ORIGINAL SCIENTIFIC ARTICLE

ASSOCIATIONS OF VITAMIN D LEVEL AND GLUCOREGULATORY PARAMETERS IN TYPE 2 DIABETES MELLITUS

Andrea Zubnar[1,2], Stanislava Nikolic[1,3], Damir Benc[1,4], Nebojsa Janjic[1,5], Mina Maricic[1,2], Danijel Slavic[6], Borislav Tapavicki[1], Aleksandra Popovic[6] and Dea Karaba Jakovljevic[6]

[1] *University of Novi Sad, Faculty of Medicine, Novi Sad, Serbia*
[2] *Institute for Child and Youth Health Care of Vojvodina, Novi Sad, Serbia*
[3] *Clinical Center of VojvodinaCenter of laboratory medicine, Novi Sad, Serbia*
[4] *Clinical center of Vojvodina, Clinic for endocrinology, Diabetes and metabolic disorders, Novi Sad, Serbia*
[5] *Center for gastroenterology and hepatology, Clinical center of Vojvodina, Novi Sad, Serbia*
[6] *University of Novi Sad, Department of Physiology, Faculty of Medicine,Novi Sad, Serbia*

Received: 24.08.2019.
Accepted: 17.10.2019.

Corresponding author:

Danijel Slavic,

Faculty of Medicine
University of Novi Sad Novi Sad, Serbia

E-mail: danijel.slavic@mf.uns.ac.rs

ABSTRACT

Vitamin D is known to affect the functions of pancreatic beta cells, but the effects of vitamin D deficiency on glucoregulatory mechanisms are still inconclusive. The aim of this study was to link vitamin D levels with parameters of insulin resistance and insulin secretion. The study included 70 male and female participants, 40 newly diagnosed patients with type 2 diabetes mellitus (T2DM) and 30 healthy controls. All participants were tested for fasting glucose, hemoglobin A1c, fasting insulin, vitamin D levels, and the HOMA indexes were calculated using HOMA2 calculator. Fasting glucose levels, insulinemia, hemoglobin A1c levels and HOMA IR were all significantly higher in the diabetic group ($p<0.001$), while vitamin D levels and HOMA S index were significantly lower ($p<0.001$). HOMA-B values did not differ between the two groups ($p=0.31$). Vitamin D levels moderately correlated with HOMA S and HOMA B indexes ($r=0.466$, $p<0.001$; $r=0.394$, $p<0.001$, respectively), whereas a negative correlation was found between vitamin D levels and HOMA IR ($r=-0.285$; $p<0.001$). Multiple regression analysis showed that vitamin D levels significantly predicted the values of HOMA B index ($p=0.001$), but they had no predictive value on HOMA IR ($p=0.26$). In conclusion, the group of newly diagnosed patients with T2DM showed significantly lower vitamin D values compared to the healthy control group. The connection between vitamin D, glucose levels, hemoglobin A1c and insulin secretion index underlines the role of this vitamin in glucoregulation.

Keywords: *vitamin D, insulin resistance, insulin secretion, HOMA index, glu-cose metabolis.*

UDK: 616.379-008.64:577.161.2
615.356:577.161.2
Ser J Exp Clin Res 2022; 23(2):141-146
DOI: 10.2478/sjecr-2019-0049

INTRODUCTION

Some of the most common chronic metabolic disorders of present time are insulin resistance and vitamin D deficiency, and this is probably due to common risk factors, such as lack of physical activity, less sun exposure, bad dietary habits based on increased intake of concentrated carbohydrates and lower intake of dairy products and fibers, as well as chronic stress (1).

A published study has shown that individuals with vitamin D deficiency have reduced insulin secretion (2). Vitamin D receptors (VDR) are found in pancreatic β cells which also possess the 1-alpha hydroxylase enzyme essential for the synthesis of vitamin D, providing autocrine and paracrine biological effects (1,3,4). Furthermore, there is a study which suggests that vitamin D promotes increased synthesis of the mRNA for the preproinsulin of the pancreatic β cells in mice (1). The foregoing facts could indicate that vitamin D plays a significant role in physiological regulation of the endocrine pancreas. Additionally, it is believed that vitamin D may also affect the sensitivity of the peripheral insulin receptors due to the existence of VDRs in skeletal muscle, adipose tissue and hepatocytes.

Data considering the effects of vitamin D substitution on insulin resistance and secretion are still inconclusive. In some studies, vitamin D supplementation did not have any effect on insulin resistance in patients with prediabetes and type 2 diabetes mellitus patients (T2DM) (5,6). On the other hand, some studies conducted on T2DM patients and patients with impaired fasting glucose, showed that vitamin D substitution results in significant decrease of blood glucose levels, as well as improvement of calculated indexes of insulin resistance and/or insulin secretion (7-9).

Therefore, the aim of this study was to establish vitamin D status in patients with newly diagnosed T2DM, and to examine the link between vitamin D levels and the parameters of insulin resistance and secretion.

PATIENTS AND METHODS

This cross-sectional study was conducted in the Center of Laboratory Medicine in cooperation with the outpatient department of the Clinic for Endocrinology, Diabetes and Metabolic Disorders, Clinical Center of Vojvodina, during the summer and autumn of 2016. The study included 70 participants (57 male and 13 female). The mean age of the participants was 56.3 ± 5.1 for males and 55.8 ± 6.0 for females. Based on the patient history, clinical examination and laboratory analysis (fasting plasma glucose > 7 mmol/L and/or HbA1c > 48 mmol/mol), 40 participants were classified into a group of T2DM (10). All T2DM patients in this study were newly diagnosed and therapy-naïve. The control group consisted of 30 healthy (age and sex matched) subjects (individuals with normal fasting plasma glucose, insulin and glycated hemoglobin A1c levels).

Criteria for exclusion from the study were: kidney disease, psychiatric disorders and the presence of metabolic bone disease, as well as supplementation with vitamin D and/or calcium.

Laboratory assay

All laboratory analyses were performed before implementing any treatment of T2DM. After an eight-hour overnight fast and 15-minute rest in the laboratory after the arrival, blood samples were taken in order to determine fasting plasma glucose (FPG) and insulin (FPI) concentration, vitamin D level (25(OH)D) and glycated hemoglobin A1c (HbA1c). Glucose level was determined by an enzyme-specific, GOD-Perid method (reference range from 4.0 to 6.1 mmol/L) and the insulin was measured on an automated system ADVIA Centaur XP (reference value is 3.0 to 25.0 mIU/L). 25(OH)D was measured by direct chemiluminescent technology on platform ADVIA Centaur XP. Vitamin D status was defined according to blood levels of 25(OH)D as follows: vitamin D deficiency as 25(OH)D below 30 nmol/L, and vitamin D insufficiency as 25(OH)D of 30 to 50 nmol/L, while normal vitamin D status as 25(OH)D concentration greater than 50 nmol/L (11). HbA1c was determined by immunoturbidimetric method of inhibition of agglutination on the microparticles, on an automated system (Abbott Architect ci 4100). HOMA indexes were estimated using HOMA 2 calculator: HOMA index for estimation of insulin resistance (HOMA-IR), insulin sensitivity (HOMA-S) and secretory capacity of pancreatic β cells (HOMA-B) (12).

Data were presented using descriptive statistical methods for continuous variables as mean and standard deviation. In order to evaluate differences in parametric variables we used Student's T-test. Relations among variables were assessed using Pearson's correlation coefficient. Using linear correlation analysis, we estimated the relationship between all examined parameters and 25(OH)D. Multiple regression analysis was performed in order to estimate the independent relation between 25(OH)D levels and calculated parameters of insulin resistance/sensitivity as well as insulin secretion. A 2-tailed p-value of less than 0.05 was considered statistically significant. Statistical analysis was performed using the Data Analysis Excel (Microsoft Corp., Redmond, WA) and MedCalc 12.1.4.0 statistical software (MedCalc Software, Mariakerke, Belgium).

RESULTS

Comparing T2DM patients with healthy controls, statistically significant difference was found for all tested parameters except for HOMA-B, ($p=0.31$). Compared with the control group, the T2DM group had significantly lower vitamin D levels ($p<0.001$) and HOMA-S ($p<0.001$) index, and significantly higher HOMA-IR index, BMI, FPG, FPI and HbA1c ($p<0.001$) (Table 1). In the T2DM group, 30 out of

40 participants were D vitamin insufficient (D vitamin below 50 nmol/l), while all healthy controls had vitamin D level above 50 nmol/l ($p<0.001$).

Using linear correlation analysis on both examined groups together (N=70), vitamin D levels moderately correlated with HOMA S and HOMA B indexes, whereas a negative correlation was found between vitamin D levels and HOMA IR. Negative correlations were also determined between 25(OH)D and FPG and HbA1c. However, we did not establish a correlation between 25(OH)D and FPI (Table 2).

Multiple regression analysis was used to assess the ability of the predictor model containing 25(OH)D and BMI to predict the values of HOMA IR, HOMA B% and HOMA S. As presented in Table 3, the model could successfully explain 48%, 41% and 13% of their respective variances. 25(OH)D was found to be a significant predictor of HOMA S and HOMA B, but not HOMA IR. As for BMI, it was a significant predictor of HOMA S and HOMA IR, but not HOMA B.

Table 1. Characteristics of glucoregulatory parameters of two examined groups

VARIABLE	T2DM group n=40	Control group n=30	P value
	X±SD	X±SD	
BMI (kg/m²)	29.94±5.77	22.05±1.89	p<0.001
FPG (mmol/L)	7.86±1.98	4.77±0.39	p<0.001
FPI (mIU/L)	17.24±7.07	7.40±4.64	p<0.001
HbA1c (mmol/mol)	47.9±17.72	31.1±4.01	p<0.001
HOMA-IR	2.82±1.12	0.96±0.58	p<0.001
HOMA-S (%)	42.07±18.98	135.86±70.88	p<0.001
HOMA-B (%)	88.15±40.19	98.45±28.74	p=0.31
25(OH)D (nmol/L)	45.17±20.65	70.7±16.33	p<0.001

Legend: BMI - body mass index; FPG - fasting plasma glucose; FPI - fasting plasma insulin; HOMA-IR - index for estimation of insulin resistance; HOMA-S - index for estimation of insulin sensitivity; HOMA-B - index for estimation of secretory capacity of pancreatic β cells; 25(OH)D - 25 hydroxyvitamin D; p - value

Table 2. Correlation between vitamin D levels and examined parameters

	25(OH)D N=70	
	r	P
BMI (kg/m²)	-0.261	p<0.001
FPG (mmol/L)	-0.600	p<0.001
FPI (mIU/L)	-0.201	p>0.05
HbA1c (mmol/mol)	-0.616	p<0.001
HOMA-IR	-0.285	p<0.001
HOMA-S (%)	0.466	p<0.001
HOMA-B% (%)	0.394	p<0.001

Legend: BMI-body mass index; FPG-fasting plasma glucose; FPI- fasting plasma insulin; HOMA-IR- index for estimation of insulin resistance; HOMA-S- index for estimation of insulin sensitivity; HOMA-B-index for estimation of secretory capacity of pancreatic β cells; 25(OH)D - 25 hydroxyvitamin D; r - correlation coefficient; p - value

Table 3. Multiple regression analysis

Dependent variable	Adjusted R^2	P value	Predictors		
N=70					
				β	p(β)
HOMA-IR	0.484	p<0.001	BMI	6.986	<0.001
			25(OH)D	-1.144	0.257
HOMA-S (%)	0.412	p<0.001	BMI	-4.693	<0.001
			25(OH)D	3.316	0.001
HOMA-B (%)	0.13	0.006	BMI	0.532	0.596
			25(OH)D	3.297	0.001

Legend: BMI - body mass index; FPG - fasting plasma glucose; FPI - fasting plasma insulin; HOMA-IR - index for estimation of insulin resistance; HOMA-S - index for estimation of insulin sensitivity; HOMA-B - index for estimation of secretory capacity of pancreatic β cells; 25(OH)D - 25 hydroxyvitamin D; Adjusted R^2 - the coefficient of determination that is consistent with the number of independent variables included in the model; β - standardized regression coefficient; p - p value of standardized regression coefficient

DISCUSSION

The main purpose of this study was to compare vitamin D levels in T2DM and healthy controls and to examine the relationship between vitamin D and insulin secretion and resistance indexes. The results showed that the T2DM group had significantly lower vitamin D levels compared with the control group. Also, 75% of patients with T2DM were vitamin D insufficient.

It has not been clarified yet if low vitamin D status is responsible for insulin resistance, or if it is just a consequence of the prediabetic state, including obesity, which exists together with insulin resistance. One of the major cohort studies concluded that reduced vitamin D levels lead to an increased risk of T2DM (13). Also, published results showed that people who have been substituting vitamin D or calcium had a lower risk of T2DM (14). The absence of the anti-inflammatory effect, which is attributed to reduced levels of vitamin D, can contribute to the development of insulin resistance, and therefore disrupt the glucoregulatory mechanisms, as well as the onset of T2DM (1). On the other hand, it is known that people with a higher BMI have lower levels of vitamin D due to the sequestration of liposoluble vitamin D in the larger adipose compartment, so this vitamin would be low in the serum (15). On the other hand, Drincic et al. (16) concluded that simple volumetric dilution in obese patients can be the explanation for the low vitamin D status in obesity.

Compared with the control group, the T2DM group had significantly higher HOMA-IR and insulin levels and lower HOMA-S values. The obtained results are expected and are in accordance with the previously known literature data. The pathophysiological basis of T2DM refers to development of insulin resistance, and therefore, significantly higher fasting insulin levels in our T2DM group represents an attempt to overcome the present insulin resistance (17). On the other hand, the T2DM group had a lower HOMA-B than the control group (88.15 ± 40.19% vs. 98.45 ± 28.14%), but without statistical significance. HOMA-B value depends on the level of insulin resistance, and the functional ability of pancreatic β cells. Thus, the reduction in insulin secretion can result from the preserved sensitivity of the peripheral tissues, and therefore a lower need for insulin secretion. On the other hand, after the hyperinsulinemic phase in T2DM, the insulin secretion becomes insufficient and reduced in relation to the level of current glycemia. Thus, HOMA-B values may be lower in healthy subjects, but also, as a consequence of the relative exhaustion of pancreatic β cells and a gradual decrease in insulin secretion, in T2DM patients. Also, knowing that HOMA-B is calculated using FPI and FPG, this index shows whether this insulin secretion is capable of maintaining fasting glucose in a normal range. We can see that in our control group HOMA-B is near the value of 100% which means that insulin secretion is optimal for that glucose level, whereas in the diabetic group, although fasting insulin is higher than in the control group, that insulin level is not capable of maintaining glucose level in euglycemic range, and that is the reason for lower HOMA B index in this group.

Results of this study also showed a statistically significant relationship between 25(OH)D and HOMA-B which is in accordance with previously published studies (18). Using multiple regression analysis, we have found that vitamin D is a predictor of insulin secretion estimated by HOMA-B index. It is known that the insulin secretion is glucose dependent due to the activation of GLUT-2 transporters, ATP synthesis, blockage of ATP-dependent potassium channels, as well as consequent depolarization of the cell membrane and influx of calcium ions. Cell influx of Ca^{2+} ions directly stimulates exocytosis and the release of insulin molecules into the

circulation. In addition to this mechanism, the second pathway of regulation of insulin secretion is also indicated, via cAMP (19,20). Besides the glucose-dependent, there are also glucose independent pathways of insulin secretion (for example the effects of incretin, amino-acids or glucagon on β-cells) (21,22). The presence of VDR receptors and 1α-hydroxylase in the pancreatic β cells suggests the possibility of a regulatory role of vitamin D on the intracellular concentration of Ca^{2+} ions, as well as the effect of vitamin D on the transcription of the preproinsulin gene (1,23). The second theory is based on the fact that insulin secretion can be independently induced by initiators (glucose, Ca^{2+}) as well as potentiators (glucagon, acetylcholine, β-adrenergic receptor agonists, cAMP) which enhance insulin secretion, but only in the presence of glucose as the most dominant initiator. According to this hypothesis, vitamin D can affect insulin secretion by acting on the efficacy of the initiator and/or potentiator, modulating the effects of the initiator of insulin secretion (23). Vitamin D deficiency causes a change in the efficiency of the glucose-dependent cAMP pathway and, consequently, its role in the regulation of insulin secretion (24). As a confirmation of this theory, numerous studies on animals have been published. They showed the influence of vitamin D on the cAMP signaling pathway, as well as changes in the activity of the cAMP-dependent protein kinase in vitamin D deficient animals (24-26). On the other hand, a clinical study which observed changes in the level of insulin secretion during parenteral administration of vitamin D concluded that an increase in insulin secretion was found only in people with elevated glycemia above the euglycemic range, whereas in subjects with normal glycemia, no increase occurred (27-29). Jeddi et al. had similar results on experimental animals (30).

According to our results, significant negative correlation was found between 25(OH)D and HOMA-IR which is in accordance with previously published data (18,29,31). However, multiple linear regression analysis has shown that the level of vitamin D influence on HOMA-IR is statistically insignificant. This finding is in line with the latest findings concerning the association between vitamin D and insulin resistance (1). Also, multiple linear regression showed that vitamin D is a significant predictor of HOMA-S, an index that represents the sensitivity of peripheral tissues to insulin. Both HOMA-IR and HOMA-S indexes are relatively close and point to the similar occurrences from a different perspective. However, the parameters used to calculate these indexes are different. In the HOMA-IR equation, only the basal values of glucose and insulin are used, whereas the estimation of HOMA-S also includes a deviation factor from normal values characteristic for healthy population (100%) (32). Different models of these indexes also point to different aspects of insulin effects on peripheral tissues (12). The effect which vitamin D potentially achieves in insulin sensitive tissues is still not well-known and explained in the literature. The local concentration of vitamin D and the production of hormone D in tissues, which directly affect the level of insulin resistance or sensitivity, cannot be fully identified with the measured level of vitamin D in systemic circulation.

CONCLUSION

Our study showed that newly diagnosed T2DM patients had significantly lower vitamin D levels compared with healthy controls. The relation between vitamin D levels and glucoregulatory parameters such as glucose levels, hemoglobin A1c and insulin secretion index (HOMA-B) underlines the glucoregulatory role of this vitamin, primarily due to its influence on the secretory function of pancreatic β cells.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

The study was conducted in accordance with the ethical standards of the committee responsible for human experimentation (institutional and national) and the Helsinki Declaration of 1975, as revised in 2013. Voluntary written and informed consent was obtained from each participant prior to enrollment in the study.

COMPETING INTERESTS

There are no conflicts of interest.

FUNDING

None.

REFERENCES

1. KAl-Shoumer ASK, Al-Essa MT. Is there a relationship between vitamin D with insulin resistance and diabetes mellitus. World J Diabetes.2015; 6(8):1057-64.
2. Gedik O, Akahn S. Effects of vitamin D deficiency and repletion on insulin and glucagon secretion in man. Diabetologia. 1986;29(3):142-5.
3. Palomer X, Gonzalez-Clemente JM, Blanco-Vaca F, Mauricio D. Role of vitamin D in the pathogenesis of type 2 diabetes mellitus. Diabetes Obes Metab. 2008; 10(3):185-97.
4. Naumovic N. Vitamin D- physiological importance. Med Pregl. 2010; 63 (5-6):301-4.
5. Witham MD, Dove FJ, Dryburgh M, Sugden JA, Morris AD, Struthers AD. The effect of different doses of vitamin D on markers of vascular health in patients with type 2 diabetes: a randomised controlled trial. Diabetologia. 2010;53(10):2112-9.
6. Jorde R, Figenschau Y. Supplementation with cholecalciferol does not improve glycaemic control in diabetic subjects with normal serum 25-hydroxyvitamin D levels. Eur J Nutr. 2009;48(6):349-54.
7. Talaei A, Mohamadi M, Adgi Z. The effect of vitamin D on insulin resistance in patients with type 2 diabetes. Diabetol Metab Syndr. 2013;5(1): 8.
8. Kumar S, Davies M, Zakaria Y, Mawer EB, Gordon C, Olukoga AO, Boulton AJ. Improvement in glucose tolerance and beta-cell function in a patient with vitamin D

deficiency during treatment with vitamin D. Postgrad Med J. 1994;70(824):440-3.
9. Pittas AG, Harris SS, Stark PC, Dawson-Hughes B. The effects of calcium and vitamin D supplementation on blood glucose and markers of inflammation in nondiabetic adults. Diabetes Care. 2007;30(4):980-6.
10. American Diabetes Association. Classification and diagnosis of diabetes. Sec. 2. In *Standards of Medical Care in Diabetes-2015*. Diabetes Care 2015;38(Suppl. 1): S8-S16
11. Munns CF, Shaw N, Kiely M, et al. Global Consensus Recommendations on Prevention and Management of Nutritional Rickets. J Clin Endocrinol Metab. 2016; 101(2):394-415.
12. Wallace T, Levy J, Matthews D. Use and abuse of HOMA modeling. Diabetes Care. 2004; 27(6):1487-95.
13. Mattila C, Knekt P, Mannist OS, Rissanen H, Maarit A. Laaksonen M, et al. Serum 25-Hydroxyvitamin D Concentration and Subsequent Risk of Type 2 Diabetes. Diabetes care.2007;30(10):2569-70.
14. Pittas AG, Dawson-Hughes B, Li T, Van Dam RM, Willett WC, Manson JE, et all. Vitamin D and calcium intake in relation to type 2 diabetes in women. Diabetes Care. 2006; 29(3):650-6.
15. Mcgill AT, Stewart JM, Lithander FE, Strik CM, Poppitt SD. Relationships of low serum vitamin D3 with anthropometry and markers of the metabolic syndrome and diabetes in overweight and obesity. Nutr J. 2008;28;7:4.
16. Drincic A, Armas L, Van Diest E, Heaney R. Volumetric Dilution, Rather Than Sequestration Best Explains the Low Vitamin D Status of Obesity. Obesity (Silver Spring). 2012;20(7):1444-8.
17. Bonora E, Formentini G, Calcaterra F, Lombardi S, Marini F, Zenari L, et all. HOMA- Estimated Insulin Resistance Is an Independent Predictor of Cardiovascular Disease in Type 2 Diabetic Subjects. .Diabetes Care. 2002; 25(7):1135-41.
18. Yoon H, Jeon DJ, Park CE, You HS, Moon AE. Relationship between homeostasis model assessment of insulin resistance and beta cell function and serum 25-hydroxyvitamin D in non-diabetic Korean adults. J Clin Biochem Nutr. 2016;59(2):139-144.
19. Schuit FC. Factors determining the glucose sensitivity and glucose responsiveness of pancreatic beta cells. Horm Res. 1996;46(3):99-106.
20. Tengholm A. Cyclic AMP dynamics in the pancreatic b-cell. Ups J Med Sci. 2012; 117(4):355-69.
21. Seino Y, Kurahachi H, Goto Y, Taminato T, Ikeda M, Imura H. Comparative insulinogenic effects of glucose, arginine and glucagon in patients with diabetes mellitus, endocrine disorders and liver disease. Acta Diabetol Lat. 1975;12(2):89-99.
22. Drucker DJ, Philippe J, Mojsov S, Chick WL, Habener JF**.** Glucagon-like peptide I stimulates insulin gene expression and increases cyclic AMP levels in a rat islet cell line**.** PNAS. 1987;84(10):3434-8.
23. Bourlon P-M, B Billaudel B, Faure-Dussert A. Influence of vitamin D3 deficiency and 1,25 dihydroxyvitamin D3 on de novo insulin biosynthesis in the islets of the rat endocrine pancreas. J Endocrinol. 1999;160(1):87-95.
24. Berg JP, Haug E.Vitamin D: a hormonal regulator of the cAMP signaling pathway. Crit Rev Biochem Mol Biol. 1999;34(5):315-23.
25. Rudack-Garcia D, Henry H. Effect of Vitamin D Status on Cyclic AMP-dependent Protein Kinase Activity and Its Heat-stable Inhibitor in Chick Kidney. J biol chem. 1981; 256(21):10781-5.
26. Vazquez G, Boland R, de Boland A. Modulation by $1,25(OH)_2$-vitamin D_3 of the adenylyl cyclase/cyclic AMP pathway in rat and chick myoblasts. BBA-mol cell res. 1995;1269(1):91-7.
27. Nilas L, Christiansen C. Treatment with vitamin D or its analogues does not change body weight or blood glucose level in postmenopausal women. Int J Obes. 1984;8(5): 407-11.
28. Boucher BJ, Mannan N, Noonan K, et al.Glucose intolerance and impairment of insulin secretion in relation to vitamin D deficiency in east London Asians. Diabetologia. 1995;38(10):1239-45.
29. von Hurst PR, Stonehouse W, Coad J. Vitamin D supplementation reduces insulin resistance in South Asian women living in New Zealand who are insulin resistant and vitamin D deficient - a randomised, placebo-controlled trial. Br J Nutr. 2010;103(4):549-55.
30. Jeddi S, Syedmoradi L, Bagheripour F, Ghasemi A. The Effects of Vitamin D on Insulin Release From Isolated Islets of Rats. IntJEndocrinolMetab. 2015;13(1):e20620.
31. Chiu KC, Chu A, Go VL, Saad MF. Hypovitaminosis D is associated with insulin resistance and beta cell dysfunction. Am J ClinNutr. 2004;79(5):820-5.
32. Hedblad B, Nilsson P, Janzon L, Berglund G: Relation between insulin resistance and carotid intima-media thickness and stenosis in non-diabetic subjects. Results from a cross-sectional study in Malmo,Sweden. Diabet Med 2000;17:299-307.