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ORIGINAL ARTICLE

EVALUATION OF CARNITINE AND LDH LEVELS IN PATIENTS WITH TYPE 2 DIABETES MELLITUS AND ITS COMPLICATIONS

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Summary

Background: Type 2 diabetes is a common condition that causes the level of sugar (glucose) in the blood to become too high. It can cause symptoms relative insulin deficit, whether due to beta-cell damage, insulin resistance. The study of carnitine and LDH levels in diabetic patients is significant because both play important roles in the metabolism of glucose and fatty acids. Carnitine is a compound that transports fatty acids into the mitochondria for energy production, while LDH (lactate dehydrogenase) is an enzyme involved in the conversion of glucose to lactate. Humans with type 2 diabetes develop lipid accumulation due to carnitine depletion. LDH is an essential physiological molecule in the glycolytic pathway, and its concentration may be indicative of the condition of cellular metabolism.

Aim: For measuring and evaluating the levels of serum carnitine and LDH in all study groups.

Method: A case-control study was done in the Al-Zahraa Teaching Hospital, Kut, Iraq on 150 Iraqi males and females as patients and control between (April 2022 and January 2023). Their ages ranged between 44 and 77 years. Among them were 120 patients divided into 4 groups 30 type 2 diabetes mellitus; 30 diabetic cardiomyopathies; 30 diabetic nephropathies; 30 diabetic retinopathies and 30 control group where control group's age and gender matched those of the patient groups. All patients gave written informed consent to participate in the clinical study. ELISA was used to measure carnitine and LDH.

Result: In present study, it was confirmed that carnitine was significantly lower than the control group and that LDH was significantly higher than the control group. the study demonstrated significant differences in fasting blood sugar and HbA1C levels among the control group, DM2, DCM, DNP, and DRP groups.

Conclusion: This case-control study revealed significant differences in carnitine levels, LDH, FBS, and HbA1C levels among patients with Type 2 diabetes mellitus (T2DM) and their complications compared to the control group. These findings suggest alterations in energy metabolism and cellular damage in patients, indicating poorer glycemic control, and supporting the presence of uncontrolled diabetes.

Key words: diabetes mellitus; nephropathy; retinopathy; cardiomyopathy; carnitine; lactate; dehydrogenase

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Introduction

Diabetes mellitus is simply described as having high blood sugar levels brought on by a whole or relative insulin deficit, whether due to β-cell damage, insulin resistance, or both. Although diabetes is divided mainly into type 1 diabetes (T1DM) and type 2 diabetes mellitus (T2DM), there are other types of diabetes, such as monogenic diabetes, gestational diabetes(GD) (1), and a late-onset autoimmune form latent autoimmune diabetes in adults (LADA) (2). Carnitine and lactate dehydrogenase (LDH) are two important factors involved in the development and management of diabetes. Carnitine is a compound that plays a crucial role in the transportation of fatty acids into the mitochondria, where they are oxidized to produce energy. Carnitine deficiency has been observed in individuals with diabetes, which can impair the utilization of fatty acids as an energy source.

Lactate dehydrogenase (LDH) is an enzyme involved in the conversion of lactate to pyruvate during anaerobic glycolysis. In individuals with diabetes, there can be an increased production of lactate due to impaired glucose utilization and mitochondrial dysfunction. This elevated lactate production can lead to a condition called lactic acidosis, which is characterized by an accumulation of lactate in the blood. T1DM is a severe chronic autoimmune condition that develops when the pancreatic cells fail to secrete endogenous insulin (typically as a result of β-cells loss) (3). T1DM is also known as insulin dependent diabetes mellitus (IDDM) (Amiri, et al., 2021). Adult-onset diabetes, or non-insulin dependent diabetes, also known as T2DM, is a metabolic condition that is characterized by insulin resistance and pancreatic β-cell failure as a result of uncontrolled hyperglycemia. GD is a condition of impaired glucose tolerance that is identified during pregnancy in women who were not previously known to have such a condition (4). A serious hyperglycemia is indicated by polydipsia, polyuria, and loss of weight, occasionally paired with polyphagia and blurred vision (5). The idea that the development of insulin resistance and T2D is correlated with tissue lipid accumulation and dysregulated fatty acid metabolism is supported by scientific findings L-carnitine transfers long-chain fatty acids from the cytosol to mitochondria, which is necessary for beta-oxidation. Fat cannot be used as fuel when carnitine is absent, so carnitine plays a significant role in the metabolism of fatty acids (6). Many theories contend that type 2 diabetic patients are at a higher risk of carnitine deficiency. However, a deficiency of carnitine leads to a deficit in energy metabolism and reduced energy output. Humans with type 2 diabetes develop lipid accumulation due to carnitine depletion (7). In people with diabetes type 2 and heart disease, carnitine has been shown to improve insulin resistance and vascular function (8). Lactate is an essential physiological molecule in the glycolytic pathway, and its concentration may be indicative of the condition of cellular metabolism (9). Its high content may represent an early indicator of insulin resistance (10). Individuals with T2DM have greater fasting plasma lactate concentrations than non-diabetics (11). Fasting plasma lactate concentrations are higher in diabetic patients with obesity than in obese non-diabetics (12).

Materials and methods

A case-control study was done in the Al-Zahraa Teaching Hospital, Kut, Iraq on 150 Iraqi males and females as patients and control between (April 2022 and January 2023). Their ages ranged between 44 and 77 years. Among them were 120 patients divided into 4 groups 30 type 2 diabetes mellitus; 30 diabetic cardiomyopathies; 30 diabetic nephropathies; 30 diabetic retinopathies and 30 control group where control group's age and gender matched those of the patient groups. All patients gave written informed consent to participate in the clinical study. Carnitine and LDH were measured by ELISA technique. ELISA Kit components for carnitine estimation are Standard (Freeze dried), Biotin-antibody (100 x concentrate), HRP-avidin (100 x concentrate), Biotin-antibody Diluent, HRP-avidin Diluent Sample Diluent, Wash Buffer (25 x concentrate), TMB Substrate, and Stop Solution. While ELISA Kit Components for LDH Estimation are Standard, Sample Diluent, HRP-Conjugate Reagent, 20 x Wash solution, Stop Solution, Chromogen Solution A, and Chromogen Solution B. Also fasting blood sugar and HbA1C were measured by automated Cobas E411/Roche. Patients were asked questions via an anonymous questionnaire form that included information such as age, height, weight, and others. Verbal informed consent was obtained from all participants.

Sample Collection

Five milliliters of blood were taken from each participant's venous puncture in sterile gel tubes and left to coagulate for a few minutes at room temperature before separating the serum from the clot by centrifugation

for 10 minutes at 2012 xg. Then they were divided into several Eppendorf tubes and immediately frozen at -80 °C until used in the bioassays.

Exclusion Criteria

Exclusion of Individuals with secondary causes of diabetes, such as pancreatic disease, hormonal disorders, or drug-induced diabetes. Pregnant women or individuals with gestational diabetes. Individuals with impaired glucose tolerance (IGT) or impaired fasting glucose (IFG) without meeting the diagnostic criteria for diabetes. Individuals with incomplete or insufficient medical records or information. Individuals with a history of severe illness or comorbidities that may impact the study outcomes or interpretation. Individuals with age, gender, compensation of DM, current pharmacotherapy of DM, disease duration. Patients were not included if they had type 1 diabetes, diabetic nephropathy, thyroid disorders, liver disorders, and autoimmune disorders.

Characteristics of the control group

It include: similar demographic characteristics, Similar baseline characteristics, no exposure to the intervention or treatment being studied, placebo or standard of care treatment, randomization, and overall.

Tests and markers Kits were used in this study

Human carnitine and Human lactate dehydrogenase (LDH) ELISA Kit (MyBioSource, USA), Glucose kit and HbA1C kit (Roche, Switzerland).

Diagnostic criteria for diabetes: Fasting Plasma Glucose (FPG) Test

Normal: FPG < 100 mg/dL, Prediabetes: FPG between 100-125mg/dL, Diabetes: FPG ≥ 126mg/dL.

Hemoglobin A1C (HbA1C) Test

Normal: HbA1C < 5.7%, Prediabetes: HbA1C between 5.7-6.4%, and Diabetes: HbA1C ≥ 6.5%.

Medication use by diabetic patients

Regarding medication use by diabetic patients, it depends on the individual's specific condition and management plan. Medications are commonly prescribed to help control blood sugar levels and manage the symptoms and complications of diabetes. The type of medication and its usage will vary based on factors such as the type of diabetes (Type 1 or Type 2), the individual's overall health, and their response to lifestyle modifications.

Statistical analysis

The results were conducted according to GraphPad prism 9.2.1 and statistical package for social sciences (SPSS) version 25 were used together, summarize, analyze, and present data. ANOVA was performed to determine the significance of differences between groups. The data were expressed as mean ± standard deviation (SD) and P value<0.05 was considered statistically significant, while P-values equal to or less than 0.01 were considered high significance using Tukey multiple comparisons.

Results

1. Determination of FBS and HbA1C between control and patient groups

A Comparison of fasting blood sugar and HbA1C among the control, DM2, DCM, DNP, and DRP groups was shown in (Table 1). There was a significant difference in mean fasting blood sugar concentration among patients and control groups ($p < 0.001$). The mean fasting blood sugar of the control group was significantly lower than the mean of DM2, DCM, DNP, and DRP groups ($p < 0.001$) (Table 1) (Figure 1), while there was a significant

difference in mean HbA1C value among patients and control groups ($p < 0.001$). The mean HbA1C of the control group was significantly lower than the mean of the DM2, DCM, and DRP groups ($p < 0.001$) (Table 1) (Figure 2).

Table 1. Comparison of fasting blood sugar and HbA1C among control, DM2, cardiomyopathy, nephropathy and retinopathy groups. There are differences between DM2 with DCM, DNP, DRP.

Characteristic	Control *n* = 30	DM2 *n* =30	DCM *n* =30	DNP *n* = 30	DRP *n* = 30	*P*
FBS (mg/dL)						
Mean ±SD	88.8 ±7.0	241.4±48.2	275.1±60.5	283±62.8	271.1±63.6	<0.001
Range	80 - 100	150 - 350	180 - 420	190 - 400	182 - 418	
HbA1C (%)						
Mean ±SD	5.26±0.25	8.46±1.28	9.64±1.67	9.4 ±1.35	9.20±1.516	<0.001
Range	4.9 - 5.4	6.7 - 12	7.3 -14	7.7 -14.3	7 - 14.4	

n**:* number of cases; **SD**: standard deviation; **: significant at $p \leq 0.05$; ******: significant at $p \leq 0.01$; *******: significant at $p \leq 0.001$; **NS**: not significant; **DM2**: diabetic mullites type 2; **DCM**: diabetic cardiomyopathy; **DNP**: diabetic nephropathy; **DRP**: diabetic retinopathy; **FBS**: fasting blood sugar.

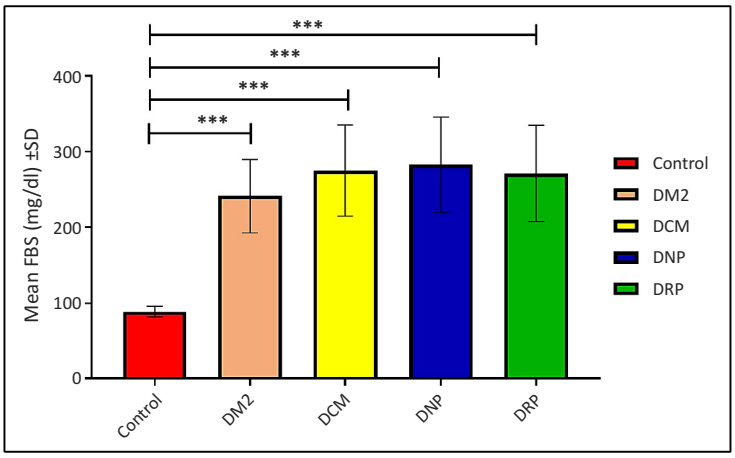

Figure 1. Comparison of mean FBS among control group, DM2 group, DCM group, DNP group and DRP group. There are differences between DM with DCM, DNP, DRP.

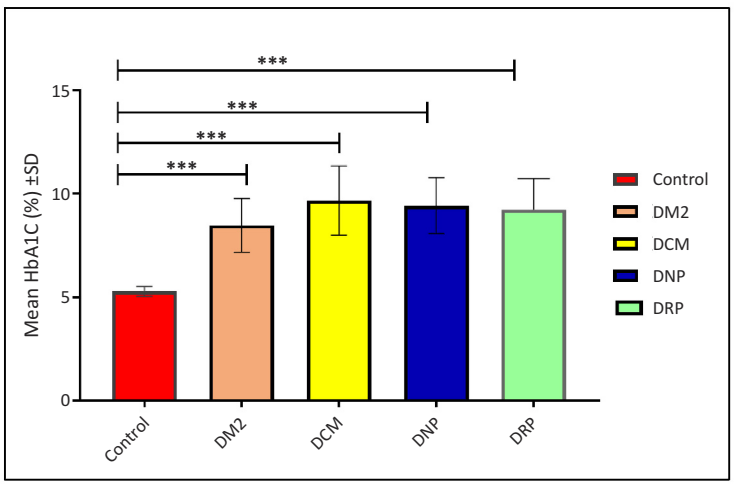

Figure 2. Comparison of mean HbA1C among control group, DM2, DCM, DRP and DNP groups. There are differences between DM with DCM, DNP, DRP and DNP.

2. Determination of Carnitine and LDH between control and patient groups

A Comparison of carnitine and LDH among the control, DM2, DCM, DNP, and DRP groups are shown in (Table 2). There was a significant difference in mean serum carnitine concentration among patients and control groups ($p < 0.001$). The mean serum carnitine of the control group was significantly higher than the mean of DM2, DCM, DNP, and DRP groups ($p < 0.001$) (Table 2) (Figure 3), while there was a significant difference in mean serum LDH activity among patients and control groups ($p < 0.001$). The mean serum LDH of the control group was significantly lower than the mean of DM2, DCM, DNP, and DRP groups ($p < 0.001$) (Table 2) (Figure 4).

Table 2. Comparison of results of Carnitine and LDH biochemical markers among control group, DM2 group, DCM group, DNP group, DRP group, and DRP group, There are differences between DM with DCM, DNP, DRP.

Characteristic	Control *n* = 30	DM2 *n* =30	DCM *n* =30	DNP *n* = 30	DRP *n* = 30	*P*
Carnitine (µmol/L)						
Mean ±SD	49.30 ±6.47	32.70 ±6.8	38.13±8.3	34.50±10.1	33.40±9.2	<0.001
Range	40-74	20-51	26-62	22-60	21-61	
LDH (U/L)						
Mean ±SD	161.5 ±31.7	230.8 ±43.6	245.6 ±47.6	251.4 ± 41.5	258.6 ±45.3	<0.001
Range	115 - 225	134 - 297	132 - 308	197 - 367	185 - 377	

n: number of cases; **SD**: standard deviation; **O**: one way **ANOVA**; *: significant at $p \leq 0.05$; **: significant at $p \leq 0.01$; ***: significant at $p \leq 0.001$; **NS**: not significant; **DM2**: diabetic mullites type 2; **DCM**: diabetic cardiomyopathy; **DNP**: diabetic nephropathy; **DRP**: diabetic retinopathy; **LDH**: lactate dehydrogenase.

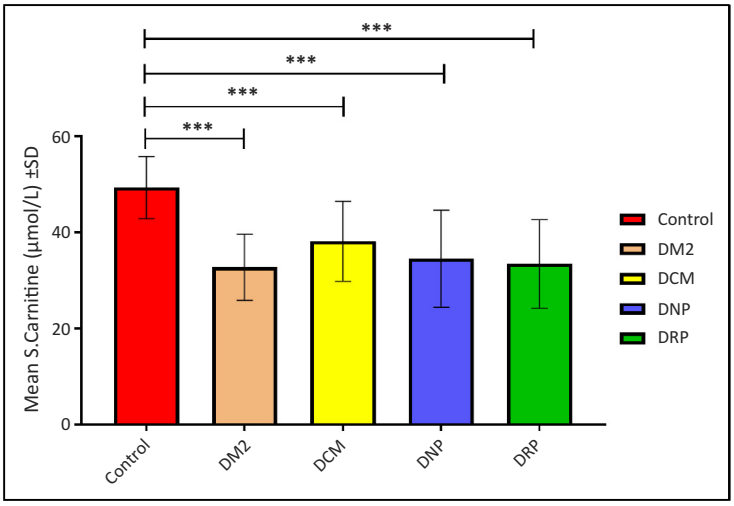

Figure 3. Comparison of mean Serum (S) Carnitine among control group, DM2 group, DCM group, DNP group and DRP group. There are differences between DM with DCM, DNP, DRP. There are differences between DM2 with DCM, DNP, DRP.

Discussion

Since these patients had diabetes mellitus type 2 and insulin resistance, fasting blood sugar would be higher at morning (13) (14). A study in the north of Iran by (15) showed a high significant FBS concentration in diabetic retinopathy patients as compared with healthy people. Also, a case-control observational study done by (16) on diabetic nephropathy patients showed high levels of FBS in these patients. The high HbA1C in the DM2 group (8.46%) indicates these patients have impaired glycemic control, and this value increases with the DCM and DRP

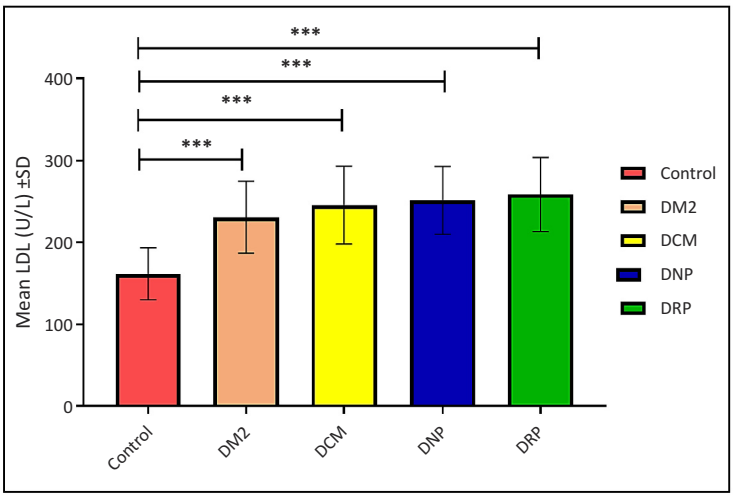

Figure 4. Comparison of mean LDH among control group, DM2 group, DCM group, DNP group and DRP group. There are differences between DM2 with DCM, DNP, DRP.

groups, which means these patients have poor glycemic control due to increased blood sugar concentration, which increases glycated protein, thus increasing HbA1C (17). A study by showed that HbA1C values were highly significant in the diabetic type 2 group as compared with the non-diabetic group. Another cross-sectional study done on 288 Saudi patients with DM2 by (18) showed these patients had poor glycemic control.

Diabetes is a condition linked with altered lipid and carbohydrate metabolism, resulting in oxidative stress. Carnitine enhances the oxidation of long-chain acyl CoA in the mitochondria, which is considered to be the source of insulin resistance in the heart and muscles; hence, it has substantial impacts on glucose metabolism. Additionally, carnitine influences the activity of glycolytic and gluconeogenic enzymes. Finally, carnitine improves glucose consumption in cells through changing the expression of genes involved in the insulin signaling cascade (19). In type II diabetes, more carnitine is used to remove the intermediates of fatty acid oxidation, which contribute to insulin resistance. Consequently, it is hypothesized that carnitine levels may be lower in people with type II diabetes compared to healthy (20). Another study showed that the serum carnitine concentrations in diabetic individuals with complications were lower than those of diabetic individuals without complications. Also, a study by (21) was found that there was a low level of carnitine in diabetic patients as compared with healthy people.

Since T2D is characterized by insulin resistance and obesity, obesity results in the production of a significant amount of lactate by adipose tissue. Reduced blood supply to adipose tissue causes hypoxia and increased lactate generation in obese individuals (22). Moreover, when adipocyte size rises and approaches the diffusion limit of oxygen, lactate generation increases. Therefore, reduced oxygen availability in adipocytes may be largely responsible for the excessive lactate generation associated with obesity. A case-control study done on 571 Singapore Chinese patients with DM2 by (23) found that LDH had no significant association between LDH and DM2.

Conclusion

This case-control study revealed significant differences in carnitine levels and LDH levels among patients with Type 2 diabetes mellitus (DM2) and their complications compared to the control group. The findings showed that patients with T2DM and their complications had decreased carnitine levels, while LDH levels increased significantly. These findings suggest alterations in energy metabolism and cellular damage in patients with T2DM and their complications. Additionally, the study demonstrated significant differences in fasting blood sugar and HbA1C levels among the control group, DM2, DCM, DNP, and DRP groups. FBS and HbA1C levels were higher in the patient groups compared to the control group, indicating poorer glycemic control, and supporting the presence of uncontrolled diabetes. Overall, this study provides valuable insights into the biochemical changes associated with DM2 and its complications. The observed differences in these tests highlight the importance of monitoring

these parameters in the management and assessment of DM2 patients. It is recommended to monitor carnitine, LDH, FBS and HbA1C levels in DM2 patients. Also, Glycemic control and the importance of a comprehensive assessment should be made.

Conflict of Interest Statemen

Regarding the publishing of this work, the authors state that they have no conflicts of interest. The study design, data analysis, and result interpretation were all done independently, without the influence of outside financing or conflicting interests. The writers don't have any personal or financial ties to people or organizations that would slant their writing. This statement guarantees the integrity and objectivity of the research described in this article.

Adherence to Ethical Standards

The study included blood sample collection and experimental procedures authorized by Al-Zahra Teaching Hospital in Kut, Iraq. Before collecting samples, all methods and procedures were followed, and rules and standards were set by the Ethical Committee at the College of Medicine at Al-Qadisiyah University. Patients have the right to know what type of tests are being performed on their samples, as well as any potential risks and benefits. Before performing any test, informed patient consent is taken and information about patients and test results is kept private and shared with authorized persons only.

References

1. Poorabbas A, Fallah F, Bagdadchi J, et al. Determination of free L-carnitine levels in type II diabetic women with and without complications. European journal of clinical nutrition. 2007;61(7):892-895.
2. Chen S, Du K, Zou C. Current progress in stem cell therapy for type 1 diabetes mellitus. Stem Cell Research & Therapy. 2020;11(1):1-13.
3. Ahlqvist E, Storm P, Käräjämäki A, et al. Novel subgroups of adult-onset diabetes and their association with outcomes: a data-driven cluster analysis of six variables. The lancet Diabetes & endocrinology. 2018;6(5):361-369.
4. McIntyre HD, Catalano P, Zhang C, et al. Gestational diabetes mellitus. Nature reviews Disease primers. 2019;5(1):47.
5. Mukhtar Y, Galalain A, Yunusa U. A modern overview on diabetes mellitus: a chronic endocrine disorder. European Journal of Biology. 2020;5(2):1-14.
6. Makrecka-Kuka M, Liepinsh E, Murray AJ, et al. Altered mitochondrial metabolism in the insulin-resistant heart. Acta Physiologica. 2020;228(3):e13430.
7. Cuomo P, Capparelli R, Iannelli A, et al. Role of branched-chain amino acid metabolism in type 2 diabetes, obesity, cardiovascular disease and non-alcoholic fatty liver disease. International Journal of Molecular Sciences. 2022;23(8):4325.
8. El-Sheikh HM, El-Haggar SM, Elbedewy TA. Comparative study to evaluate the effect of l-carnitine plus glimepiride versus glimepiride alone on insulin resistance in type 2 diabetic patients. Diabetes & Metabolic Syndrome: Clinical Research & Reviews. 2019;13(1):167-173.
9. Li X, Yang Y, Zhang B, et al. Lactate metabolism in human health and disease. Signal Transduction and Targeted Therapy. 2022;7(1):305.
10. Jones TE, Pories WJ, Houmard JA, et al. Plasma lactate as a marker of metabolic health: Implications of elevated lactate for impairment of aerobic metabolism in the metabolic syndrome. Surgery. 2019;166(5):861-866.
11. Xin N, Jianfei E, He H, et al. (2022). Fasting plasma lactate: A diagnosis and staging biomarker with superior performance in adult nonalcoholic fatty liver disease.
12. Rehman SSU, Karimi H, Gillani SA, et al. Effects of supervised structured aerobic exercise training programme on level of Exertion, dyspnoea, VO2 max and Body Mass Index in patients with type 2 diabetes mellitus. J. Pak. Med. Assoc. 2017;67:1670-1673.
13. Zheng F, Liu S, Liu Y, et al. (2019). Effects of an outpatient diabetes self-management education on patients with type 2 diabetes in China: a randomized controlled trial. Journal of diabetes research, 2019.
14. Huang F-Y, Deng T, Meng L-X, et al. Dietary ginger as a traditional therapy for blood sugar control in patients with type 2 diabetes mellitus: A systematic review and meta-analysis. 2019;98(13):e15054.

15. Rasoulinejad SA, Meftah N, Maniati MS, et al. (2022). High levels of FBS and HbA1c and their association with diabetic retinopathy: a study in the north of Iran. Journal of Diabetes & Metabolic Disorders. 2022;21(1):399-406.
16. Lee S, Liu T, Zhou J, et al. (2021). Predictions of diabetes complications and mortality using hba1c variability: a 10-year observational cohort study. Acta Diabetologica. 2021;58:71-180.
17. Abera RG, Demesse ES, Boko WD. Evaluation of glycemic control and related factors among outpatients with type 2 diabetes at Tikur Anbessa Specialized Hospital, Addis Ababa, Ethiopia: a cross-sectional study. BMC endocrine disorders. 2022;22(1):54.
18. Badedi M, Solan Y, Darraj H, et al. Factors associated with long-term control of type 2 diabetes mellitus. Journal of diabetes research. doi: 10.1155/2016/2109542.
19. Shaukat D, Haroon ZH, Khalid UB, et al. Establishing Carnitine Deficiency In Patients Of Type-Ii Diabetes Mellitus With Early Diabetic Retinopathy. Pakistan Journal of Pathology. 2021;32(4):152-156.
20. Ramazani M, Qujeq D, Moazezi Z. (2019). Assessing the levels of L-carnitine and total antioxidant capacity in adults with newly diagnosed and long-standing type 2 diabetes. Canadian journal of diabetes. 2019;43(1):46-50. e41.
21. Kılıçlı F, Dökmetaş S, Candan F, et al. Inspiratory muscle strength is correlated with carnitine levels in type 2 diabetes. Endocrine research. 2010;35(2):51-58.
22. Kiran SA, Baghel M, Kandwal M, et al. Association of serum lactate levels among patients with Type 2 diabetes mellitus of a tertiary care hospital, Uttar Pradesh. National Journal of Physiology, Pharmacy and Pharmacology. 2022;12(5):620-623.
23. Wang Y-L, Koh W-P, Yuan J-M, et al. Association between liver enzymes and incident type 2 diabetes in Singapore Chinese men and women. BMJ Open Diabetes Research and Care. 2016;4(1):e000296.