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EVALUATION OF SERUM ZINC LEVELS AND ITS RELATIONSHIP TO GLYCEMIC CONTROL AND LIPID PROFILE IN TYPE (2) DIABETES MELLITUS

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Due to the importance of Zinc in glucose and lipid metabolism, it has been aimed in this study to investigate the association of serum zinc levels with glycemic control and lipid profile in type 2 diabetes mellitus patients (T2DM). This study has been conducted on 129 adult patients with diabetes at Tishreen University Hospital (TUH) and The Diabetes Center of Latakia City in Syria, and 51 matched healthy subjects. The results have shown a statistically significant decrease in serum zinc levels in T2DM patients compared with controls. A statistically significant negative correlation has been identified between zinc and all of HbA1C, BMI, duration of disease, and triglycerides, while a positive correlation has been observed with HDL levels in the diabetic group. In conclusion, this study has demonstrated that low zinc level is associated with poor glycemic control and it may give a cause to explain a possible association with dyslipidemia observed in T2DM patients.

Keywords: *Type 2 Diabetes Mellitus, Serum Zinc, Glycemic Control, Dyslipidemia*

INTRODUCTION

Type 2 Diabetes Mellitus (T2DM) is a chronic metabolic disorder characterized by persistent hyperglycemia, which is caused by a combination of peripheral insulin resistance (IR) and pancreatic beta-cell dysfunction. It is the most common form of diabetes, approximately 90% of all patients with diabetes mellitus have T2DM[1]. IR can also alter systemic lipid metabolism, which leads to the development of dyslipidemia[2]. Several theories have been proposed to understand the mechanisms associated with IR including genetic defects, physical inactivity, obesity, family history, oxidative stress, and inflammation[3]. Recently, researchers have become interested in studying the association between trace elements and diabetes mellitus[4]. Zinc is a micronutrient that plays a fundamental role in metabolic and immunological processes, wound healing, and recently as an important mediator in hemostasis and thrombosis[5&6]. Zinc also plays an important role in lipid and glucose metabolism. Insulin is stored and crystallized in pancreatic beta cells as a hexamer, consisting of six insulin monomers with two zinc ions in the center[7]. In addition, it is required for insulin secretion and activation of the insulin signaling pathway by inhibiting the activity of protein tyrosine phosphatase 1B (PTP1B); a negative regulator of the insulin signaling pathway[8]. Zinc also influences the expression of peroxisome proliferator-activated receptors α (PPAR-α), which regulates the expression of several genes critical for lipoprotein metabolism such as lipoprotein lipase (LPL); the enzyme that breaks down triglycerides in very-low-density lipoprotein (VLDL) into fatty acids and glycerol for the cell to use[9]. Furthermore, zinc is important for the biosynthesis of zinc alpha-2 glycoprotein (ZAG); an adipokine secreted by various organs such as the liver, breast, and adipose tissue[10]. The most known biological role of ZAG is inducing lipolysis by stimulating the expression of lipolytic enzymes such as hormone-sensitive lipase (HSL) and reducing

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the expression of lipogenic enzymes such as Fatty acid synthase (FAS) [11&12]. ZAG also appears to have a protective role against IR by stimulating adiponectin secretion in adipocytes; an adipokine that increases the sensitivity of insulin in peripheral organs[13]. Primary deficiency of zinc (inadequate dietary intake and/or poor availability) and secondary zinc deficiency (states of decreased absorption and/or excessive excretion) are characterized by impaired immune function and altering lipid and glucose metabolism[14&15]. Due to the prevalence of zinc deficiency in developing countries, discrepancies in results regarding the effect of zinc on lipid profile, lack of previous evaluation of serum zinc levels in patients with diabetes in Syria, it has been aimed in this study to evaluate serum zinc levels among T2DM patients group compared with healthy subjects and investigate the association of serum zinc levels with glycemic control and lipid profile in T2DM patients.

Material and Methods

The present study was conducted at Tishreen University Hospital (TUH) and The Diabetes Center of Lattakia City in Syria from November 2020 to December 2021. An informed written consent was taken from each participant in the study.

Ethical approval

All procedures were approved by the Institutional Review Board of Tishreen University. The decision involved Ethical Approval (Decision Number: **2774** in September 2020). An informed written consent was taken from each participant in the study.

Patients

One hundred twenty-nine adult patients (62 Males and 67 females), diagnosed with T2DM, were enrolled in this study, and 51 age–matched healthy subjects as a control group. A questionnaire with questions about age and medical history was completed by all participants. Weight, height, Systolic Blood Pressure (SBP), and Diastolic Blood Pressure (DBP) were measured. Body Mass Index (BMI) was calculated as the ratio of an individual's weight in kilograms (kg) divided by the height in meters squared (m^2)[16].

Participants were divided according to the World Health Organization (WHO) classification of BMI for adults as normal weight (BMI < 25 kg/m^2), overweight (BMI from 25-29.9 kg/m^2), obesity: BMI ≥ 30 kg/m^2.

Exclusion criteria

Patients with chronic diseases that interfere with zinc absorption such as Crohn's disease, who reported receiving zinc supplements, were excluded.

Analytical methods and instrumentation

HbA1C has been determined using the technique of High-Performance Liquid Chromatography (HPLC) by Tosoh Automated Glycohemoglobin Analyzer (HLC®-723GX)/India. Biochemical assays for lipid profiles have been performed with commercially available kits from Biosystem® (Spain) by a Mindary BS-380 clinical chemistry analyzer from China (Total cholesterol; Cat. No. 11505, Triglycerides; Cat. No. 11528, LDL-C; Cat. No. 23585 and HDL-C; Cat. No. 23557). Serum zinc concentrations and fasting blood glucose have been analyzed using a kit from Medichem Middle East® (Syria) and Biosystem®, respectively with a Semi-Automatic Microprocessor Controlled Photometer (HumaLyzer Primus) from Germany: (Glucose; Cat. No. 11503 and Zinc; Cat. No. 12650).

Statistical analysis

Data have been analyzed using the Statistical Package for Social Sciences (SPSS) version 20 for Windows. Data are expressed as mean ± standard deviation (SD). The student's t-test has been used to compare the means of different variables between two independent samples (the diabetes group and the healthy group). Analysis of variance (ANOVA) of one factor has been used to identify differences in means in serum zinc levels between categories of FBG, HbA1C, TG, total cholesterol, BMI, and duration of disease. Pearson's coefficient has been performed to show the linear correlation between different variables. Results have been expressed as a correlation coefficient (r) and a P-value Less than or equal to 0.05, which is considered statistically significant.

RESULTS AND DISCUSSION

Results

Table1: Demographic Data for Subjects Included in This Study.

Demographic characteristics	Diabetes	Controls
Number of subjects	n= 129	n= 51
Male	n= 62	n= 23
Female	n= 67	n= 28
Age (years; mean ± SD)	46.63 ± 11.35	45.19 ± 10.60
Duration of disease (years; mean ± SD)	5.52 ± 3.89	-

(Table 1) shows the demographic characteristics of the study subjects. The distribution of participants by sex was homogeneous in both groups. In the diabetic group 48% (n= 62) of subjects were men and 52% (n= 67) were women, while in the control group 45% (n= 23) of subjects were men and 55% (n= 28) were women. According to this data, the mean age was similar in the two groups (46.63 ± 11.35 years for the diabetes group and 45.19 ± 10.60 years for the controls).

(Table 2) shows a comparison between the diabetes group and the control group according to the parameters examined. It could be noticed that FBG, HbA1c, LDL cholesterol, and BMI were higher in the diabetic group. There was a significant difference between the means of the two groups ($p< 0.001$). Triglycerides and total cholesterol were also higher in patients with diabetes and significant differences between the mean of the two groups were identified ($p< 0.05$). On the other hand, it was noticed that the serum zinc levels and HDL cholesterol concentrations were lower in the diabetic group compared with the controls and the differences between the means of the two groups were significant ($p< 0.001$). SBP and DBP values were lower in the control group. However, there were no significant differences between the means of the two groups in terms of blood pressure values ($p> 0.05$). Depending on the reference range of the kit used (Medichem Middle East®), the normal serological values of zinc range from 46 µg/dL to 150 µg/dL for adults[17]. According to the zinc values, Diabetes were divided into two groups; group1: diabetics with zinc < 46 µg/dL (n=41) and group2: diabetics with zinc ≥ 46 µg/dL (n=88). It was found that HbA1C, TG, and BMI levels were significantly higher in group 1 ($p< 0.001$). Group 1 also has higher FBG concentrations compared with group 2 ($p< 0.05$) while HDL levels were significantly higher in group 2 ($p< 0.001$). On the other hand, there were no significant differences between the mean of the two groups in terms of total cholesterol, LDL cholesterol, SBP, and DBP ($p> 0.05$).

Table 2: Comparison of the Studied Parameters between Diabetics and Controls.

parameter	**Diabetes**		**Control**		**p-value***
	Mean	**SD**	**Mean**	**SD**	
FBG (mg/dL)	146.37	56.34	79.47	11.08	**0.000**
HbA1C %	8.25	1.68	5.22	0.71	**0.000**
Zinc (µg/dL)	86.20	34.99	132.86	24.89	**0.000**
TG(mg/dL)	171.13	58.60	143.86	40.12	**0.002**
Total cholesterol (mg/dL)	196.15	28.50	177.25	28.54	**0.031**
LDL-C(mg/dL)	98.61	22.78	77.94	14.30	**0.000**
HDL-C(mg/dL)	57.62	15.05	68.09	20.02	**0.000**
BMI (Kg/m^2)	29.05	4.42	24.73	1.07	**0.000**
SBP(mmHg)	121.02	17.53	119.03	9.35	0.345
DBP(mmHg)	84.06	7.3	83.62	7.49	0.323

*T-test to independent samples, **FBG**; Fasting Blood Glucose, **TG**; Tri-glycerides**, LDL-C**; Low Density Lipoprotein Cholesterol, **HDL-C**; High Density Lipoprotein Cholesterol; **BMI;** body mass index, **SBP**; Sytosolic Blood Pressure, **DBP**; Diastolic Blood Pressure .

Table 3: Comparison of Two Patients Groups based on Serum Zinc Levels.

Parameter	Group1 (Zn < 46μg/dL) *n*= 41	Group2 (Zn ≥ 46μg/dL) *n*= 88	p-value*
FBG (mg/dL)	187 ± 70.79	149.06 ± 36.29	**0.001**
HbA1C %	9.31 ± 1.57	7.82 ± 1.52	**0.000**
Zinc (μg/dL)	37.27 ± 8.77	105.89 ± 17.95	**0.000**
TG (mg/dL)	213.00 ± 66.26	154.30 ± 45.76	**0.000**
Total cholesterol(mg/dL)	195.56 ± 27.02	196.39 ± 29.22	0.883
LDL-C (mg/dL)	91.97 ± 14.93	89.28 ± 24.82	0.352
HDL-C(mg/dL)	49.62 ± 10.88	61.20 ± 14.32	**0.000**
BMI (Kg/m^2)	32.97 ± 4.59	27.47 ± 3.17	**0.000**
SBP(mmHg)	119.72 ± 9.52	121.54 ± 19.75	0.597
DBP(mmHg)	83.81 ± 7.79	84.17 ± 7.13	0.801

*T-test to independent samples

(Table 4) shows the association between serum zinc levels and studied parameters in patients with diabetes. Diabetes mellitus patients were divided into three categories based on FBG and HbA1C levels. It was observed that mean zinc levels were lower in patients with high levels of FBG and HbA1C. Moreover, zinc levels were compared by FBG and HbA1C and showed significant differences ($p< 0.05$ and $p< 0.001$, respectively). The relation between serum zinc levels and lipid profile was also analyzed. It was found that a high level of triglyceride was associated with the lowest level of zinc and the difference was significant ($P< 0.05$), while there was no association between zinc concentration and both total cholesterol and LDL levels ($P> 0.05$). On the other hand, it was found that diabetes with low zinc levels had low HDL cholesterol levels compared with those with elevated zinc concentration. The difference in serum zinc levels by HDL cholesterol was significant ($p< 0.05$). According to the BMI score, it was noticed that serum zinc levels were lower in obese diabetics compared with overweight and normal-weight diabetics. The differences in zinc levels by BMI were significant ($p< 0.001$). On the other hand, there were no statistically significant differences in zinc concentration by SBP, DBP, and sex ($p>0.05$). The relationship between serum zinc concentrations and the duration of diabetes was also analyzed. Results showed that patients with at least ten years of diabetes mellitus had significantly lower zinc concentrations compared with those with fewer years of disease ($p< 0.001$). As a result of Pearson's test, a significant inverse correlation was found between zinc and FBG (Fig.1), HbA1C% (Fig.2), TG (Fig.3), BMI (Fig.4) and duration of disease (Fig.5), while on the other hand a positive correlation was found with HDL levels (Fig.6) in patients with diabetes. The rest of the correlations were not significant (Table 4).

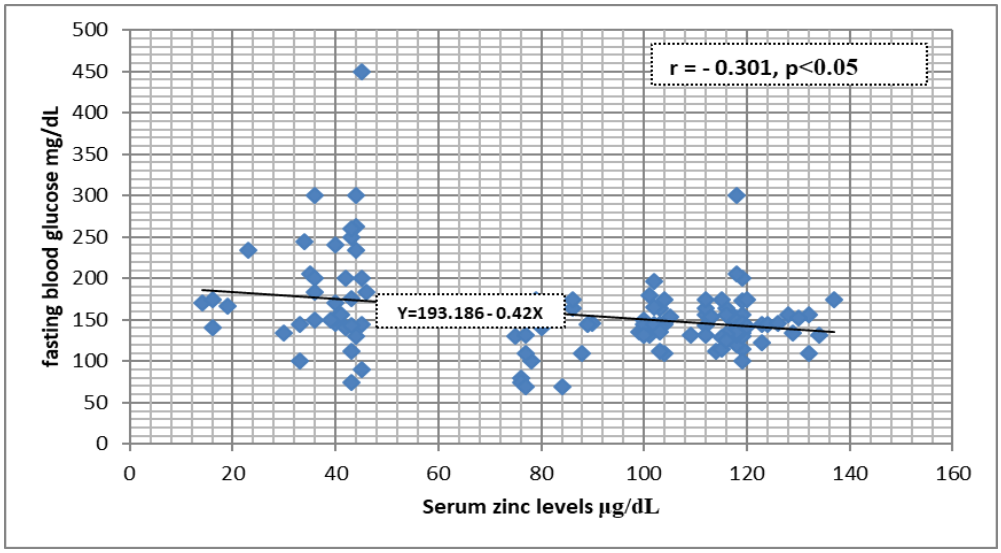

Fig.1: Correlation between serum zinc levels and fasting blood glucose (r = - 0.301, p<0.05).

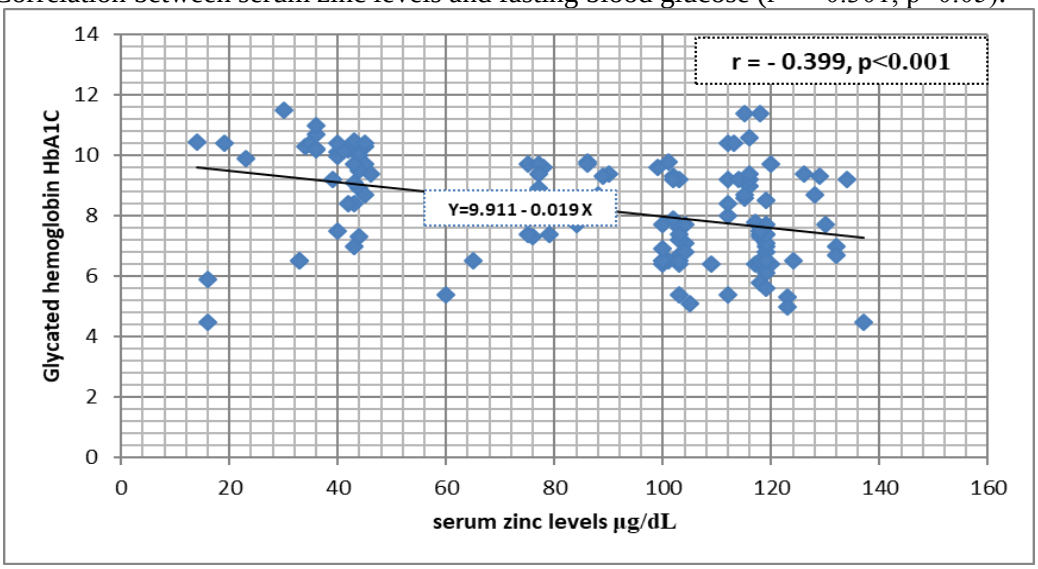

Fig. 2 : Correlation of serum zinc levels and Glycated hemoglobin HbA1C (r = - 0.399, p<0.001).

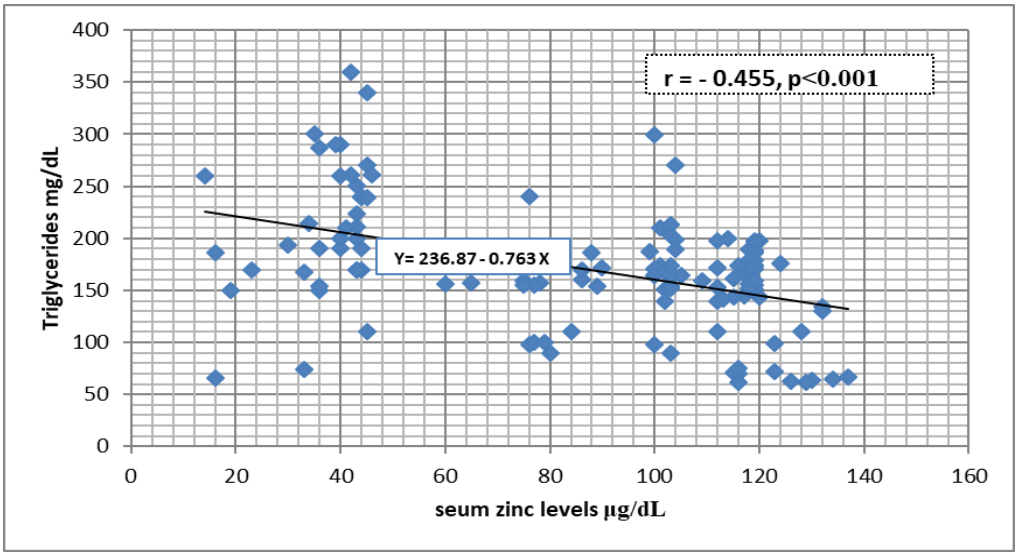

Fig. 3 : Correlation of serum zinc levels and triglycerides (r = - 0.455, p<0.001).

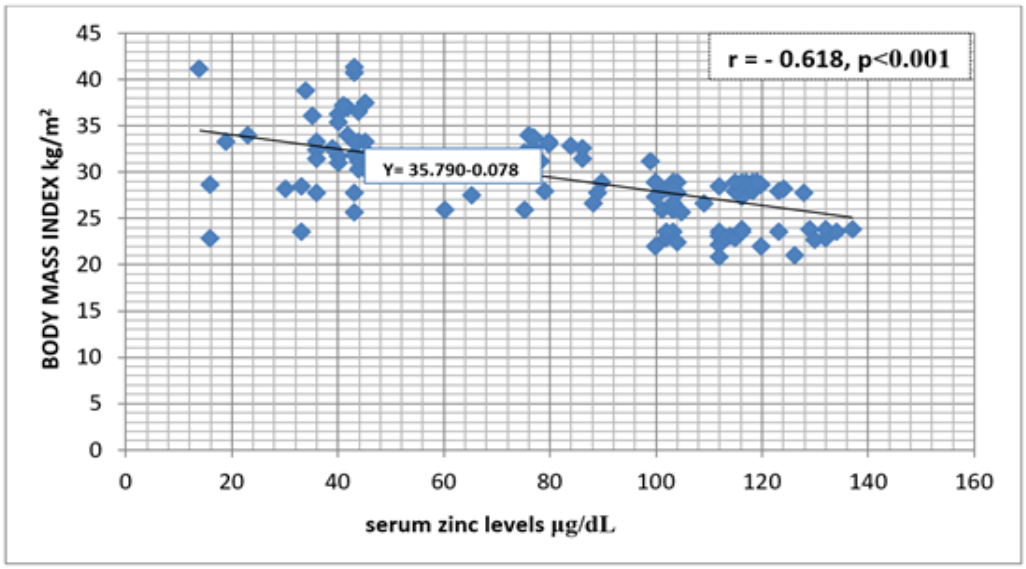

Fig. 4: Correlation between serum zinc levels and BMI (r = - 0.618, p<0.001).

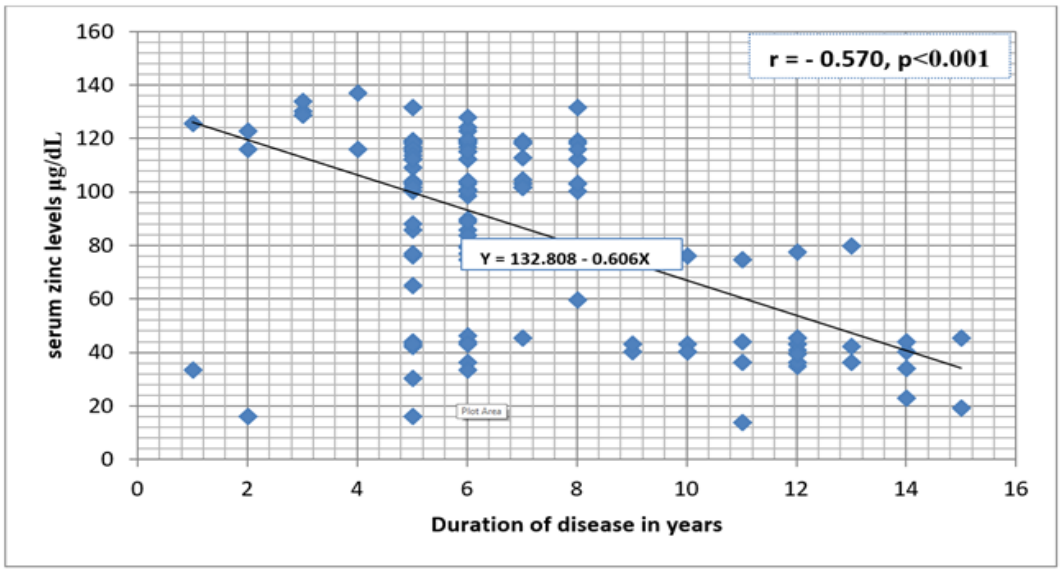

Fig. 5: correlation of serum zinc levels and duration of disease (r = - 0.570, p<0.001).

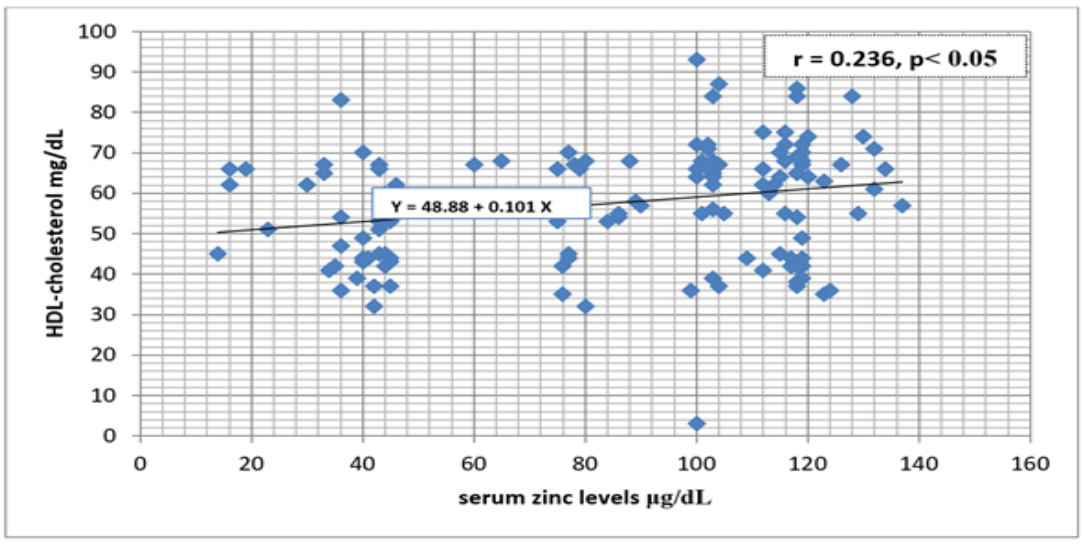

Fig. 6: Correlation of serum zinc levels with HDL cholesterol (r = 0.236, p< 0.05).

Table 4: Correlation of Serum Zinc Levels with Various Parameters in the Diabetic Group.

parameter	values		n	serum zinc levels (µg/dl)			
				Mean ± SD	P-value	Correlation Coefficient	P-value***
FBG (mg/dL)	< 150		70	93.89±29.96	**0.005****	- 0.301	**0.001**
	150-250		53	79.13±38.54			
	> 250		6	55.00±31.03			
HbA1C %	< 8		60	99.95±3.79	**0.000****	- 0.399	**0.000**
	8-10		46	31.52±4.64			
	> 10		23	33.82±7.05			
TG (mg/dL)	< 150		33	104.60±30.7	**0.000****	- 0.455	**0.000**
	150-200		68	92.10±28.78			
	> 200		28	50.21±27.99			
Total cholesterol (mg/dL)	< 200		72	96.72±29.73	0.217**	- 0.274	0.253
	200-250		46	87.78±33.16			
	> 250		11	84.62±36.65			
LDL-C (mg/dL)	< 100		61	103.52±33.5	0.321*	- 0.211	0.257
	≥ 100		68	99.38±30.94			
HDL-C (mg/dL)	male	≤ 45	25	64.72±31.58	**0.000***	0.236	**0.007**
		> 45	37	101.48±26.2			
	female	≤ 55	30	65.35±33.78	**0.000***		
		>55	37	102.64±29.4			
BMI (kg/m^2)	< 25		25	109.80±27.9	**0.000****	- 0.618	**0.000**
	25 - 29.9		61	100.81±26.4			
	≥ 30		43	51.76±20.72			
SBP (mmHg)	< 120		60	99.03±34.71	0.208 **	- 0.164	0.415
	120-140		51	99.98±34.45			
	>140		18	101.89±29.6			
DBP (mmHg)	< 80		58	97.48±35.16	0.153**	- 0.471	0.216
	80-90		61	96.05±24.05			
	> 90		10	98.67±31.99			
Gender	Male		62	85.66±33.60	0.806*	-	-
	Female		67	86.66±36.48			
Duration of disease (years)	< 5 years		25	123.62±25.8	**0.000****	- 0.570	**0.000**
	5-9 years		71	97.32±51.61			
	≥ 10 years		33	36.33±24.16			

*T-test to independent samples, ** ANOVA of a factor, *** Pearson's Correlation.

Discussion

Zinc has been studied as an essential trace element in the diet for its biological importance in different processes associated with the body's homeostasis[18]. Serum zinc levels were estimated in all subjects; patients with diabetes appeared to have lower serum zinc levels compared with controls, and that was consistent with the results of previous studies[19&20] The results of Pearson's test have shown that a decrease in serum zinc level was associated with poor glycemic control in diabetes. A similar result was reached in a previous study[21]. This study also showed that a decrease in serum zinc level occurs with the increase in the duration of the disease. Previous studies have shown that hypozincemia observed in diabetics was associated with hyperzincuria, which could be explained by renal function disorder as the years of illness increase, especially in the case of uncontrolled disease[22&23]. The association between serum zinc levels and lipid profile has been controversial. This study demonstrated a negative correlation between serum zinc levels and triglycerides and a positive correlation between serum zinc levels and HDL cholesterol, while no association of zinc concentration with total cholesterol and LDL cholesterol was demonstrated. In the case of lipid profile, Ma et al. reported a positive correlation of serum zinc levels with HDL cholesterol[24]. Yang et al. reported a negative association of zinc levels with HDL cholesterol in men and women[25]. Dasarathan et al. reported

that zinc had a negative association with triglyceride and total cholesterol in diabetes[19]. The discrepancies in results could be explained by the difference in study design and participants' characteristics (the mean of serum zinc levels, diet, smoking, alcohol intake, and other diseases that could affect lipid profile). Overweight and obesity are considered to be the most important modifiable risk factors for IR[26]. This study showed that 47.28% (n= 61) of diabetes were overweight and 33.33% (n= 43) were obese. A Negative correlation was observed between serum zinc concentrations and BMI; these data are consistent with the result of a similar study[19]. Zinc participates in ZAG biosynthesis. Therefore, a decrease in zinc levels could affect the biological function of ZAG as a lipid mobilizing factor[27]. The effect of zinc supplementation on blood sugar levels and lipid profile has been tested, a previous study has been reported that diabetics, who received zinc supplementation (30 mg/day), have a significant reduction in HbA1C concentrations and decreased levels of total cholesterol, LDL cholesterol, and triglycerides compared with placebo group[28]. Zinc supplementation improves insulin sensitivity in adipose tissue by activating hormone signaling pathways. Therefore, this mineral stimulates the translocation of glucose transporter-4 (GLUT-4) to the plasma membrane[29]. Zinc also influences the expression of PPARs-α, which plays a major regulator in lipoprotein metabolism[30]. The serum zinc concentrations were slightly higher in females than in males. However, no significant difference was found in serum zinc concentrations by gender, which is consistent with a report of a previous study[31].

Conclusion

This study suggests that T2DM patients with decreased serum zinc levels have higher levels of HbA1C, fasting blood glucose, triglycerides, and lower levels of HDL cholesterol. In addition, obese patients have lower zinc values compared with normal-weight patients. No other association was found between serum zinc concentrations with total cholesterol, LDL cholesterol, SBP, DBP, and sex. However, this does not rule out the possibility that low serum zinc concentrations may affect blood pressure, total cholesterol, and LDL cholesterol concentrations. Today, more large-scale studies are needed to explore the role of zinc in chronic metabolic diseases.

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نشرة العلوم الصيدلية
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تقييم المستويات المصلية للزنك وعلاقته مع ضبط سكر الدم والصيغة اللبيدية لدى مرضى الداء السكري من النمط الثاني

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نظراً لأهميّةِ الزنكِ في استقلابِ الجلوكوز والدسم، كانَ الهدفُ من هذهِ الدراسة هو التحقق من علاقةِ المستوياتِ المصليّةِ للزنكِ مع ضبطِ سكرِ الدم والصيغةِ اللبيديّة لدى مرضى الداء السكريِّ من النمطِ الثاني. تمَّ اجراءُ هذه الدراسة على ١٢٩ بالغاً من مرضى الداء السكري في مستشفى تشرين الجامعي ومركز السكري في مدينة اللاذقيّة/ سوريا مع ٥١ شخصاً من الشواهدِ الأصحاء. أظهرتِ النتائجُ انخفاضاً ذو دلالةٍ احصائيّةٍ هامّة في مستويات الزنكِ لدى مجموعةِ السكريِّ بالمقارنةِ مع مجموعةِ الشواهدِ الأصحاء. تمَ تحديدُ وجودِ علاقة ارتباطٍ سلبيّة هامَة احصائيّاً بين الزنك وكل من الخضاب الجلوكوزي، مؤشر كتلة الجسم، مدة الإصابة بالمرض، والشحوم الثلاثية بينما تم ملاحظة وجودِ علاقة ارتباطٍ ايجابيّةٍ مع مستويات الكوليسترول عالي الكثافة لدى مجموعة مرضى السكري. ومنهُ، أشارتِ هذهِ الدراسة إلى أنَّ انخفاضَ مستوياتِ الزنكِ قد ترافقَ مع ضبطٍ سيءٍ لسُكرِ الدم، وأنهُ قد يعطي سبباً لتفسيرِ اضطرابِ شحوم الدم المشاهدِ لدى مرضى الداء السكريِّ من النمطِ الثاني.