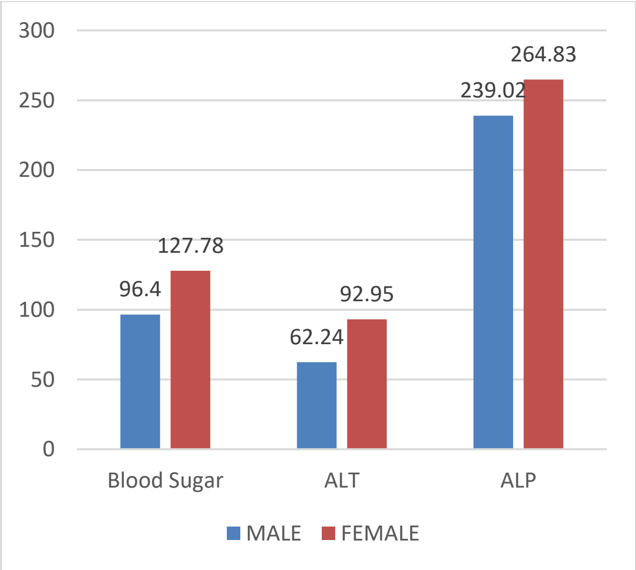


Table 1
Chi-Square Test for all the respective parameters for the prevalence of diabetes mellitus.

Parameters	Category	Positive	Tested	Prevalence %	95% C.I	O.R (95% C.I)	Chi-Square Value	P-value
Sex	Female	33	78	42.30%	31.96-53.38	4.32 2.01-9.26	15.50	0.0001
	Male	10	102	9.80%	5.41-17.11			
Age	> 2 year	19	65	29.23%	19.58-41.20	2.83 0.79-10.12	3.02	0.3891
	1-2 year	14	51	7.45%	17.11-40.95	2.65 0.72-9.82		
	6 mon-1 year	7	35	20%	10.04-35.89	1.93 0.47-7.99		
	Up-to 6 months	3	29	10.34%	3.58-26.38			
Body Weight	>45 kg	21	50	42%	29.38-55.77	6.72 1.51-29.99	9.47	0.0237
	30-45 kg	11	47	23.40%	13.60-37.22	3.74 0.79-17.65		
	15-30 kg	9	51	17%	9.57-30.26	2.82 0.59-13.61		
	Up to 15 kg	2	32	6.25%	1.73-20.15			
Body Condition Score	>6 Score	19	57	33.33%	22.49-46.27	4.08 1.31-12.69	6.62	0.0849
	5-6 Score	15	53	28.30%	17.96-41.57	3.47 1.09-11.05		
	3-4 Score	5	21	23.80%	10.63-45.09	2.92 0.73-11.72		
	Up to 2 Score	4	49	8.16%	3.22-19.18			
Neutering status	Spay	23	64	35.93%	25.29-48.18	3.07 1.34-7.08	7.39	0.0248
	Intact	11	39	28.20%	16.55-43.78	2.41 0.93-6.26		
	Neuter	9	77	11.68%	6.27-20.75			
Diet	Homemade	23	77	29.87%	20.80-40.85	1.61 0.79-3.31	1.72	0.4225
	Mixed	5	22	22.72%	10.12-43.44	1.23 0.41-3.68		
	Dog Food	15	81	18.51%	11.56-28.33			
Vaccination Status	Irregular	21	58	36.20%	25.06-49.08	2.53 1.14-5.64	5.31	0.0703
	Regular	11	45	24.44%	14.23-38.67	1.71 0.69-4.24		
	Not Vaccinated	11	77	14.28%	8.17-23.80			
Breeds	Rottweiler	9	25	36%	20.25-55.48	7.56 0.93-61.66	7.63	0.3660
	German Shepherd	13	41	31.70%	19.57-46.99	6.66 0.85-51.99		
	Golden Retriever	6	19	31.57%	15.37-53.99	6.63 0.77-57.30		
	Labrador	7	23	30.43%	15.60-50.86	6.39 0.76-53.74		
	Mixed breed	3	20	15%	5.24-36.04	3.15 0.32-31.15		
	Terrier	2	15	13.33%	3.73-37.88	2.80 0.25-31.71		
	Black Shepherd	2	16	12.5%	3.50-36.02	2.62 0.23-29.68		
	Husky	1	21	4.76%	0.85-22.67			
Glucometer	Accu-Chek	27	81	33.33%	24.03-44.14	2.25 0.73-6.90	4.43	0.1094
	EasyGluco	12	72	16.66%	9.80-26.91	1.13 0.34-3.73		
	D-Check	4	27	14.81%	5.91-32.47			

Age Wise Prevalence

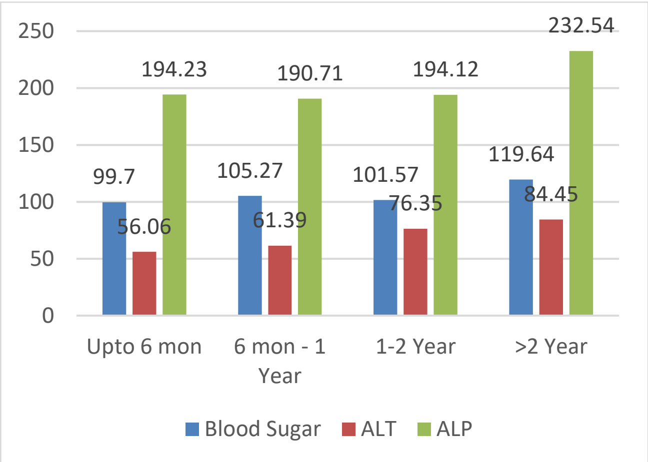
The age wise distribution of dogs with diabetes mellitus were as follows: 3 (10.34%) dogs with diabetes mellitus aged <6 months, 7 (20%) aged 6 months–1 year, 14 (27.45%) aged > 1–2 years and 19 (29.23%) over the age of 2 years. The prevalence of diabetes mellitus in dogs with age >2 years, increased non-significantly (P > 0.05) having 95% C.I limits (19.58-41.20), from the dogs with age >1–2 years having 95% C.I limits (17.11–40.95), dogs with age 6 months–1 year having 95% C.I limits

(10.04–35.89) and dogs with age < 6 months having 95% C.I limits (3.58–26.38) (Table 1). Age wise mean values of blood sugar, ALT and ALP were less in dogs with age < 6 months (99.7, 56.06, 194.23) and high in > 2 years of age dogs (119.64, 84.45, 232.54) (Table 2, Figure 6).

Table 2
Mean values of blood sugar, ALT and ALP with respect to different parameters.

Parameters	Category	Blood Sugar	ALT	ALP
Sex	Male	96.4	62.24	239.02
	Female	127.78	92.95	264.83
Age	Up-to 6 Month	99.7	56.06	194.23
	6 Month - 1 Year	105.27	61.39	190.71
	1-2 Year	101.57	76.35	194.12
	>2 Year	119.64	84.45	232.54
Body Weight	Up-to 15 kg	101.6	58.56	218.57
	15-30 kg	105	70.38	214.29
	30-45 kg	112.63	74.49	191.88
	> 45 kg	134	125.61	248.28
BCS	Up-to 2 BCS	125.11	72.3	286.82
	3-4 BCS	97.56	68.56	223.97
	5-6 BCS	118.89	75.82	251.81
	> 6 BCS	145.66	142.93	268.53
Neutering Status	Spay	113	92.77	279.64
	Neuter	88.26	49.4	194.9
	Intact	116.35	80.55	254.43
Diet	Dog Food	92.87	70.5	211.71
	Homemade Food	115.93	77.81	259.97
	Mixed Food	105.8	66.04	229.59
Vaccination Status	Regular Vaccinated	104.51	73.77	214.74
	Irregular Vaccination	125.64	78.31	383.27
	Not Vaccinated	108.25	40.92	276.97
Breed	German Shepherd	120.45	85.96	254.85
	Husky	102.14	67.82	392.61
	Labrador	115.8	63.18	142.62
	Golden Retriever	119.12	93.15	380.47
	Rottweiler	115.57	58.27	130.88
	Black Shepherd	99.5	66.4	232.81
	Terrier	106	68	187.15
	Mixed Breed	104.72	54.6	233.34
Glucometer	Accu-Chek	109.39	78.93	260.73
	EasyGluco	108.05	75.67	241.66
	D-eck	108.75	57.85	221.3

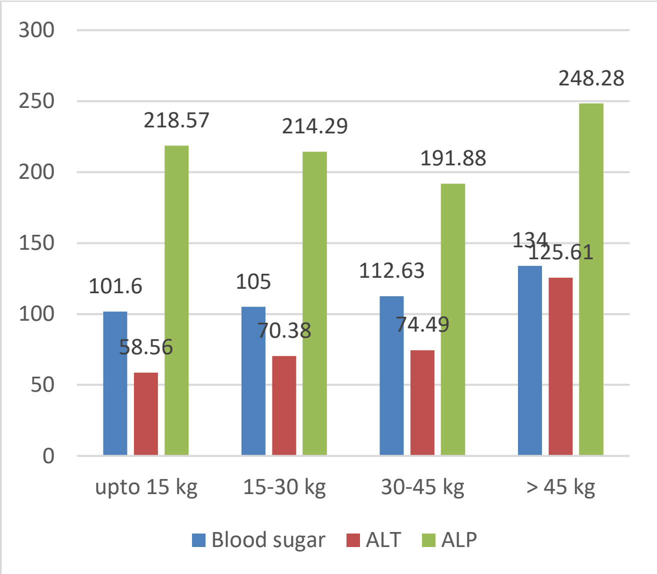
Figure 6: *Age wise prevalence of diabetes mellitus with relation to ALT and ALP*



Body Weight Wise Prevalence

The body weight wise distribution of dogs with diabetes mellitus were as follows: 2 (6.25%) dogs with up to 15 kg body weight, 9 (17%) dogs with 15–30 kg body weight, 11 (23.40%) dogs with 30–45 kg body weight and 21 (42%) dogs with > 45 kg body weight. The occurrence of diabetes mellitus in dogs with > 45 kg of body weight, increased non-significantly (P = 0.0237) having 95% C.I limits (29.38-55.77), from the dogs with 30-45 kg body weight having 95% C.I limits (13.60–37.22), dogs with 15–30 kg body weight having 95% C.I limits (9.57–30.26) and dogs with < 15 kg body weight having 95% C.I limits (1.73–20.15) (Table 1). Dogs weighing > 45 kg were at an increased risk of diabetes mellitus compared with dogs weighing < 15 kg (P = 0.0237). Body weight wise mean values of blood sugar, ALT and ALP were less in dogs with < 15 kg body weight (101.6, 58.56, 218.57) and high in > 45 kg of body weight dogs (134, 125.61, 248.28) (Table 2, Figure 7).

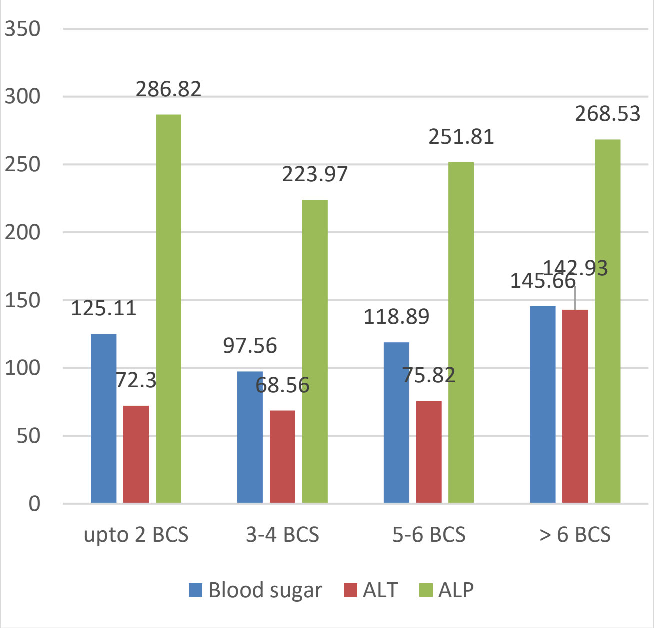
Figure 7: *Body weight wise prevalence of diabetes mellitus with relation to ALT and ALP*



Body Condition Score Wise Prevalence

The body condition score wise distribution of dogs with diabetes mellitus were as follows: 4 (8.16%) dogs with score up to 2, 5 (23.80%) dogs with score 3-4, 15 (28.30%) dogs with score 5-6 and 19 (33.33%) dogs with > 6 body condition score. The prevalence of diabetes mellitus in dogs with > 6 body condition score, increased non-significantly (P = 0.0849) having 95% C.I limits (22.49–46.27), from the dogs with score 5-6 having 95% C.I limits (17.96–41.57), dogs with score 3-4 having 95% C.I limits (10.63–45.09) and dogs with up to 2 body condition score having 95% C.I limits (3.22–19.18) (Table 1). Body condition score wise mean values of blood sugar, ALT and ALP were less in dogs with score 3-4 (97.56, 68.56, 223.97) and high in dogs with > 6 score (145.66, 142.93, 268.53) (Table 2, Figure 8).

Figure 8: *Body condition score wise prevalence of diabetes mellitus with relation to ALT and ALP*



Neutering Status Wise Prevalence

The neutering status wise distribution of dogs with diabetes mellitus were as follows: 23 (35.93%) female spayed dogs, 9 (11.68%) male neutered dogs and 11 (28.20%) intact male and female dogs. The prevalence of diabetes mellitus in spayed dogs, increased non-significantly (P = 0.0248) having 95% C.I limits (25.29-48.18), from the intact dogs having 95% C.I limits (16.55-43.78) and neuter dogs having 95% C.I limits (6.27-20.75) (Table 1). Intact male dogs were at a significantly greater risk (odd ratio = 2.41) than neutered male dogs, but there was no significant difference in risk between neutered and intact females (P = 0.0248). Neutering status wise mean values of blood sugar, ALT and ALP were less in neuter dogs (88.26, 49.4, 194.9) and high in intact dogs (166.35, 80.55, 254.43) (Table 2, Figure 9).

Figure 9: *Neutering status wise prevalence of diabetes mellitus with relation to ALT and ALP*

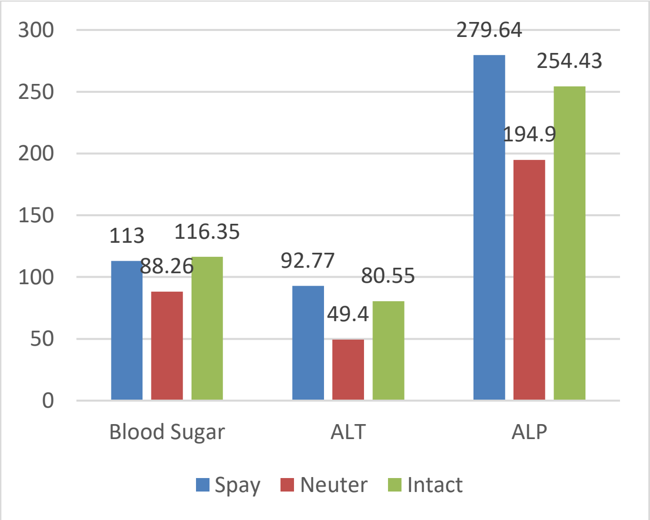


Figure 10: *Diet wise prevalence of diabetes mellitus with relation to ALT and ALP*

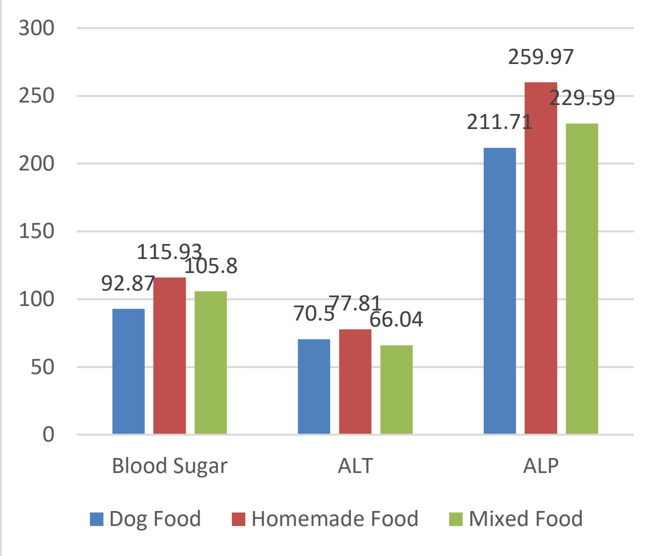
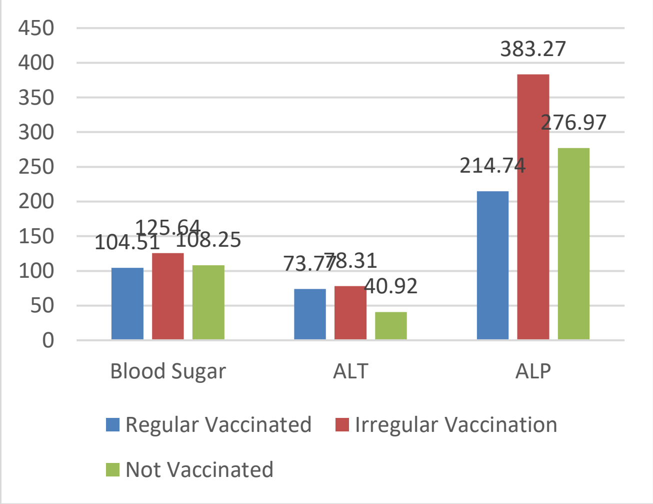


Figure 11: *Vaccination status wise prevalence of diabetes mellitus with relation to ALT and ALP*



Breed Wise Prevalence

The breed wise distribution of dogs with diabetes mellitus were as follows: 13 (31.70%) Germen shepherds, 7 (30.43%) Labrador, 6 (31.57%) Golden Retriever, 9 (436%) Rottweiler, 2 (12.5%) Black shepherd, 2 (13.33%) Terrier, 1 (4.76%) Husky and 3 (15%) mixed breeds. The prevalence of diabetes mellitus in Rottweiler dogs, increased non-significantly (P = 0.3660) having 95% C.I limits (20.25-55.48), from the Germen Shepherd dogs having 95% C.I limits (19.57-46.99), Golden Retriever dogs having 95% C.I limits (15.37-53.99), Labrador dogs having 95% C.I limits (15.60-50.86), mixed breeds having 95% C.I limits (5.24-36.04), Terrier dogs having 95% C.I limits (3.73-37.88), Black Shepherd dogs having 95% C.I limits (3.50-36.02) and husky dogs having 95% C.I limits (0.85-22.67) (Table 1). The breed at highest risk of diabetes mellitus was the Rottweiler (odds ratio = 3.00) while the Black shepherd breed had the lowest risk (odds ratio = 7.56). Breed wise mean values of blood sugar, ALT and ALP were less in Black shepherd dogs (99.5, 66.4, 232.81) and high in Germen Shepherd dogs (120.45, 85.96, 254.85) (Table 2, Figure 12, 13).

Figure 12: *Breed wise prevalence of diabetes mellitus with relation to ALT and ALP*

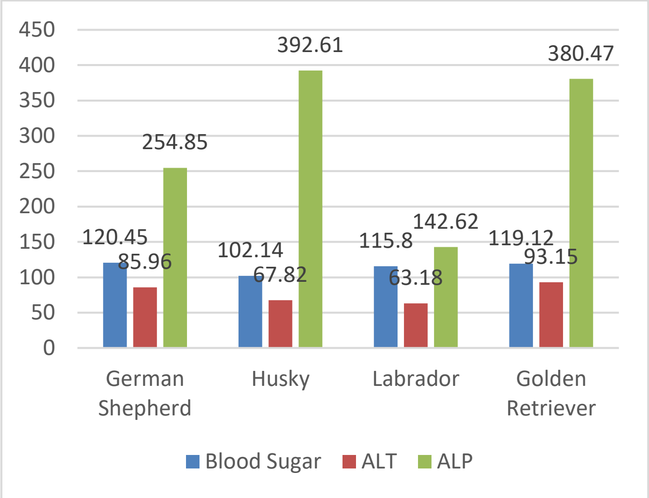


Figure 13: *Breed wise prevalence of diabetes mellitus with relation to ALT and ALP*

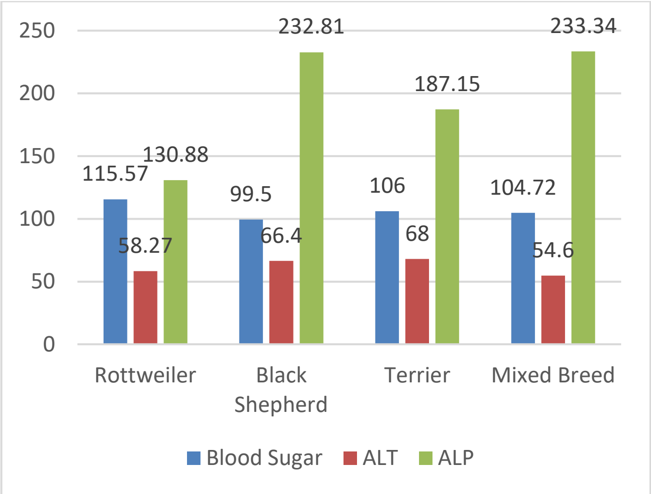
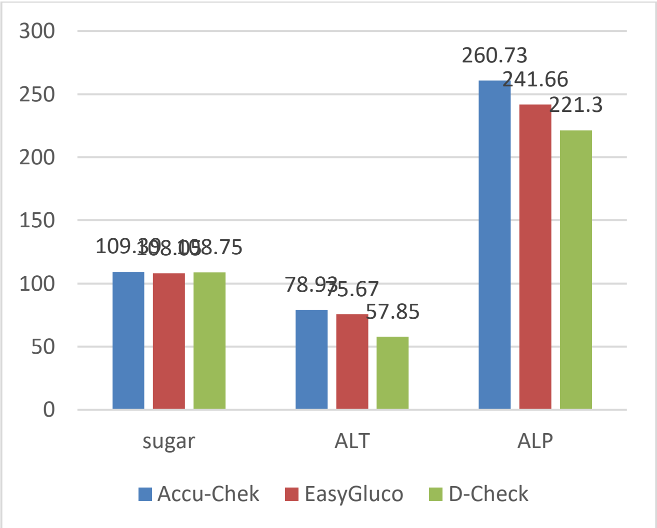


Figure 14: *Glucometer wise prevalence of diabetes mellitus with relation to ALT and ALP*



DISCUSSION

The One Health approach operates efficiently through an organized reporting mechanism, enabling the real-time tracking and communication of health events in the domains of animals, humans, and science-policy. This facilitates the promotion of comprehensive health initiatives. In Pakistan, there exists a blended healthcare system where both public and private sectors collaborate to deliver healthcare services to animals (Afzal *et al.*, 2022). Epidemiological analysis of a group of diabetic dogs determined that age, gender and breed were altogether essential factors accompanying with the presence of diabetes mellitus. The conclusions of this study also aided to emphasize the fraction of diabetic dogs which were young (20% were less than 6 months) and the composite correlation between age and gender related with the illness. As a whole prevalence of diabetes mellitus in the current reading were 30% and 39.28% for dogs aged more than 2 years, and therefore may be indirectly comparable to the lesser prevalence values of 0.32 to 0.36% reported by other UK and Australian studies measuring the total population dogs within a primary-care setting (Catchpole *et al.*, 2005; Mattin *et al.*, 2014; Yoon *et al.*, 2020). Canine diabetes mellitus has a stated prevalence of 0.26 to 1.33% in the Sweden, USA, UK and Italy (Guptill *et al.*, 2003; Mattin *et al.*, 2014; Heeley *et al.*, 2020). A number of studies have described a higher prevalence of diabetes mellitus in specific dog breeds, although other breeds express an apparent safety from diabetes mellitus (Fall *et al.*, 2007; Catchpole *et al.*, 2013), provided that suggestion for a considerable genetic component to disease vulnerability. Geneti risk for the illness reveals greater risk for a specific canine (e.g. golden retrievers and Labradors) (German, 2006; Lund *et al.*, 2006; Corbee, 2013). In the present study, diabetes mellitus were more in pure breeds like Rottweiler (42.85%), Labrador (40%), Golden Retriever (37.5%) and Germen Shepherd (31.81%). Some pure breed dogs were at lower risk for diabetes mellitus which includes Black Shepherd (16.6%) and Husky (14.28%). While mixed breed dogs (27.27%) were in between the both higher and lower risk pure breed dogs. Further risk factors include sex, neutering position and middle age. Middle-aged neutered female dogs have more chances to be overweight or obese (McGreevy *et al.*, 2005; German, 2006; Laflamme, 2006; Lund *et al.*, 2006). In the current study, female dogs were more prevalent (50%) than male dogs (16.66%). On the basis of age, diabetes mellitus were more in dogs with age > 2 years (39.28%) and less in dogs with age < 6 months (20%). Neutered female dogs were at high risk (44.4%) as compared to the neutered male dogs (10.52%) while intact male dogs were at much higher risk (35.71%) than neuter male dogs but intact female dogs were considerably less at risk than neuter females. In dogs, decreased daily workout has been correlated with obesity (Bland *et al.*, 2009; Courcier *et al.*, 2010). The population prevalence of overweight and obese has been expected to be 19.7 to 59.3% (McGreevy *et al.*, 2005; Lund *et al.*, 2006; Hill, 2009; Courcier *et al.*, 2010; Corbee, 2013; Mao *et al.*, 2013). In the present study, the prevalence of diabetes mellitus were higher in dogs with body weight > 45 kg (50%) than the dogs with body weight <15 kg (20%). Similarly, dogs with > 6 body condition score were at higher risk (66.6%) than dogs with < 2 body condition score (44.44%). Although the prevalence of diabetes mellitus were less in 3-4 body condition score dogs (20.51%), while there was non-significant difference between dogs with 5-6 body condition score (36.84%) and dogs with < 2 body condition score (44.44%). Diseases reported simultaneously with 0verweight or 0besity include diabetes mellitus, endocrine abnormalities (e.g. hypothyroidism, hyperadrenocorticism), arthritis, ligaments rupture like cruciate ligament rupture, lower urinary tract infection, 0ral infections, pancreatitis and neoplasms (Laflamme, 2006; Lund *et al.*, 2006; Marshall *et al.*, 2009).

High-fat nutrition is linked with overweight or obesity in dogs (Laflamme, 2006). Metabolic dysfunction leads to various liver disorders, including nonalcoholic fatty liver disease (NAFLD). Insulin resistance triggered by inflammation plays a pivotal role in NAFLD development, characterized by elevated insulin and blood sugar levels. The rising incidence of obesity mirrors the growing prevalence of NAFLD (Ishtiaq *et al.*, 2022). The basis of food (commercially primed or home-made) was not an important feature for canine obesity in some studies, but a non-commercial source of food was a risk factor in a different study (Mao *et al.*, 2013). In our study, diabetes mellitus were more in dogs which were on homemade diet (34.37%) and dogs which were on dog food are less prevalent (12.5%). While dogs which were on mixed food showed prevalence (30%) in between the both, more and less prevalent dogs. The price of pet foodstuff has been found to be correlated with 0besity; possessors of obese dogs are more likely to point out the significance of inexpensive foods. Obesity has also been associated with the number and quantity of meals and snacks served and feeding of kitchen/table scraps, fresh meat and commercial delights for dogs (Bland *et al.*, 2009; Sallander *et al.*, 2010).

Almost all diabetic dogs have type-1 diabetes mellitus and were insulin reliant while diagnosed. The percentage of diagnosis of diabetes mellitus in dogs is about 0.6% (Guptill *et al.*, 2003). A latest study of American Eskimo dogs used a logistic regression model to evaluate a heritability of 0.62 for DM in this specific breed (Cai *et al.*, 2019) and following complex segregation analysis proposed a polygenic means of inheritance. In case of dogs, there was an enormously extensive spectrum of exposure for a single species, when comparing breeds at more risk, such as the Samoyed (odds ratio = 35.84), and those at low risk, such as the Boxer (odds ratio = 0.07) (Catchpole *et al.*, 2013). This recommends that there was a variation in the biological manners of pancreatic beta cells in the dog population, with breeds at the extremes of the range at high risk of either insulin deficiency (beta cell death) or excess (beta cell malignant transformation). Diabetes mellitus in dogs was usually diagnosed between 5 and 12 years of age (Guptill *et al.*, 2003). Diagnosis of canine diabetes mellitus was made depending upon hyperglycemia, usually along with glycosuria and associated clinical signs of polyuria, polydipsia, polyphagia and weight loss (Behrend *et al.*, 2018). A few number of juvenile-onset diabetes cases (i.e. with onset in dogs less than 12 months of age) have been reported, with more prevalence in some breeds, like Labrador retrievers (Catchpole *et al.*, 2008; Alvarez *et al.*, 2015), although the genetic origin of this disorder has not yet been well-known.

CONCLUSIONS

This study is conducted first time in Pakistan and would be helpful in future for further testing related to diabetes mellitus in dogs. The portable glucose meter examined is an effective, safe, accurate, and financially feasible alternative for glycaemia measures in dogs, either in hypoglycemia, normoglycemia, or hyperglycemia conditions. Some benefits of portable glucose meter over enzymatic method include its simplicity, applicability, and ability to do field testing for rapid diagnosis, prevention, and monitoring of disorders that change glycaemia.

Ethical Responsibility

The study pertained to M. Phil thesis duly approved from the department of Clinical Medicine and Surgery, University of Agriculture, Faisalabad.

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