Vol 25 No. 2 March 2019

p-ISSN 0854-4263
e-ISSN 4277-4685

INDONESIAN JOURNAL OF
CLINICAL PATHOLOGY AND MEDICAL LABORATORY
Majalah Patologi Klinik Indonesia dan Laboratorium Medik

CONTENTS

RESEARCH

LITERATURE REVIEW

CASE REPORT

INDONESIAN JOURNAL OF
CLINICAL PATHOLOGY AND MEDICAL LABORATORY
Majalah Patologi Klinik Indonesia dan Laboratorium Medik

2019 March; 25(2): 150-154
p-ISSN 0854-4263 e-ISSN 2477-4685
Available at www.indonesianjournalofclinicalpathology.org

ANALYSIS OF VITAMIN D IN PATIENTS WITH TYPE 2 DIABETES MELLITUS

Arfandhy Sanda[1], Uleng Bahrun[1], Ruland DN. Pakasi[1], Andi Makbul Aman[2]

[1] Department of Clinical Pathology, Faculty of Medicine, Hasanuddin University/Wahidin Sudirohusodo Hospital, Makassar, Indonesia. E-mail: arfandhysanda@gmail.com
[2] Department of Internal Medicine, Faculty of Medicine, Hasanuddin University/Wahidin Sudirohusodo Hospital, Makassar, Indonesia

ABSTRACT

Diabetes Mellitus (DM) is a metabolic disease which is still a public health problem until now. World Health Organization (WHO) and the International Diabetes Federation (IDF) predict an increase in the number of people with diabetes which become one of the global threats. Some evidence showed that vitamin D deficiency might influence the pathogenesis of type 2 DM caused by insulin resistance and dysfunction of the pancreatic beta-cell. This study was aimed to analyze the differences of vitamin D levels in three groups of DM (prediabetic, controlled, and uncontrolled DM), and to identify the correlation of vitamin D levels with severity of DM. The method used was a cross-sectional study. During the study period, total of 93 subjects of type 2 DM patients (who met the study criteria) were obtained. The subjects consisted of 33 prediabetic subjects, 30 controlled types 2 DM subjects, and 30 subjects of uncontrolled types 2 DM. The study sample consisted of 49(52.7%) males and 44(47.3%) females with a minimum age was 20 years, and a maximum age was 79 years (mean 56.59+12.15 years). The minimum HbA1c level was 4.8%, and the maximum level was 12.9% (mean 6.95+1.81%). The minimum vitamin D level was 9.07 ng/mL, and the maximum level was 66.49 ng/mL (mean 26.85+9.30 ng/mL). Kruskal-Wallis test showed a p-value=0.132, Spearman correlation test showed a p-value>0.05. The conclusion from this study there was no significant correlation between vitamin D level and type 2 DM consisting of a prediabetic group, controlled type 2 DM, and uncontrolled type 2 DM.

Key words: Type 2 diabetes mellitus, vitamin D, HbA1c

INTRODUCTION

Diabetes Mellitus (DM) is a metabolic disease with ahyperglycemic manifestation caused by insulin secretion disorder, insulin function, and both. Type 2 DM is a hyperglycemic disease caused by cell insensitivity towards insulin. This disease is also called Non-Insulin Dependent Diabetes Mellitus (NIDDM) because insulin is not capable of exciting the glucose absorption in peripheral organ targets like muscles and fat which results to hyperinsulinemia, glucose intolerance, and insulin resistance.[1-3]

Some epidemiological research showed the increase of DM type 2 incidence in the whole world. World Health Organization (WHO) predicts an increase in DM patients which has become a global health threat. The WHO predicted an increase in the population of DM patients in Indonesia from 8,4 million in 2000 to 21,3 million in 2030. Another world organization like the Internation Diabetes Federation (IDF) predicted an increased population of DM patients in Indonesia from 9,1 million in 2014 to 14,1 million in 2035.[2,4]

Diabetes Mellitus diagnosis is based on the American Diabetes Association (ADA) 2013 which consists of: glycated hemoglobin (HbA1c) level >6.5% with fasting blood glucose (GDP)>126 mg/dL, or 2 hours post-prandial blood glucose (GD2PP) >200 mg/dL, or patients with classic symptoms of hyperglycemia (polyuria, polydipsia, polyphagia). In the last few decades, it has been reported about many non-skeletal diseases dealing with vitamin D deficiency, one of them is type 2 DM. Vitamin D deficiency is a risk factor of diabetes in many Americans and Africans, besides other risk factors such as obesity, dietary habit, and lack of physical activities. Some evidence showed that vitamin D deficiency might be influential in type 2 DM pathogenesis caused by insulin resistance and pancreatic beta cell dysfunction. Otherwise, adequate consumption of vitamin D supplement could improve the insulin secretion and decrease the risk of insulin resistance.[3-5]

Research by Chiu *et al*. showed a positive correlation between vitamin D level with insulin sensitivity and pancreatic beta-cell function associated with vitamin D deficiency. This research

showed that hypovitaminosis D could be the independent risk factor of insulin resistance, type 2 DM, and metabolic syndrome. Study research by Tsur *et al*. showed that vitamin D could be the independent risk factor for the development of impaired glucose tolerance and diabetes. Another analysis by Deleskog *et al*. showed that high vitamin D levels in the blood could predict the decrease of type 2 DM risk in prediabetic individuals, but not on normal glucose tolerance, so it showed that vitamin D supplementation must be evaluated to preventing type 2 DM in prediabetic individuals.[4,6,7]

The adequacy of vitamin D consumption has been proven to avoid the risk of type 2 DM. This effect was particularly connected with the immunomodulator function of vitamin D. The lack of vitamin D consumption caused decreased insulin secretion. Besides that, the presence of Vitamin D Receptor (VDR) and vitamin D-Binding Protein (DBP) could affect glucose tolerance and insulin secretion causing the risk of type 2 DM.[8,9]

Prospective research by Pittas *et al*. showed a connection between vitamin D consumption and DM. The role of vitamin D in the regulation of pancreatic beta cell has been proven by the invention of VDR in pancreatic beta-cell capacity and disruption of insulin secretion which has a lack of vitamin D functional receptors. A study in New Zealand reported that new patients who were newly diagnosed with type 2 DM or impaired glucose tolerance had low vitamin D levels compared with healthy control subjects. An epidemiologic data in Bangladesh showed the decrease levels of vitamin D in a risk population of type 2 DM.[10-12]

Incidence of DM patients, and studies about vitamin D levels in controlled and uncontrolled type 2 DM, had never been done in Indonesia, so the authors were interested to do a research about vitamin D levels in prediabetic, controlled and uncontrolled DM patients in order to find the connection between vitamin D levels and incidence of DM type 2. This study was aimed to analyze the differences of vitamin D levels in three groups of DM (prediabetic, controlled, and uncontrolled DM), and to identify the correlation of vitamin D levels with severity of DM.

METHODS

The study design was anobservational study with across-sectional study approach. The samples were taken in the Internal Disease Outpatient subdivision of Metabolic Endocrine and Internal Disease Room Care Wahidin Sudirohusodo Hospital Makassar, Makassar city Regional Hospital, one hundred twenty-three Clinics, and Laboratories in Makassar. Laboratory examination was done in the Clinical Pathology Laboratory Installation of Wahidin Sudirohusodo Hospital Makassar, Hasanuddin University Educational Hospital, and Research Laboratory Unit, Medical Faculty of Hasanuddin University. The study was started in August 2017 until the amount of the sample was enough.

The study population was all type 2 DM patients who underwent examination in Endocrine Outpatient Clinic and Internal Medicine Ward of Wahidin Sudirohusodo Hospital, Hasanuddin University Educational Hospital, Makassar city regional hospital, and one hundred twenty-three Clinics, and Laboratories in Makassar. Inclusion criteria were adults with age 18-60 years who came to the Endocrine Outpatient Clinic of the Wahidin Sudirohusodo Hospital and after they underwent physical and laboratory examination, were diagnosed with type 2 DM patients, willing to take part in the study by filling in and signing the informed consent. Exclusion criteria were type 2 DM patients who had vitamin D and calcium therapy, who had chronic kidney disease, liver failure, obesity, and gastrointestinal disorder, a medical history such as consuming anti-seizure drugs, glucocorticoid, rifampicin, antiretroviral drugs, lysed, hemolytic, and icteric serum.

RESULT AND DISCUSSION

About 93 subjects were studied, who consisted of 33 prediabetic subjects, 30 controlled types 2 DM subjects, and 30 uncontrolled types 2 DM subjects. The characteristics such as age, gender, HbA1c, and vitamin D level is shown in Table 1.

Table 2 explained the characteristics of subjects, HbA1c level, and vitamin D showing results about the age range between 20-79 years with a mean of 56.6 years, HbA1c level range between 4.8-12.9% with a mean of 6,95%, and vitamin D level range showing results about the range between 4.8-12.9% with a mean of 6.95%, and the range of vitamin D levels between 9.07-66.49 ng/mL with a vitamin D mean level of 26.85 ng/mL.

The comparison of vitamin D levels in the prediabetic, controlled DM, and uncontrolled DM group is stated inTable 3.

Table 3 showed that the vitamin D levels (median and mean) were found lower in prediabetic than controlled DM and uncontrolled DM, but statistically, test results showed that the difference was not significant ($p>0.05$). The comparison of vitamin D

Table 1. Sample characteristics based on age and gender group

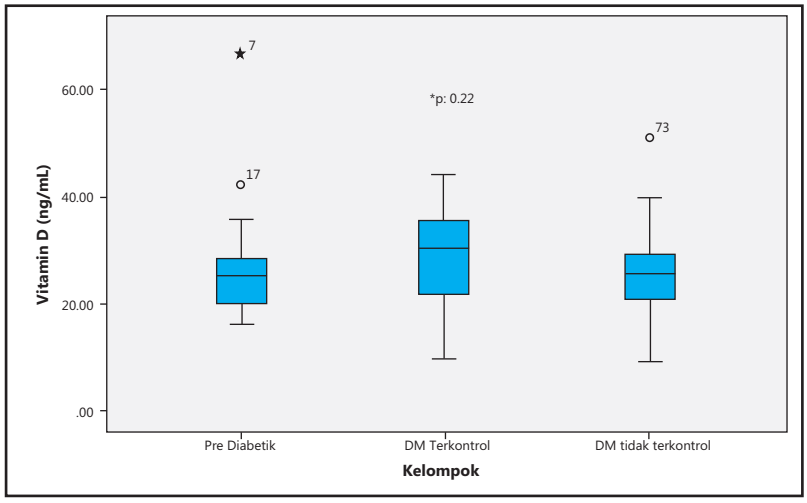
Variable	Sampel group (n=93)			Total (n=93)
	Prediabetic (n=33)	Controlled DM (n=30)	Uncontrolled DM (n=30)	
Age group (years)				
<40	6 (18.2%)	2 (6.7%)	2 (6.7%)	10 (10.8%)
40-49	4 (12.1%)	5 (16.7%)	5 (16.7%)	14 (15.1%)
50-59	12 (36.4%)	6 (20%)	9 (30%)	27 (29.0%)
60-69	9 (27.3%)	9 (30%)	11 (36.7%)	29 (31.2%)
>70	2 (6.1%)	8 (26.7%)	3 (10.0%)	13 (14.0%)
Gender				
Males	16 (48.5%)	23 (76.7%)	10 (33.3%)	49 (52.7%)
Females	17 (51.5%)	7 (23.3%)	20 (66.7%)	44 (47.3%)

Table 2. Sample data distribution based on age, HbA1c, and vitamin D levels

Variable	Minimum	Maximum	Mean	Mean	SD	Distribution data*
Age (years)	20	79	58.00	56.59	12.15	Normal
HbA1c (%)	4.8	12.9	6.30	6.95	1.81	abnormal
Vitamin D (ng/mL)	9.07	66.49	25.87	26.85	9.30	abnormal

Table 3. Vitamin D levels in sample groups

Group	Vitamin D levels (ng/mL)				
	Minimum	Maximum	Median	Mean	SD
Pre-diabetic	15.71	66.49	25.30	26.32	9.21
Controlled DM	9.40	44.14	30.32	28.87	10.03
Uncontrolled DM	9.07	51.29	25.65	25.41	8.56



*Kruskal-Wallis test

Figure 1. Comparison of vitamin D levels in prediabetic, controlled DM, and uncontrolled DM

level in prediabetic, controlled DM, and uncontrolled DM group is shown in Figure 1.

Spearman test showed there was no correlation between HbA1c and vitamin D (p=0.739) as shown in Table 4. This results revealed that there was no connection between HbA1c level variation and vitamin D level variation in subjects of type 2 DM.

Table 4. Correlation between HbA1c and vitamin D levels (n=93)

Variable	Vitamin D	
	r	*p
HbA1c	0.035	0.739

: *Spearman's test

Data in Table 4 showed that subject presentation with vitamin D deficiency was found higher in uncontrolled DM (24,2%) and lower in controlled DM (16,7%) but the statistic test result with Chi-Square test showed that the difference was not significant between vitamin D levels and DM.

As it is known, vitamin D is a fat steroid consisting of two forms of vitamin D2 (ergocalciferol) and vitamin D3 (cholecalciferol). Both forms of these vitamins are in a state of inactivity and must undergo hydroxylation process as much as two times in the liver and kidney. The liver hydroxylates vitamin D2 or D3 to 25 (OH) D3 or cholecalciferol, a compound which is stable in the blood. Then 25 (OH) D3 will be hydroxylated for the second time in the kidney and converted to 1.25 (OH) 2D3 or calcitriol. the active compound functioning to increase the absorption of calcium and phosphate from the gut for osteoclast formation in bone metabolism.[7,8,11]

The association of vitamin D with type 2 DM is as follows: there is central insulin resistance in pancreatic beta cells triggers gluconeogenesis in pancreatic alpha cells so that glucose production in the basal state by the liver increases and glucose use in peripheral tissues is impaired. This process affects the peripheral insulin resistance of insulin receptor receptors in muscle, brain and adipose tissues resulting in absorption decreased of glucose for energy requirement in peripheral tissue, increased lipolysis of adipose tissue, increased deficiency of the gastrointestinal and increased glucose adsorption in the kidney resulting in impaired glucose tolerance.[8,12,13]

Several studies have shown that vitamin D has an important role in the work of pancreatic beta-cells, insulin sensitivity and insulin secretion by direct and indirect mechanisms. The discovery of vitamin D receptor (VDR) and vitamin D binding protein (DBP) in beta-cells support the role of vitamin D in insulin secretion. In the direct mechanism, vitamin D works on VDR by transcriptional activation of the insulin gene that increases insulin secretion, and stimulates expression of insulin receptors and increases insulin sensitivity. In the indirect mechanism, active form vitamin D (1.25 (OH) 2D3) regulates calbindin, a calcium-binding protein found in pancreatic beta-cells. With such regulation, vitamin D will act as a depolarization modulator of the beta-cells pancreas to secrete insulin in sufficient quantities.[14-16]

Vitamin D also promotes insulin sensitivity in peripheral tissue with anulcer, stimulates expression of insulin receptors and activates peroxisome receptor proliferator activated receptor δ (PPAR-δ) involved in fatty acid metabolism and glucose transport in muscles to convert to adenosine triphosphate (ATP) as a source of energy. In the event of vitamin D deficiency, it will affect intracellular calcium in the target tissue insulin, causing peripheral insulin resistance that impacts the transport of glucose to the muscle which is reduced. Vitamin D deficiency also affects insulin resistance indirectly through the aldosterone renin-angiotensin system. Angiotensin inhibits insulin in the vasculature and muscle have impaired glucose absorption and reduced energy demand.[12,14]

Based on the Spearman correlation test it was shown that there was no significant relationship between HbA1c levels with vitamin D levels ($p > 0.05$, $r = 0.035$). Chi-Square test also showed no significant correlation between vitamin D deficiency and type 2 diabetes ($p = 0,733$), but on subject presentation, the group of controlled DM patients showed a greater than the controlled and prediabetic group of DM. The results of this study differed from those obtained by Silambanan *et al*. which showed a significant correlation between vitamin D deficiency and DM type 2 ($p = 0.000$) in which the healthy control group had a mean and standard deviation of vitamin D 51.47 + 26.21 while vitamin D deficiency group had mean and standard deviation of 16.35 + 5.26. The different results in this study may be caused by several factors such as the history of frequent exposure to sunlight, skin color of the patient, duration of patients suffering from DM, and history of taking drugs, and intake of food or beverages that affect vitamin D levels.[14,15]

The high frequency of sun exposure to individuals living in the tropics such as Indonesia can affect most of the vitamin D metabolism in the body of the target organ, one of which is the pancreatic beta-cell. Individuals who are often exposed to sunlight will affect the path of vitamin D metabolism to pancreatic beta-cells directly by activating 7-dehydrocholesterol found in the skin to vitamin D3. Vitamin D3 will undergo a process of hydroxylation in two stages of the liver and kidney to 1.25 (OH) 2D3 which then directly activate the pancreatic beta-cells with the intermediate down regulation Fas-related pathways (Fas/Fas-L) so that the beta-cells produce insulin in sufficient quantities. The mechanism explains why in this study, controlled and controlled DM patients are more likely to have adequate vitamin D levels than those with vitamin D deficiency.[12,15,16]

The results of this study were very different when compared with other studies conducted in the

United States and some countries in continental Europe those continents have a climate of four seasons and residents who live there are rarely exposed to sunlight which affects the metabolism of vitamin D in the body. This reason explains why the people who live there are more likely to suffer from vitamin D deficiency and therefore have a high risk of having type 2 diabetes.[9,12,17]

CONCLUSIONS AND SUGGESTIONS

Based on the study, it can be concluded that vitamin D levels are not related with type 2 diabetes mellitus, controlled DM, and uncontrolled DM, and variation of HbA1c level is not influenced by variation of vitamin D level. The researchers suggest, that it is better to analyze the characteristic pattern of differences between vitamin D levels with severity of DM (glucose level or HbA1c) and analyze another risk factors involved in vitamin D metabolism blood sugar control for type 2 DM.

REFERENCES

1. Afzal S, Bojesen SE, Nordestgaard BG. Low 25(OH)D and risk of type 2 diabetes mellitus: A prospective cohort study and meta-analysis. Clin Chem, 2013; 59(2): 381-91.
2. American Diabetes Association. Standard of medical care in diabetes. Diabetes Care, 2013; 36 (1): s11-s50.
3. Carvalho EF, Griz LHM, Bandeira F, Gabbay MA, Dib SA. Vitamin D and diabetes mellitus: An update 2013. Arq Bras Endocrinology Metabolic. 2014; 58(1): 1-8.
4. Chiu CK, Chu A, Saad FM. Hypovitaminosis D is associated with insulin resistance and cell dysfunction, Am J Clin Nutr, 2004; 79(5): 820-5.
5. Corwin EJ. The pancreas and diabetes mellitus in Handbook of Pathophysiology, 3rd Ed., Philadelphia, Lippincott Williams and Wilkins, 2008; 550-573.
6. Tsur A, Feldman BS. Decreased serum concentrations of 25-hydroxycalciferol are associated with increased risk of progression to impaired fasting glucose and diabetes. Diabetes Care, 2013; 36(5): 1361-67.
7. Deleskog A, Hilding A, Birismar K, Hamsten A, Efendic S, Ostenson CG. Low serum 25(OH)D predicts progression to type 2 diabetes in individuals with prediabetes but not with normal glucose tolerance. Diabetologia, 2012; 55(6): 1668-78.
8. Eliades M, Pittas AG. Vitamin D and type 2 diabetes. Clinic Rev Bone Miner Metabolic. 2009; 7(2): 185-198.
9. Epstein S. Vitamin D: Metabolism. Endocrinology and Metabolism Clinics in North America, 2010; 39(2): 243-446.
10. Pittas AG, Joseph L, Hughes DB. Review: The role of vitamin D and calcium in type 2 diabetes. A Systematic review and meta-analysis. JCEM, 2007; 92(6): 2017-29.
11. Griz LHM, Bandeira F, Andrade M, Gabbay L, Dib SA, de Carvalho EF. Vitamin D and diabetes mellitus: An update-2013. Arq Bras Endocrinol Metab, 2014; 58(1): 1-14.
12. Harinarayan CV. Vitamin D and diabetes mellitus. Hormones, 2014; 13(2): 163-81.
13. Hesmat R, Malazy OT. Effect of vitamin D on insulin resistance and anthropometric parameters in type 2 diabetes; A randomized double-blind clinical trial. Journal of Pharmaceutical Sciences, 2012; 20(1): 10.
14. Silambanan S, Jothimalar LG. Vitamin D status in type 2 diabetes mellitus. International Journal of Clinical Biochemistry and Research. 2015; 2(3): 140-142.
15. Lui J. Vitamin D and diabetes mellitus: What do we know?. Journal of Hypo and Hyperglycemia. 2013; 1: 1.
16. Masharani U. Diabetes mellitus and hypoglycemia in current medical diagnosis & treatment. 46th Ed., Toronto, Mc-Graw Hill Medical. 2007; 1219-21.
17. Mathieu C, Gysemans C, Guiletti A. Vitamin D, and diabetes. Diabetologia, 2005; 48(7): 1247-57.