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ORIGINAL: Evaluation of the Relationship between Vitamin D Deficiency and Atherogenic Factors in Diabetic Patients with Metabolic Syndrome

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ABSTRACT

Introduction: Metabolic syndrome includes a range of disorders that increase the risk of cardiovascular disease and diabetes mellitus. In this study, we examined the serum level of vitamin D3 in diabetic individuals with metabolic syndrome compared with non-diabetic individuals without metabolic syndrome and the association of serum vitamin D3 levels with metabolic syndrome and atherogenic factor (LDL/HDL).

Material and Methods: In a case-control study, we included 110 women with metabolic syndrome according to ATP III criteria and 127 healthy women as a control group. Serum concentration of total cholesterol, LDL-C, FBS, HDL-C and serum triglyceride (TG) determined by enzymatic method and colorimetric and, serum level 25-(OH) vitamin D determined by ELISA.

Results: It was found that the two healthy and metabolic groups were significantly different in terms of total cholesterol levels, LDL and TG levels, HDL, VLDL, FBS, atherogenic index (LDL/HDL) and vitamin D levels ($P<0.05$). All participants in the control group and the patient and the whole study population were divided into two categories of insufficient and sufficient based on their measured serum concentrations of 25-(OH) vitamin D. There was a significant difference between the group with insufficient levels of vitamin D in comparison with the group with sufficient levels of vitamin D in terms of total cholesterol, LDL and triglyceride levels, HDL, VLDL, FBS and atherogenic index (LDL/HDL) ($P= 0.000$).

Conclusion: The present results showed that there is a significant relationship between level 25-(OH) D and atherogenic index (LDL/HDL) and the incidence of metabolic syndrome.

Introduction

Metabolic syndrome (insulin resistance syndrome) is a set of disorders that increase the risk of cardiovascular disease and diabetes mellitus. The main

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factors of metabolic syndrome include obesity in the central parts of the body, hypertriglyceridemia, hypercholesterolemia and low HDL, hyperglycemia and hypertension (1). According to the criteria of the National Cholesterol Education Program for Adults Treatment Panel III (ATP III), the presence of at least three of the following disorders is considered as a metabolic syndrome: abdominal obesity ≥88 cm for women and 102 cm for men; High-density lipoprotein cholesterol (HDL-c) level <50 mg/dL for women and <40 mg/dL for men, increased triglyceride (TG) level ≥150 mg/dL, Fasting Blood Sugar (FBS) >100 mg/dl, high blood pressure (systolic) blood pressure ≥130 mm Hg or diastolic blood pressure ≥85 mm Hg (2).

The prevalence of metabolic syndrome in Iran based on the definitions of ATPIII, IDF, and WHO is estimated at 50.8, 41.9 and 41.8%, respectively. The underlying disorder of this syndrome appears to be insulin resistance. Obesity, which is a major component of this syndrome, is associated with an increased risk of diabetes, even in mild forms, which is exacerbated in the presence of insulin resistance (3). Type 2 diabetes is a group of disorders that are associated with varying degrees of insulin resistance, impaired insulin secretion, and increased glucose production. Insulin resistance and impaired insulin secretion are among the factors involved in the pathogenesis of type 2 diabetes (4).

Over the past decades, a large number of non-skeletal diseases associated with vitamin D deficiency, including type 2 diabetes, have been identified (4). Vitamin D (vit D) reduces insulin resistance by affecting calcium metabolism and regulating insulin receptor genes. Vit D-mediated increase in cytosolic calcium in muscles tissue appears to be responsible for increased glucose transport to muscle. If insulin secretion is impaired by vit D deficiency, it is improved by administration of 1, 25 $(OH)_2$-D3. Clinical and empirical evidence suggests that serum concentrations of 25-(OH) vit D may be inversely associated with some cancers, type 2 diabetes, metabolic syndrome, and cardiovascular disease (5). In addition, vit D deficiency has been suggested as a possible risk factor for dyslipidemia. Dyslipidemia, characterized by abnormal levels of lipid, is a modifiable risk factor for cardiovascular disease, which is the most common cause of death worldwide and accounts for 32% of worldwide deaths (6). The obvious link between serum vit D levels and metabolic syndrome components has been studied in several studies, some of which have been confirmed, while others have not. One study found that low serum vit D3 levels were inversely related to weight gain and BMI; however, it has no significant relationship with other components of metabolic syndrome (7).

The aim of this study was to evaluate the serum level of vit D3 in diabetic individuals with metabolic syndrome compared to non-diabetic individuals without metabolic syndrome, and to investigate the relationship between serum vit D3 levels with metabolic syndrome and atherogenic indexs (LDL/HDL).

Methods

The present study is a case-control study. We randomly admitted 110 women with definite metabolic syndrome based on clinical findings recorded in Khatam Laboratory of Neka County as a case group. Also, we randomly enrolled 127 healthy women from healthy clients as a control group. They were included in the study if they met the following criteria: not taking any vit D or calcium supplements, and no known illnesses associated with vit D deficiency such as rickets. Exclusion criteria are taking vit D supplements and all known vit D-related disorders. Under the criteria of the National Cholesterol Education Program for Adults in Treatment Panel III (ATP III), metabolic syndrome is defined in the presence of three or more of the following components: waist circumference in men over 102 cm and in women over 88, fasting glycemia ≥100 mg/dl, serum TG levels above 150 mg/dL, HDL-c levels <50 mg/dL in women and <40 mg/dL in men, and hypertension Systolic

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≥130 and/or diastolic blood pressure ≥85 mm Hg). Before starting, we informed all participants about the objectives and details of this study. None of the participants were excluded from our study. This study was designed, compiled and written based on the data recorded in Khatam Laboratory of Neka city.

Clinical measurements

After 12-14 hours of fasting, 10 ml of patients' blood samples were collected and sent to the laboratory to measure lipid profile, FBS and 25-hydroxyvitamin D levels. Blood samples were centrifuged at room temperature for 10 minutes at 3000 rpm. Serum concentrations of total cholesterol, LDL-C, FBS, HDL-C and serum TG were determined by enzymatic and colorimetric methods (Yasin Teb, Karaj, Iran). Serum levels of 25-(OH) vit D are determined by ELISA (Diagnostic Immune Systems, Paris, France). All experiments were performed in Khatam Neka laboratory.

Statistical analysis

Mean data for continuous variables were compared using independent t-test when data were normally distributed. Otherwise, the Mann-Whitney U test will be used. The fit of the classification variables will be compared using the Chi-square test. The ratios of the classification variables will be compared using the Chi-square test or the Fisher's exact test. P-values less than 0.05 are used to determine statistical significance. All analyzes were performed using SPSS 16 software.

Results

In this study, 127 healthy individuals were evaluated as a control group and 110 diabetic patients with metabolic syndrome as a patient group. The mean age of the 237 participants in the study was 47.37 years for people with diabetes with metabolic syndrome and 47.09 years for the control group with an age range of 35 to 60 years. All participants in the study were women. The results were expressed as mean ± standard deviation in both groups. According to the results reported in ***Table 1***, it was found that the two healthy and metabolic groups were significantly different in terms of total cholesterol levels, LDL and TG levels, HDL, VLDL, FBS, atherogenic index (LDL/HDL) and vit D levels (P<0.05).

Table 1. Comparison of studied parameters between two diabetic groups with metabolic syndrome and healthy individuals

Parameter	Mean ± SD		P-value
	Metabolic group (N=110)	Healthy group (N=127)	
Age	47.37±12.15	47.09±12.60	0.8
FBS	120.68±18.22	93.82±12.25	0.000
TG	196.31±51.90	124.06±55.31	0.000
Cholesterol	210.03±33.69	169.43±26.72	0.000
HDL	37.55±5.10	43.22±5.50	0.000
Vit D	18.73±8.54	34.18±4.39	0.000
LDL	133.21±35.35	101.41±31.19	0.000
VLDL	39.26±10.38	24.81±11.26	0.000
Atrogenic index	3.63±1.17	2.43±0.98	0.000

All participants in the control and case groups and the whole study population were divided into two categories of insufficient and sufficient based on their measured serum concentrations of 25-(OH) vit D. According to the results of ***Table 2***, significant differences were observed between the group with insufficient levels of vit D in comparison with the group with sufficient levels of vit D in terms of total cholesterol, LDL and TG levels, HDL, VLDL, FBS and atherogenic index (LDL/HDL) (P= 0.000). ***Table 3*** examined the relationship between vitamin D levels and other parameters in the

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general population. According to ***Table 3***, there was a significant positive correlation between vit D and HDL and a significant negative correlation between vit D and LDL, cholesterol, TG, FBS, VLDL, and atherogenic index.

Table 2. Comparison of the studied parameters in the metabolic syndrome and healthy groups and the whole population based on insufficient and sufficient serum concentrations of 25-(OH) vit D

Parameter	Mean ± SD					
	Metabolic group		Healthy group		Total	
	Vit D <30ng/dl (N=97)	Vit D >30ng/dl (N=13)	Vit D <30ng/dl (N=31)	Vit D >30ng/dl (N=96)	Vit D <30ng/dl (N=128)	Vit D >30ng/dl (N=109)
Age	47.77±12.04	44.38±13.09	47.58±11.58	46.93±12.97	47.72±11.88	46.63±12.95
FBS	121.22±19.29	116.69±4.40	96.41±13.76	92.98±11.67	115.21±20.97	95.81±13.47
TG	195.66±52.77	201.15±46.50	140.16±51.14	118.88±57.16	182.22±57.38	128.69±61.90
Cholesterol	207.75±33.51	227±31.20	164.81±21.2	170.93±28.21	197.35±36.01	177.61±33.79
HDL	37.62±5.28	37±3.6	45.83±5.24	42.35±6.66	39.61±6.33	41.71±6.60
Vit D	16.42±5.93	36±4.01	28.93±0.82	35.88±3.67	19.45±7.46	35.89±3.69
LDL	130.99±34.86	149.77±35.98	90.93±26.64	104.80±31.92	121.29±37.19	110.16±35.42
VLDL	39.13±10.55	40.23±9.30	28.03±10.22	23.77±11.43	36.44±11.47	25.73±12.38
Atrogenic index	3.57±1.15	4.13±1.25	2.04±0.71	2.56±1.03	3.21±1.25	2.75±1.17

Table 3. Correlations of studied parameters with vitamin D in whole study populations

		Age	FBS	TG	Chol	HDL	LDL	VLDL	Atrogenic Index
Vit D (N= 237)	Pearson correlation	-.090	-.546**	-.394**	-.321**	.292**	-.229**	-.394**	-.288**
	Sig. (2-tailed)	.166	.000	.000	.000	.000	.000	.000	.000

** Correlation is significant at the 0.01 level (2-tailed).

Discussion

We found that serum levels of vit D and HDL in patients with metabolic syndrome were significantly lower than the normal group and total cholesterol, LDL and triglyceride, VLDL and FBS levels and atherogenic index (LDL/HDL) in the metabolic syndrome group in Comparison with the control group was significantly higher. Metabolic syndrome (MS) and its consequences are a growing health problem in the last century. Clinical conditions defined by the occurrence of a number of metabolic and vascular changes include central obesity, hypertension, hyperlipidemia, hyperglycemia, insulin resistance, and a prothrombotic state. The main effect of these changes is to increase the risk of type 2 diabetes (8). Metabolic syndrome also increases the risk of atherosclerotic cardiovascular disease and mortality (9). According to NCEPATP III criteria, recent studies have shown that the prevalence of metabolic syndrome in Iran is 41.1% (10). Significant studies have been performed on the relationship between vit D levels and the clinical findings of metabolic syndrome. Some studies have shown an inverse relationship between serum 25-(OH) vit D and insulin resistance, diabetes, and metabolic syndrome (11, 12). Previous studies have shown the role of vit D nuclear receptors in pancreatic β cells in insulin synthesis and secretion. Vit D deficiency can increase the risk of type 2 diabetes, obesity and cardiovascular disease (12, 13). Decreased or no absorption of vit D in the diet, increased catabolism and lack of exposure to sunlight, and kidney or liver disease can cause vit D deficiency (12).

Many studies have shown that vit D deficiency is associated with obesity (14-17). Evidence suggests that obesity is a risk factor for vit D deficiency (18). Liu et al. found that inadequate levels of vit D were higher in obese children than in non-obese children (19). Studies have shown that vit D deficiency is associated with insulin

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resistance and metabolic syndrome (20, 21). On the other hand, other studies did not show an association between inadequate vit D and lipid profiles. One meta-analysis (22) showed that serum levels above 25-(OH) vit D were associated with better lipid profile in children, and another study (14) found that high levels of total cholesterol (TC) and LDL were associated with vit D deficiency. No significant relationship was observed. However, Erol et al. in their research on vit D deficiency and insulin resistance as risk factors for dyslipidemia in obese children showed that vit D deficiency is a risk factor for hyperlipidemia (16). Another study reported that those with vit D deficiency had higher cholesterol and triglycerides than those with sufficient vit D (19, 23). A meta-analysis of 28 studies involving 99,745 participants reported that serum high 25-(OH) vit D levels of 55%, 51%, and 33% were associated with a reduced risk of diabetes, metabolic syndrome, and cardiovascular disease, respectively (24). In their study, Gagnon et al. studied 4,164 adults (mean age 50 years) and reported that serum 25-(OH) vit D was inversely related to waist circumference, TG, and serum FBS levels, but not to HDL-C levels. Blood pressure is not related (25).

However, the association of vit D with glycemic index has been shown in a number of studies (26, 27). The chairman et al. stated that vit D levels in US adolescents were significantly associated with hyperglycemia and metabolic syndrome (27). A 10-year cohort study also showed that serum vit D levels were linked to future glucose levels. Other studies have reported that vit D deficiency is associated with the prevalence of metabolic syndrome (28, 29), but found that there was no association between vit D levels (14). Vit D deficiency is very common in the Iranian population and an inverse relationship between serum vit D and various skeletal and non-skeletal diseases has been reported in the Iranian female population (31, 30). Women with vit D deficiency significantly suffer from skeletal and non-skeletal disorders such as osteoporosis, fractures, and diabetes, which may improve with interventions such as vit D (31). Significant improvement in serum FBS and insulin after vit D treatment was reported in 100 patients with type 2 diabetes in an Arak diabetes clinic (30). In the present study, we examined serum levels of 25-(OH) vit D, FBS, cholesterol, HDL, TG, VLDL and LDL, and atherogenic index (LDL/HDL) in patients with metabolic syndrome compared with controls. All subjects in the control group and metabolic syndrome were divided into two groups based on their serum levels of 25-(OH) vit D sufficient (> 30 ng/ml) levels and insufficient (<30 ng/ml). Inadequate levels of 25-(OH) vit D had the highest level in the patient group compared to the control. LDL, FBS, VLDL, cholesterol, and TG and atherogenic index (LDL/HDL) were significantly higher and HDL was significantly lower in the metabolic syndrome group compared to the control group. We also examined the relationship between metabolic parameters and vit D in the entire study population. In the whole population, there was a significant positive correlation between vit D and serum HDL, but there was a significant negative correlation between vit D with LDL, FBS, VLDL, cholesterol, and TG and serum LDL/HDL atherogenic index.

Conclusion

As a result, the results of current research showed that there is a significant relationship between level 25-(OH) vit D and atherogenic index (LDL/HDL) and the incidence of metabolic syndrome. The findings of this study also showed that the level of 25-(OH) vit D in the metabolic syndrome group is significantly lower compared to the control group.

Ethical standards statement

This study was approved by the Research Ethics Committee of Mazandaran University of Medical Sciences (IR.MAZUMS.REC. 1400.349).

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Conflicts of interest

The authors declare no conflict of interest regarding publication of this article.

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Authors' contributions

All authors have intellectually committed to the study design and process. The final manuscript was revised and accepted by all authors.

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