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Impact of Vitamin D Deficiency on Diabetic Obese Patients with Non-Alcoholic Fatty Liver Disease

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Abstract

Background: Vitamin D is considered an extremely important physiological regulator other than its classical role in skeletal homeostasis. There is growing evidence that connects vitamin D to liver disease.

Aim of Study: Aim of this work is to study the clinical utility of vitamin D among the diabetic obese patients and to assess the effect of vitamin D deficiency on NAFLD.

Material and Methods: Our study was conducted on 150 older diabetic obese adult patients, patients were divided into two groups, group with normal vitamin level (NVDD) and group with decreased vitamin level (VDD). Both groups included a percent of NAFLD patients.

Results: Highly significantly increased percentage of NAFLD in the group with vitamin D deficiency (VDD). Also, the prevalence of hyperlipidemia was higher in patients with vitamin D deficiency as compared to patients with a normal vitamin D.

Conclusion: Vitamin D deficiency considered to be an important risk factor for development of NAFLD.

Key Words: *Fibrosis – NAFLD – Vitamin D – VDD – NVDD.*

Introduction

NON-ALCOHOLIC fatty liver disease (NAFLD) is now considered to be the most common chronic liver disease worldwide with significant consequences that markedly affect health and economic systems of Western countries *[1,2]*.

NAFLD is defined by excessive fat deposition in hepatocytes in individuls who do not drink excessive amounts of alcohol. It include a range of diseases of the liver, from non-alcoholic steatohepatitis (NASH), which can cause cirrhosis and hepatocellular carcinoma, to simple hepatic steatosis *[3]*. Estimates indicate that NAFLD affects about 25% of the world's population, and it has gradual rising prevalence nowadays *[4]*.

The pathogenesis of NAFLD is primarily linked to multiple metabolic impairments and alteration of the glucose–insulin homeostasis. For this tight connection between metabolic diseases and NAFLD, this condition has been recently re-named as metabolic (dysfunction)-associated fatty liver disease (MAFLD) *[5]*. As a vicious circle, once MAFLD is established it increases the hepatic insulin resistance, which, in turn, can be triggered in about 30–40% of cases, MAFLD evolution towards steato-hepatitis (NASH), and eventually, cirrhosis, liver failure, and hepatocarcinoma *[6]*.

MAFLD also promotes systemic low-grade inflammation with impaired insulin sensitivity in extra-hepatic tissues. Finally, MAFLD increases the risk of type 2 diabetes with risk of increase its complications and is considered as an established risk factor for cardiovascular morbidity and mortality *[7]*.

Vitamin D is one of the fat-soluble hormones which is obtained from sunlight exposure, diet, and also from some health supplements. Dietary sources include oily fish, shitake mushrooms and fortified foods which can include cereals and milk, and supplements *[8]*. Sunlight may be responsible for about 90% of the requirement of vitamin D in most people *[9]*.

In addition to its essential role in bone and calcium homeostasis, there is an expanding volume of data regarding associations between vitamin D deficiency and several medical diseases including multiple sclerosis, myopathy, Alzheimer's disease, and cancer. Liver disease discovered to be strongly linked to vitamin D deficiency, and the development

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of the vitamin D receptor (VDR) knockout murine model has greatly increased our understanding of vitamin D's role in liver disease [10].

Aim of the work:

To investigate the impact of vitamin D deficiency on the variable studied parameter among the diabetic obese patients, and to assess the effect of vitamin D deficiency on NAFLD.

Material and Methods

This cross-sectional study was conducted on 150 adult diabetic obese patients from outpatient clinic of Ain Shams University, they were divided into two groups, according to the level of vitamin D, group with normal vitamin level (NVDD. This group included 55 patients and group with decreased vitamin level (VDD). This group included 95 patients.

We included also NAFLD patients within both groups (NVDD and VDD) in order to evaluate the association between vitamin D and NAFLD. NAFLD patients was diagnosed based on to ultrasound findings.

All recruited patients were obese with a BMI $\geq 30 kg/m^2$. Diabetes were diagnosed according to guidelines. From all patients, we obtained the followings; full medical history, clinical examination, and laboratory investigations including liver function tests, lipid profile and CBC. Vitamin D was assayed using electrochemiluminescence (ECL) on Cobas e 411 (Roche Diagnostics).

Statistical analysis:

Data were analyzed using Statistical Program for Social Science (SPSS) version 24. Qualitative data were expressed as frequency and percentage. Quantitative data were expressed as median and IQR in a case of data was not normally distributed. Also, the following tests were done; Mann Whitney U test (MW) to compare between two groups (for abnormally distributed data), Chi-square test was used when comparing between non-parametric data, and pearson's correlation coefficient (r) test was used for correlating data p-value, p-value <0.001 was considered as highly significant and p-value >0.05 was considered insignificant.

Results

Our results included 55 patients with NVDD (36.7%) and 95 patients with VDD (3.3%). A statistically significant difference was found between NVDD and VDD groups as regards serum cholesterol, LDL-cholesterol, and hemoglobin (p-value <0.05) as in Table (1).

A statistically significant difference was found between NVDD and VDD groups as regards US findings (p-value <0.05). In NVDD group, there were 35 normal patients (63.6%), 11 patients (20%) with bright fatty liver normal size and 9 patients (16.4%) with enlarged fatty liver. In VDD group, there were 36 normal patients (37.9%), 31 patients (32.6%) with bright fatty liver normal size, 26 patients (27.4%) with enlarged fatty liver and 2 patients (2.1%) with irregular coarse liver. Furthermore, 63.2% of patients with VDD was having NAFLD as in Table (2).

In NAFLD group, astatistically significant negative correlation between the levels of vitamin D and each of the followings; serum creatinine, WBCs, and hemoglobin (p-value <0.05). On the other hand a statistically significant positive correlation was found between vitamin D and platelets (p-value <0.05). No statistically significant correlation was found between vitamin D and other studied data. In NAFLD group, vitamin D level was statistically significant negative correlated with both ALT and LDL-cholesterol.

The multivariate logistic regression analysis for factors predictive of vitamin D status in all studied patients showed that sex, serum cholesterol, LDL-cholesterol, hemoglobin and presence of NAFLD could be used as predictive factors for vitamin D status in the studied patients (p-value <0.05).

Discussion

NAFLD is not simply an outcome of insulin resistance and metabolic derangements; instead, it is a disease with complicated underlying pathogenesis. Moreover, deficiency of vitamin D has been associated with NAFLD development and increased susceptibility to more severe liver diseases. Derangement in vitamins correlates to the lipotoxic hepatic environment, immune system alteration, oxidative stress, unwarranted inflammation, gene mutations, epigenetic modification, and gut dysbiosis seen in NAFLD [11].

Previous studies have shown conflicting results regarding the association between hypovitaminosis D and NAFLD. The aim of this study is to evaluate the effect of hypovitaminosis D and metabolic syndrome on NAFLD. Deficiency of vitamin D inadequacy is often observed in obese individuals, which can be causes by vitamin D sequestration in adipose tissue, resulting in decreased availability, and diluted volumetric due to larger adipose tissue mass [12].

In the present study we performed a cross-sectional study on 150 older diabetic obese adult patients.

Our comparative study between the 2 groups (NVDD & VDD groups), revealed that there was a highly statistically significant difference as regards sex. In NVDD group, there were 10 males (18.2%) and 45 females (81.8%). In VDD group, there were 42 males (44.2%) and 53 females (55.8%).

In a similar study, they reported a higher risk of vitamin D deficiency in women with metabolic syndrome [13], same findings were found in another study which stated that vitamin D deficiency is more severe in women in a cross-sectional study which conducted to assess vitamin D deficiency and the associated risk factors in 166 women aged 30–65 years and they recommend to raise public awareness among women about the benefit of rich dietary sources of vitamin D, and the sun exposure [14].

Moreover, our study reported a statistically significant higher percentage of NAFLD in VDD group (60 patients, 63.2%) as compared to NVDD group (20 patients, 36.4%). The same findings were found in many previous studies [15-17]. Another convincing experimental data showed that the vitamin D/VDR axis is directly involved in the modulation of many metabolic and inflammatory pathways associated with the development of NAFLD in overweight and obesity [18].

This also was confirmed by Tingwan Du et al., who documented that in the early stage of fatty liver, low serum 25(OH)D3 concentration was associated with an increased risk of NAFLD. On the other hand the adequate vitamin D state (25(OH) D3 >20ng/mL) has a significant protective effect regarding the NAFLD [19].

Li et al. [20] demonstrated that treatment with 100 nM calcitriol protects against hepatic steatosis by inducing autophagy. The differences in these studies may be due to focusing on many different pathways. The benefits of vitamin D supplementation were described in a previous meta-analysis study based on a large population which reports an inverse correlation between serum 25(OH)D3 and NAFLD in European individuals [21].

In another pervious study, the presence of a low vitamin D level was a risk factor for NAFLD/steatosis with no advanced fibrosis (FIB-4 <1.3)[19] this is consistent with our study as our results showed a statistically significantly low levels of vitamin D among NAFLD patients however in our study vitamin D did not show any significant correlation with the grade of liver fibrosis (FIB-4 score).This could be explained by a fewer number of patients recruited in our study (as previous study minimally enrolled 350 patients versus 150 patients in our study) [19]. However, Xiu et al. [21] reported that levels of vitamin D decrease with the progression of hepatic fibrosis. These differences may be attributed to several reasons such as different sources of patients, presence of the early stage of fatty liver disease without liver fibrosis [19] whereas in the study performed by Xiu et al., they enrolled the majority of patients having liver fibrosis [22].

Nonetheless, further studies on larger populations of individuals may be needed before making general conclusions on the importance and benefit of vitamin D supplementation in patients with fatty liver disease.

Our study showed a statistically significant negative correlation between vitamin D and ALT, this finding was in agreement to Geier et al., who observed that 48-week vitamin D3 treatment (2100 IU vitamin D3 daily) leads to significantly decreased serum ALT in twenty individuals with biopsy-proven NASH [23].

Similarly, another study noticed modest improvement of ALT and enhanced liver fibrosis score, along with a trend towards reduced insulin-resistance-estimated by the Homeostatic Model Assessment for Insulin Resistance (HOMA-IR) index in the actively vitamin D treated patients group versus placebo [24].

Also, many previous supported the hypothesis that vitamin D supplementation may exerts beneficial effects mostly in younger individuals, with shorter disease duration and mild to moderate liver damage [25] and/or in addition to anti-fibrotic agents [26]. This also is consistent with a previous meta-analysis, which reported that vitamin D supplementation decreases serum ALT [27]. This was further confirmedby another study who demonstrated that 25(OH)D3 was negatively correlated with ALT [19].

Our study showeda statistically significant difference between studied groups (NVDD & VDD groups) as regards the serum cholesterol, LDL-cholesterol in VDD groups as compared to NVDD.

Many previous studies evaluated the effect Vitamin D on the lipid profile and revealed that vitamin D had a significant effect on the lipid profiles [28,29] In contrast, Barchetta et al., which reported that vitamin D had a non-significant on the lipid profile [30].

Another study indicated that vitamin D supplementation (alone or with co-supplementation) could statistically improve lipid metabolism and concluded that supplying vitamin D could improve the serum lipid profile, and supplementation of vitamin D ≤5,000 IU/day seemed to have a statistical effect compared with higher doses and this study recommended larger prospective studies to further validate their results [31].

Our study showed a statistically significantly positive between vitamin D and platletes, this was not consistent with a previous study which included 78 participants, reporting a significant negative relationship between vitamin D level and platelet count among CAD patients [32].

In another study, there was a negative relationship between the vitamin D levels and the platelet count in gestational diabetes mellitus [33], the same

results observed also by Yon Chul Park et al.، who enrolled 3190 adult subjects, as the platelet count and mean platelet volume were found to be inversely associated with vitamin D levels in adults *[34]*, while Cumhur et al. *[35]* reported a non-significant relationship between vitamin D deficiency and platelet count on 434 healthy participants. Very few studies explored the association between vitamin D level and platelet count, further studies are needed to clarify the mechanisms by which vitamin D level can affects platelet count.

Another study suggested that the effect of oxidative stress may be related to the high platelet counts *[36]*. The elevated Interleukin-6 (IL-6) level, as an inflammatory cytokine, results in increased oxidative stress and increase megakaryocyte production and maturation *[37]*. There was an inverse correlation between vitamin D and IL-6 levels in a study on 1381 healthy participants *[38]*. Lower vitamin D levels were thought to be related with inflammation and elevated cytokine levels that would result in increased thrombocyte count *[39]*.

Our study had certain limitations such as using liver enzymes and abdominal sonar as markers for NAFLD diagnosis which is controversial, and there are no other noninvasive methods to use in trials but we use FIB-4 score to differentiate fibrosis index in NAFLD patient and exclude presence of any fibrosis in normal control patients.

Conclusion:

Vitamin D deficiency considered to be an important risk factor for development of NAFLD. Also, the prevalence of hyperlipidemia was higher in patients with vitamin D deficiency as compared to patients with a normal vitamin D.

The Ethical Committee of Ain Shams University approved the protocol of this study (approval number FMASU R242/2023).

Conflict of interest: None.

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تاثير نقص فيتامين د على مرضى السكر المصابين بالسمنه وتشحم الكبد الغير كحولى

يعتبر مرض تشحم الكبد من اكثر امراض الكبد انتشاراً وتاثيرا على الصحه العامه والذى قد يؤدى إلى التهاب الكبد الدهنى والذى قد ينتهى بتليف الكبد وسرطان الكبد. ومع وجود مرض الكبد الدهنى يزداد نسبه الاصابه بمقاومه الانسولين وداء البول السكرى.

يعتبر فيتامين د من الفيتامينات المهمه والتى تلعب دور مهم فى حدوث تشحم الكبد بالاضافه إلى دوره وتاثيره المهم فى أمراض العظام.

الهدف من البحث: دراسه العلاقه بين فيتامين د ومرض تشحم الكبد فى مرضى داء البول السكرى المصابين بالسمنه.

طريقه البحث: تم عمل الدراسه على ١٥٠ مريض مصابين بالسمنه وداء البول السكرى وتم قياس فيتامين د لكل المرضى ويشمل المرضى مرضى مصابين بتشحم الكبد ومرضى طبيعيين.

نتيجه البحث: يوجد زياده في عدد الحالات المصابه بتشحم الكبد فى مرضي نقص فيتامين د وكذلك يوجد زياده فى عدد الحالات المصابه بارتفاع نسبه الكوليسترول فى الدم فى المرضى المصابين بنقص فيتامين د واستنتج البحث ان نقص فيتامين د يعتبر من عوامل الخطوره للاصابه بمرض تشحم الكبد الغير كحولى مع زياده نسبه الاصابه بارتفاع نسبه الكوليسترول فى الدم فى حاله الاصابه بنقص فيتامين د.