The Pharma Innovation Journal 2022; 11(6): 2410-2415

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; 11(6): 2410-2415

www.thepharmajournal.com
Received: 09-04-2022
Accepted: 12-05-2022

Abarna K
Department of Veterinary Biochemistry, Madras Veterinary College, Vepery, Chennai, Tamil Nadu, India

Pandiyan V
Department of Veterinary Biochemistry, Madras Veterinary College, Vepery, Chennai, Tamil Nadu, India

Parthiban M
Department of Biotechnology, Madras Veterinary College, Vepery, Chennai, Tamil Nadu, India

Balagangathara Thilagar M
Department of Veterinary Clinical Medicine, Madras Veterinary College, Vepery, Chennai, Tamil Nadu, India

K Padmanath
Department of Veterinary Biochemistry, Madras Veterinary College, Vepery, Chennai, Tamil Nadu, India

Corresponding Author:
K Padmanath
Department of Veterinary Biochemistry, Madras Veterinary College, Vepery, Chennai, Tamil Nadu, India

Hematological and biochemical changes in diabetic dogs supplemented with mustard oil

Abarna K, Pandiyan V, Parthiban M, Balagangathara Thilagar M and K Padmanath

Abstract
The present experiment was carried out to study the effect oral administration of mustard oil in insulin treated diabetic dogs. Dogs visiting Small Animal Clinics, were taken for the study. Six healthy dogs were treated as group-I. Six diabetic dogs treated with insulin were treated as group-II. Six diabetic dogs treated with insulin along with oral administration of mustard oil were treated as group-III. Dogs in group-II and III were treated daily with insulin at the dose rate of 0.2 to 1 U/kg body weight by subcutaneous route once. Group-III dogs were administered orally with mustard oil at the dose rate of 0.25g per kg body weight. The study was carried out for a period of sixty days. Blood samples collected were used for haematological and biochemical assays. There was no significant change in the hemogram and leukogram values, except platelet count which was increased significantly in treatment groups. There was a reduction in blood glucose, HbA1c level and increase in body weight in mustard oil treated diabetic dogs. A significant increase in serum total cholesterol and HDL-cholesterol level were noticed in mustard oil treated groups. There was no significant difference in the levels of total protein, albumin and globulin among the groups. Administration of mustard oil significantly reduced the serum BUN and creatinine level in diabetic dogs treated with insulin and mustard oil. This indicates the protective role of the mustard oil in kidney function, due to the anti-hyperglycemic activity. There was no significant change in the serum electrolytes values except serum calcium level which was a significant reduced in mustard oil treated group, which may be due to precipitation of calcium by excess fatty acids present. The findings suggest that mustard oil supplementation can improve the glycemic control in diabetic dogs which may be due to mono and polyunsaturated fatty acids present.

Keywords: Canine diabetes, mustard oil, serum glucose and HbA1c

Introduction
Diabetes mellitus (DM) is a metabolic disorder characterized by chronic hyperglycemia stemming from defects in insulin secretion, insulin action, or both, affecting carbohydrate, fat, and protein metabolism. This condition leads to long-term damage, dysfunction, and organ failure (WHO, 1999) [38].

Diabetes can affect various animal species, including companion animals like dogs and cats. In dogs, diabetes mellitus is a common metabolic disorder prevalent in middle-aged to geriatric animals. It is marked by hyperglycemia, glucosuria, and weight loss due to either absolute or relative insulin deficiency. The incidence of spontaneous cases of canine DM has increased globally, influenced by factors such as obesity, changes in feeding practices, lack of owner attention, insufficient awareness, and advancements in diagnostic techniques. In India, there is limited research on breed predisposition, incidence rates, susceptibility factors, clinical characteristics, and responses to available interventions for managing and controlling diabetes in canine populations. The estimated prevalence of canine diabetes is around 0.231% (Prathaban, 1990) [29], with approximately 1 in 500 dogs and 1 in 250 house cats developing the disease (Catchpole *et al*, 2005; McCann *et al.*, 2007) [24, 11].

Insulin deficiency diabetes (IDD) is the predominant form of diabetes in dogs, characterized by pancreatic beta cell destruction resulting in absolute insulin deficiency (Montgomery *et al.*, 1996) [25]. Primary IDD in dogs involves a progressive loss of pancreatic beta cells. Insulin resistance diabetes (IRD), induced by states of insulin resistance such as hyperadrenocorticism and acromegaly, is a less common type. Iatrogenic insulin resistance can also lead to diabetes, often caused by chronic corticosteroid therapy (Catchpole *et al*, 2005) [11]. Numerous studies have demonstrated that diet can influence the fatty acid composition of cell membranes in humans and other animals.

Changes induced by dietary fats in membrane composition affect membrane-associated protein functions (Abbott *et al*, 2010) [1]. The quality of dietary fats, primarily from vegetable oils, significantly impacts cell membrane fatty acid composition and functions such as membrane fluidity, ion permeability, insulin receptor binding/affinity, and interactions of glucose transporters with second messengers (Ginsberg *et al.*, 1981; Storlien *et al.*, 1996) [16, 36]. Vegetable oils in diets have been implicated in enhancing insulin sensitivity and reducing the risk of diabetes and its complications.

Mustard oil, a traditional oil widely used in India and Bangladesh for centuries, is renowned for its medicinal properties (Dasgupta and Bhattacharyya, 2007) [13]. Derived from seeds of *Brassica juncea* and other Cruciferae family members, mustard oil is rich in essential fatty acids like linoleic and linolenic acids, along with essential vitamins. It contains lower amounts of saturated fatty acids compared to unsaturated fatty acids (Altaf *et al.*, 2013) [3]. Long-term consumption of mustard oil has been associated with various health benefits such as preventing dyslipidemia, coronary artery disease, atherosclerosis, and colon cancer (Degirolamo *et al*, 2010) [15]. Studies have shown that aqueous extracts of mustard oil significantly reduce blood glucose levels (Yadav *et al*, 2004) [40] in streptozotocin-induced diabetic rats at a dosage of 200 mg/kg body weight (Anand *et al*, 2007) [5].

Research in rat models has indicated that dietary incorporation of mustard oil enhances insulin secretion and reduces blood glucose levels. It also up-regulates the expression of the glucose transporter (GLUT4) gene by 8.456-fold compared to diabetic control groups (Sukanya *et al.*, 2020) [37]. Another study reported that mustard oil supplementation activates the insulin receptor signaling pathway, further lowering blood glucose levels in rats (Anjali Devi *et al.*, 2022) [6]. However, similar studies in dogs have not been conducted to date. Therefore, this study aimed to investigate, for the first time, the effects of mustard oil on hematological and serum biochemical parameters in diabetic dogs.

Materials and Method

Materials

Diagnostic kits for biochemical assays were procured from M/s Agappe Diagnostics, Ernakulam, Kerala, India. Insulin (Human mixtard) was obtained from a local pharmacy, and mustard oil was purchased from a local market.

Methods

Experimental Animals

Animals presenting clinical signs such as polyuria, polydipsia, obesity, bilateral rapidly developing cataracts, rapid weight loss, pyometra, mammary tumor, non-responsive dermatoses, senility, and those in diestrus were screened for diabetes mellitus (DM) using a glucometer (OneTouch) at the Small Animal Out-patient Unit, Madras Veterinary College Hospital. Animals exhibiting hyperglycemia and suggestive symptoms of diabetes were further subjected to serum biochemical analysis and HbA1c estimation. Dogs diagnosed with diabetes mellitus were included in the study (Table-I).

Experimental Design

Healthy dogs brought to the clinics for routine health check-ups or vaccinations comprised group-I. Diabetic dogs in group-II received insulin (Human mixtard) subcutaneously once daily at a dose of 0.2 to 1.0 U/kg body weight. Diabetic dogs in group-III were treated daily with the same insulin regimen as group-II, along with oral administration of mustard oil at a dose of 0.25 gm per kg body weight for sixty days.

Determination of Hematological and Biochemical Parameters

Body weights were recorded during each clinic visit. Blood samples were collected via venepuncture of the saphenous or cephalic vein after the sixty-day experimental period. Whole blood samples were collected in vacutainers coated with 10% ethylenediaminetetraacetic acid (EDTA) and immediately used for blood counts. Serum samples were obtained by centrifugation of part of the blood at 3000 rpm for 15 minutes and stored at-20 °C until analysis. Serum samples were used for estimating biochemical parameters and fatty acid assays.

Hematology

Hemoglobin concentration (Hb), packed cell volume (PCV), total erythrocyte count (TEC), total leukocyte count (TLC), and platelet count were determined using an automated blood cell analyzer (Mindray, RR 28103439). Differential leukocyte counts (DLC) were performed using smears according to the method described by (Coles, 1986) [12].

Biochemical Parameters

Serum samples were analyzed for biochemical assays including glucose, total protein, albumin, cholesterol, triacylglycerol, low-density lipoprotein (LDL), high-density lipoprotein (HDL), urea, creatinine, aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), lactate dehydrogenase (LDH-P), calcium, phosphorus, sodium, potassium, chloride, and HbA1c using an Auto-analyzer (Biosystems, A15). Serum insulin levels were determined using the Insulin IRMA (Immuno Radio Metric Assay) kit purchased from M/s Anand Brothers, Delhi, India.

Statistical Analysis

Results were presented as mean ± S.E. Statistical analysis was performed using SPSS package version 20. One-way analysis of variance (ANOVA) followed by Duncan's multiple comparison test (Snedecor and Cochran, 1994) was used to analyze the data. Statistical significance was considered at $p < 0.05$ and $p<0.01$.

Results

Body weight

The results of body weight, blood glucose, HbA1c and insulin levels are presented in table-I. There is no significant difference in the body weight between the groups. But, the group-III showed a slight increase in body weight when compared to that of group-II. (Table-II).

Hematological profile

There was no significant difference between the groups with respect to parameters like Hb concentration, PCV, TEC, TLC and DLC (Table-III). However, there was a significant increase in the platelet count in the treatment groups.

The mean percentage of monocytes observed in group-I, group-II and group-III was 15.33±1.72 per cent, 9.67±1.30

per cent and 8.50±1.76 per cent respectively. There was a significant decrease (p<0.05) in the monocytes per cent of group-II dogs when compared to that of group-I and there was a slight decrease in the monocyte per cent in group-III when compared to that of group-II.

Serum biochemical profile

There was no significant difference between the groups with respect to parameters like triglycerides, total protein, albumin, globulin, BUN, creatinine, ALT, AST, ALP, LDH, phosphorus, sodium, potassium and chloride (Table-IV). However, there was a significant decrease in the parameters like serum glucose, BUN and calcium in group-III dogs when compared to group-II dogs. There was a highly significant increase in the HbA1c in group-II and group-III dogs. The HbA1c value in group-III was slightly decreased when compared to group-II.

Discussion

Effect of Mustard Oil on Body Weight

In our study, we observed a decrease in body weight in group-II compared to group-I. This weight loss is a common feature of diabetes, where proteins and fats are metabolized excessively for gluconeogenesis. Diabetic animals experience significant energy loss through glycosuria. Glucose is a major energy source in healthy animals and contributes to weight maintenance. Insulin plays a crucial role in regulating overall protein breakdown in mammals, and its deficiency can accelerate protein catabolism by 30-150% in various tissues (Goldberg and John, 1976) [17]. Therefore, the reduced body weight observed in our study may be attributed to heightened protein and lipid degradation.

Interestingly, we noted a mild increase in body weight in group-III dogs supplemented with mustard oil compared to group-II. Previous studies in diabetic rats have reported significant weight gain with dietary mustard oil supplementation, suggesting a reversal of lipid and protein metabolism and inhibition of gluconeogenesis (Kumar *et al.*, 2013; Ahmad *et al.*, 2014) [22, 2]. Mustard oil is known to reduce proteolysis and lipolysis while enhancing insulin's anabolic effects, promoting protein synthesis and reducing protein breakdown (Murray *et al.*, 1999) [27]. Insulin also suppresses adipose tissue lipolysis, which can further inhibit muscle tissue breakdown (Campbell *et al.*, 1999) [9]. The improvement in body weight observed in group-III may therefore be due to reduced protein degradation.

Effect of mustard oil on hematological parameters

In our study, we observed a significant increase in platelet count in the treatment groups compared to group-I dogs. This finding aligns with previous reports of increased platelet counts in streptozotocin-induced diabetic rats, suggesting enhanced platelet activation and altered coagulation profiles in diabetes (Carr, 2001) [10]. Similar results were reported in rats supplemented with mustard oil, supporting our findings (Khan *et al.*, 2009) [20].

Effect of mustard oil on serum biochemical parameters

We observed a significant increase in blood glucose levels in group-II compared to group-I. Diabetes mellitus disrupts glucose uptake and metabolism due to defects in insulin secretion or action (Koenig *et al.*, 1976) [21]. Despite insulin therapy, group-II dogs showed persistently elevated blood glucose levels, likely reflecting varying degrees of insulin resistance among diabetic dogs. Insulin doses are often adjusted based on individual responses to therapy, aiming to achieve normoglycemia, which remains challenging in clinical practice.

By the 60th day of treatment, dogs in group-III (insulin + mustard oil) exhibited reduced blood glucose levels compared to those in group-II. This reduction may be attributed to mustard oil's ability to enhance insulin sensitivity through its content of unsaturated mono and polyunsaturated fatty acids. Mustard oil has been shown to stimulate insulin secretion, increase insulin receptor signaling, and promote GLUT-4 translocation to the cell membrane for enhanced glucose transport and metabolism, even in insulin-resistant conditions (Sukanya *et al.*, 2020; Anjali Devi *et al.*, 2022; Kumar *et al.*, 2013) [37, 6, 22].

HbA1c serves as a marker of protein glycation in diabetes, reflecting long-term glycemic control. In our study, group-II dogs showed a significant increase in HbA1c levels compared to group-I, indicative of poor glycemic control. In contrast, group-III dogs exhibited lower HbA1c levels than group-II, suggesting the potential anti-diabetic effects of mustard oil. Similar findings have been reported in diabetic rats supplemented with mustard oil, which reduced plasma HbA1c levels (Kumar *et al.*, 2013) [22].

Diabetes mellitus is characterized by chronic hyperglycemia and disruptions in carbohydrate, lipid, and protein metabolism (Saad *et al.*, 2016) [30]. Dyslipidemia is a major risk factor for cardiovascular complications, the leading cause of morbidity and mortality in diabetes and insulin resistance (Davidson, 1981). In our study, lipoprotein analysis of group-II dogs revealed significant increases in total cholesterol (TC) and high-density lipoprotein cholesterol (HDL-C) compared to group-I. Elevated lipid levels are common in diabetes and contribute to coronary heart disease risk (Murali, 2002) [26]. Insulin deficiency is associated with hypercholesterolemia and hypertriglyceridemia, consistent with our results and previous findings in diabetic rat models (Matson and Grundy, 1985) [23].

Interestingly, triglyceride levels did not differ significantly between groups in our study. However, group-III dogs showed significant increases in TC and HDL-C compared to group-II. This contrasts with previous studies reporting reductions in triglycerides and cholesterol levels and improvements in HDL-cholesterol in diabetic rats fed mustard oil-supplemented diets. These effects were attributed to increased insulin secretion and inhibition of lipolysis, mediated by mustard oil's unsaturated fatty acids (Sukanya *et al.*, 2020; Kumar *et al.*, 2013) [37, 22].

Serum electrolyte levels reflect renal function, crucial in diabetes-associated kidney complications. In our study, there were no significant changes in serum phosphorus, sodium, potassium, or chloride levels among groups. However, dogs in groups II and III showed decreased calcium levels compared to group-I. Diabetes can lead to decreased calcium levels due to bone mineral loss and pancreatitis-induced calcium soap formation from fatty acid hydrolysis (Patton and Carey, 1979; Aub *et al.*, 1937) [28, 7]. The type and amount of dietary fats can also influence calcium utilization (Shokeen *et al.*, 2008) [33]. Furthermore, group-II diabetic dogs exhibited significantly elevated serum urea and creatinine levels compared to group-I, indicative of renal dysfunction. Hyperglycemia in diabetes can cause renal damage such as

glomerulonephritis and nephrosclerosis, elevating serum urea and creatinine levels (Ya Chen *et al.*, 2020; Jaramillo, *et al.*, 2008) [39, 19]. In contrast, group-III showed marked decreases in serum urea levels, consistent with previous findings in mustard oil-supplemented rats, suggesting protective effects on kidney function (Sukanya *et al.*, 2020; Anjali Devi *et al.*, 2022) [37, 6].

No significant changes were observed in total protein, albumin, globulin, ALT, AST, ALP, LDH, phosphorus, sodium, potassium, or chloride levels among groups, indicating no adverse effects of oral mustard oil administration.

Conclusion

In conclusion, our study demonstrates that mustard oil supplementation in diabetic dogs treated with insulin can improve glycemic control, as evidenced by reduced blood glucose and HbA1c levels. Mustard oil's potential to enhance insulin sensitivity and protect against diabetes-related complications such as dyslipidemia and renal dysfunction warrants further investigation in long-term trials to validate its therapeutic benefits in diabetic management.

Tables

Table 1: Grouping of the experimental dogs

Groups	Number of dogs
Group-I (Normal control)	6
Group-II (Diabetic animals treated with insulin)	6
Group-III (Diabetic animals treated with insulin and mustard oil)	6

Table 2: Effect of mustard oil on body weight, serum glucose, HbA1c and insulin levels in control and experimental dogs

Parameters	Group-I	Group-II	Group-III
Body weight (kg)	20.50±5.57	17.00±3.08	21.42±4.29
Serum glucose *(mg/dL)	90.66^{b}±4.43	220.50^{a}±20.00	196^{a}±14.10
HbA1c **(%)	2.66 ^{a} ±0.09	6.050 ^{b} ±0.08	5.88 ^{b}±0.09
Insulin (µIU/ml)	1.57 ^{b} ±0.09	0.52 ^{a} ±0.04	0.65 ^{a}±0.01

Table 3: Effect of mustard oil on hematological profile in control and experimental groups.

Parameters	Group-I	Group-II	Group-III
Hb (g/dL)	12.80 ±0.90	13.93 ±0.79	14.23 ±1.30
PCV (%)	34.50 ±1.81	37.63 ±1.33	37.56 ±3.91
RBC (m/cmm)	5.20 ±0.47	6.35 ±0.14	5.76 ±0.54
Platelets */cmm	167500 ^{b} ±5457	192783.33 ^{ab}±30082	280333.33 ^{a} ±41280
WBC/cmm	12083.33 ±168	14700.00 ±2097	10816.67 ±841
Neutrophils (%)	74.00 ±4.22	75.67 ±2.10	72.67 ±2.87
Lymphocytes (%)	15.33 ±1.72	13.00 ±2.43	17.17 ±1.13
Monocytes *(%)	15.33 ^{a} ±1.72	9.67 ^{b} ±1.30	8.50 ^{b} ±1.76
Eosinophils (%)	4.00 ±1.31	1.67 ±.84	1.83 ±1.24

**-Highly significant (p<0.01), *-Significant (p<0.05) and ^{NS}-Not significant
Means bearing different superscripts in a row differ significantly between groups

Table 4: Effect of mustard oil on serum biochemical profile in control and experimental groups.

Parameters	Group-I	Group-II	Group-III
Serum total protein (g/dL)	6.90±0.50	6.88±0.33	6.48 ±0.32
Serum albumin (g/dL)	2.57±0.19	2.82±0.18	3.17±0.14
Serum globulin (g/dL)	4.33±0.34	4.06±0.34	3.30±0.20
Total cholesterol **(mg/dL)	121.33 ^{a} ±14.82	213.83 ^{b} ±9.50	357 ^{c} ±20.81
Triglycerides (mg/dL)	111.83±9.90	121.67±39.88	156.17 ±8.88
LDL-cholesterol**(mg/dL)	28.77 ^{a} ±4.92	58.60 ^{b} ±3.04	62.17 ^{b} ±4.69
HDL-cholesterol **(mg/dL)	44.56 ^{a} ±6.00	68.50 ^{a} ±17.94	114.00 ^{b} ±14.33
ALT (U/L)	63.33±12.31	41.33±7.00	52.42±0.11
AST (U/L)	62.50±16.65	52.00±2.54	48.30±0.99
ALP** (U/L)	34.17 ^{a} ±4.71	234.50 ^{b} ±64.10	221.83 ^{b}±4.24
LDH (U/L)	40.70 ±4.32	103.11±38.78	89.41±0.13
Calcium *(mg/dL)	10.80 ^{b} ±0.44	9.93 ^{ab} ±0.91	8.29 ^{a} ±0.34
Phosphorus (mg/dL)	4.05±0.55	4.68±0.45	4.38±0.29
Sodium (mEq/L)	146.57 ±2.74	143.20 ±2.15	142.63 ±1.37
Potassium (mEq/L)	4.90±0.18	4.90 ±0.23	4.77±0.01
Chloride (mEq/L)	110.70±1.74	110.08±2.01	109.80±1.02
BUN *(mg/dL)	22.35 ^{a} ±2.01	52.41 ^{b} ±7.18	40.36 ^{ab} ±8.94
Creatinine (mg/dL)	0.86±0.10	2.04±0.89	1.07±0.11

**-Highly significant (p<0.01), *-Significant (p<0.05) and ^{NS}-Not significant
Means bearing different superscripts in a row differ significantly between groups

Acknowledgement/S

The authors would like to express their sincere gratitude to Tamil Nadu Veterinary and Animal Sciences University for funding this research project and for providing the necessary facilities and resources.

References

1. Abbott SK, Else PL, Hulbert AJ. Membrane fatty acid composition of rat skeletal muscle is most responsive to the balance of dietary n-3 and n-6 PUFA. Br J Nutr. 2010;103:522-9.

2. Ahmad M, Prawz S, Sultana M, Raina R, Pankaj NK, Verma PK, *et al*. Anti-hyperglycemic, anti-hyperlipidic and antioxidant potential of alcoholic extract of Sida cordifolia (areal part) in streptozotocin-induced diabetes in Wistar rats. Proc Natl Acad Sci India Sect B Biol Sci. 2014;84(2):397-405.
3. Altaf K, Sankhyan P, Kumar S. Biochemical characterization of mustard oil (*Brassica campestris* L.) with special reference to its fatty acid composition. Asian J Adv Basic Sci. 2013;1(1):1-9.
4. American Diabetes Association. Diagnosis and classification of diabetes mellitus. Diabetes Care. 2013;36(1)
5. Anand P, Murali KY, Tandan V, Chandra R, Murthy S. Preliminary studies on antihyperglycemic effect of aqueous extracts of *Brassica nigra* (L) Koch in streptozotocin-induced diabetic rats. Indian J Exp Biol. 2007;45:696-701.
6. Anjali Devi P, Pandiyan V, Kumar TMA, Padmanath K. Dietary supplementation of mustard oil reduces blood glucose levels by triggering insulin receptor signaling pathway. Int J Diabetes Dev Ctries. 2022;42:126-37.
7. Aub JC, Tibbetts DM, McLean R. The influence of parathyroid hormone, urea, sodium chloride, fat and of intestinal activity upon calcium balance. J Nutr. 1937;13:635-55.
8. Bairati I, Roy L, Mayer F. Effect of fish oil supplement on blood pressure and serum lipids in patients treated for coronary artery disease. Can J Cardiol. 1992;8:416.
9. Campbell SM, Roland MO, Shekelle PG, Cantrill JA, Buetow SA, Cragg DK. Development of review criteria for assessing the quality of management of stable angina, adult asthma, and non-insulin dependent diabetes mellitus in general practice. BMJ Qual Saf. 1999;8(1):6-15.
10. Carr ME. Diabetes mellitus: a hypercoagulable state. J Diabet Complications. 2001;15(1):44-54.
11. Catchpole B, Ristic JM, Fleeman LM, Davison LJ. Canine diabetes mellitus: can old dogs teach us new tricks? Diabetologia. 2005;48(10):1948-56.
12. Coles EH. Veterinary Clinical Pathology. 4th ed. Philadelphia: W.B. Saunders Company; c1986. p. 77-8.
13. Dasgupta S, Bhattacharyya DK. Dietary effect of gamma-linolenic acid on the lipid profile of rat fed erucic acid-rich oil. J Oleo Sci. 2007;56(11):569-77.
14. Davidson MB. Diabetes mellitus: diagnosis and treatment. New York: Wiley. 1981;1:109.
15. Degirolamo C, Kelly KL, Wilson MD, Rudel LL. Dietary n-3 LCPUFA from fish oil but not alpha-linolenic acid-derived LCPUFA confers atheroprotection in mice. J Lipid Res. 2010;51:1897-905.
16. Ginsberg BH, Brown TJ, Simon I, Spector AA. Effect of the membrane lipid environment on the properties of insulin receptors. Diabetes. 1981;30:773-80.
17. Goldberg AL, St John AC. Intracellular protein degradation in mammalian and bacterial cells. Annu Rev Biochem. 1976;45(1):747-804.
18. Hough S, Avioli AV. Alteration of bone and mineral metabolism in diabetes. In: Nattrass M, Santiago JV, editors. Recent advances in diabetes. London: Churchill Livingstone; c1984, p. 223-9.
19. Juarez JF, Vaquez RML, Sanchez RAR, Martinez MC, Ortiz GG. Acute renal failure induced by carbon tetrachloride in rats with hepatic cirrhosis. Ann Hepatol. 2008;7(4):331-8.
20. Khan A, Zaman G, Anderson RA. Bay leaves improve glucose and lipid profile of people with type 2 diabetes. J Clin Biochem Nutr. 2009;44(1):52-6.
21. Koenig RJ, Peterson CM, Jones RL, Saudek C, Lehrman M, Cerami A. Correlation of glucose regulation and hemoglobin A1c in diabetes mellitus. N Engl J Med. 1976;295(8):417-20.
22. Kumar V, Ahmed D, Gupta PS, Anwar F, Mujeeb M. Anti-diabetic, anti-oxidant and antihyperlipidemic activities of *Melastoma malabathricum* Linn leaves in streptozotocin-induced diabetic rats. BMC Complement Altern Med. 2013;13:222.
23. Matson FH, Grundy SM. Comparison effect of dietary saturated, monounsaturated and polyunsaturated fatty acids on plasma lipids and lipoproteins in man. J Lipid Res. 1985;25:194-202.
24. McCann TM, Simpson KE, Shaw DJ, Butt JA, Moore DAG. Feline diabetes mellitus in the UK: The prevalence within an insured cat population and a questionnaire-based putative risk factor analysis. J Feline Med Surg. 2007;9(4):289-99.
25. Montgomery TM, Nelson RW, Feldman EC, Robertson K, Polonsky KS. Basal and glucagon-stimulated plasma C-peptide concentrations in healthy dogs, dogs with diabetes mellitus, and dogs with hyperadrenocorticism. J Vet Intern Med. 1996;10(3):116-22.
26. Murali B, Upadhyaya UM, Goyal RK. Effect of chronic treatment with Enicosistemma littorale in non-insulin-dependent diabetic (NIDDM) rats. J Ethnopharmacol. 2002;81:199-204.
27. Murray RR, Granner DK, Mayes PA, Rodwell VW. Harper's Biochemistry. 25th Ed. Stamford: Appleton and Lange; 1999, 610-7.
28. Paton JS, Carey MC. Watching fat digestion. Science. 1979;204(4389):145-8.
29. Prathaban S. Study on clinical and clinicopathological aspects of canine diabetes mellitus. Ph.D. dissertation, Tamil Nadu Veterinary Animal Sciences University, Chennai; c1990.
30. Saad MJA, Santos A, Prada PO. Linking gut microbiota and inflammation to obesity and insulin resistance. Physiology. 2016;31(4):283-93.
31. Sengupta A, Ghosh M. Modulation of platelet aggregation, haematological and histological parameters by structured lipids on hypercholesterolaemic rats. Lipids. 2010;45(5):393-400.
32. Sheela CG, Augusti KT. Antidiabetic effects of S-allyl cysteine sulphoxide isolated from garlic Allium sativum Linn. Indian J Exp Biol. 1992;30(6):523-6.
33. Shokeen P, Anand P, Murali YK, Tandon V. Antidiabetic activity of 50% ethanolic extracts of Ricinus communis and its purified fractions. Food Chem Toxicol. 2008;46(11):3458-66.
34. Snedecor GW, Cochran WG. Statistical method. J Educ Behav Stat. 1994;19(3):304-7.
35. Stambe C, Atkins RC, Tesch GH, Kapoun AM, Hill PA, Schreiner GF, Paterson NDJ. Blockade of p38 (α) MAPK ameliorates acute inflammatory renal injury in rat anti-GBM glomerulonephritis. Clin J Am Soc Nephrol. 2003;14(2):338-51.
36. Storlien LH, Pan DA, Kriketos AD, O′Connor J, Caterson ID, Cooney GJ *et al*. Skeletal muscle membrane

lipid and insulin resistance. Lipids. 1996, 31
37. Sukanya V, Pandiyan V, Vijayarani K, Padmanath K. A study on insulin levels and the expression of Glut 4 in streptozotocin (STZ) induced diabetic rats treated with mustard oil diet. Indian J Clin Biochem. 2020;35:488-96.
38. World Health Organization (Expert Committee). Definition, diagnosis and classification of diabetes mellitus and its complications. Report of a WHO consultation, Part 1: Diagnosis and classification of diabetes mellitus. Geneva: WHO, 1999.
39. Chen Y, Lee K, Ni Z, He JC. Diabetic kidney disease: challenges, advances, and opportunities. Kidney Dis. 2020;6(4):215-25.
40. Yadav SP, Vasta V, Ammini AC, Grover JK. *Brassica juncea* (Rai) significantly prevented the development of insulin resistance in rats fed fructose-enriched diet. J Ethnopharmacol. 2004;93:113-6.