VITAMIN D AS A PREVENTION FOR DIABETES

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Abstract: Vitamin D deficiency predisposes individuals to type 1 and type 2 diabetes and has been shown to impair insulin secretion and insulin sensitivity in humans and in animal models of diabetes, suggesting a role in the development of both types of diabetes. Furthermore, epidemiological studies suggest a link between vitamin D deficiency in early life and the later onset of type 1 diabetes. In some populations, type 1 diabetes is associated with certain polymorphisms within the vitamin D receptor gene. Vitamin D deficiency may, therefore, be involved in the pathogenesis of both forms of diabetes, and a better understanding of the mechanisms involved could lead to the development of preventive strategies. The aim of this study is to systematize and highlight the links between vitamin D and the development of diabetic diseases.

Keywords: Vitamin D, 1α,25-dihydroxyVitamin D_3, Diabetes, insulin resistance, calcium, plasma glucose,

Introduction

Vitamin D is also known as the "sunny vitamin". It belongs to a group of fat-soluble vitamins, but in nature it is extremely rare. The active form of vitamin D exists mostly in animal products, including fish liver oils, liver, animal fats, butter and egg yolk. It is important to note that Vitamin D is not a simple vitamin, but a steroid hormone, which has the ability to affect about 2000 genes in the human body [38]. Nowadays, vitamin D is known for its many benefits. It regulates the immune system, plays an important role in building and maintaining healthy cells, it is essential for absorption of calcium from the body, and also promotes bone growth [2, 31].

The general way to obtain Vitamin D is through UVB exposure. Recommended Dietary Allowances (RDAs) of Vitamin D for ages 1–70 years is 600 IU/d and for ages above 71 - it is 800 IU/d. This corresponds to a serum 25-Hydroxyvitamin D level of about 20 ng/ml (50 nmol/liter) [39].

The main symptoms of Vitamin D deficiency include skeletal pain and lack of muscle strength. In some people, however, the symptoms may be more subtle and difficult to distinguish. Nevertheless, even without tangible signs, small amounts of Vitamin D in the body can contribute to the development or increased incidence of cardiovascular diseases, diabetes type 1 and 2, cancer, multiple sclerosis, cognitive impairment in the elderly, severe asthma in children and high blood pressure [46].

Vitamin D deficiency predisposes individuals to type 1 and type 2 diabetes and receptors for its activated form, 1α,25-dihydroxy vitamin D_3 ($1,25(OH)_2D_3$), have been

identified in both β-cells and immune cells [8, 20, 28, 36]. Vitamin D deficiency has been shown to impair insulin synthesis and secretion in humans and in animal models of diabetes, suggesting a role in the development of type 2 diabetes [26]. Furthermore, epidemiological studies suggest a link between vitamin D deficiency in early life and the later onset of type 1 diabetes [51]. In some populations, type 1 diabetes is associated with certain polymorphisms within the Vitamin D receptor gene [36]. Vitamin D deficiency may, therefore, be involved in the pathogenesis of both forms of diabetes, and a better understanding of the mechanisms involved could lead to the development of preventive strategies.

Insulin secretion

Insulin secretion from β-cells of pancreatic islets is subjected to very precise and complex regulation by metabolites, hormones and the autonomic nervous system. In recent years it is shown that vitamin D also has an impact on this regulatory process. Kostoglou-Athanassiou *et al*. in 2016, showed the effect of Vitamin D on the proliferation of rat pancreatic β-cells in vitro. Insulin-1 (INS-1) rat pancreatic β-cells were incubated with progressively decreasing concentrations of $1,25(OH)_2D_3$, at an initial concentration of 100 nM to assess the proliferation of INS-1 rat β-cells. They found that $1,25(OH)_2D_3$ reduced the proliferation of INS-1 rat β-cells in vitro and thus concluded that vitamin D was found to modulate the proliferation of rat pancreatic β-cells in vitro [21].

Another study aimed to determine whether combined vitamin D and calcium supplementation improved insulin sensitivity, insulin secretion, β-cell function, inflammation and metabolic markers. Their trial included 95 adults with serum $1,25(OH)_2D_3$ ≤55 nmol/L at risk of T2DM (with AUSDRISK score ≥15). They provided the patients with daily calcium carbonate (1,200 mg) and cholecalciferol [2,000–6,000 IU to target $1,25(OH)_2D_3$ >75 nmol/L] or matching placebos for 6 months. Mean serum $1,25(OH)_2D_3$ concentration increased from 48 to 95 nmol/L in the treatment group, but remained unchanged in controls. They found no significant changes in insulin sensitivity, insulin secretion and β-cell function, or in inflammatory and metabolic markers between or within the groups. However, in a post hoc analysis restricted to participants with prediabetes, a significant beneficial effect of vitamin D and calcium supplementation on insulin sensitivity was observed [13].

Oosterwerff *et al.* studied the effect of vitamin D supplementation on insulin sensitivity and β cell function. Their study included 130 non-Western immigrants with prediabetes and vitamin D deficiency that were randomly given either cholecalciferol (1200 IU/d) or a placebo for 16 weeks. They found that mean serum $1,25(OH)_2D_3$ concentrations increased significantly in the vitamin D groups compared with placebo groups. There was no significant effect on insulin sensitivity and β-cell function. However, in a post hoc analysis, when patients with diabetes at baseline were excluded, a significant increase in the insulinogenic index was observed in participants who obtained a $1,25(OH)_2D_3$ concentration ≥60 nmol/L [35].

There was a need for the consideration of the PTH/$1,25(OH)_2D_3$ axis when studying the associations between vitamin D and glucose homeostasis. Kramer *et al.* studied the associations of $1,25(OH)_2D_3$ and parathyroid hormone at 3 months postpartum with β-cell function and glycemia at 12 months postpartum in 494 subjects. They found that 32% of subjects with prediabetes/diabetes mellitus at 12 months postpartum had both vitamin D deficiency and PTH in the highest tertile at 3 months postpartum. They also found that vitamin D deficiency with PTH in the highest tertile at 3 months predicted poorer β-cell function and insulin sensitivity and increased fasting and 2-h glucose at 12 months postpartum. It is important to note, however, that they found vitamin D deficiency with lower PTH did not predict the same outcomes. [22].

Vitamin D Receptor and diabetes mellitus

The effects of vitamin D are mediated

by nuclear receptors (VDR) that are distributed in many tissues and cells [31]. In 2016, Sisley *et al.* hypothesized that activation of central VDR links vitamin D to the regulation of glucose and energy homeostasis. Indeed, they found that small doses of $1,25(OH)_2D_3$ into the third-ventricle of the brain improved glucose tolerance and markedly increased hepatic insulin sensitivity, an effect that is dependent upon VDR within the paraventricular nucleus of the hypothalamus. Incidentally, they also found that chronic central administration of $1,25(OH)_2D_3$ dramatically decreased body weight by lowering food intake in obese rodents. They found that VDR co-localized with and activated key appetite regulating neurons in nucleus arcuatus, namely pro-opiomelanocortin neurons. Specifically, their data suggest that vitamin D regulates glucose homeostasis via the paraventricular nuclei and energy homeostasis via nucleus arcuatus [43].

Sentinelli *et al.* in 2016, performed an association study between rs11568820 polymorphism in the VDR gene, and type 2 diabetes mellitus (T2DM) in a cohort of Italian adults with T2DM and in non-diabetic controls. As many as 1788 adults were genotyped for the polymorphism. The AA genotype was significantly more frequent in adults with T2DM compared to controls (7.5% vs. 4.6%) and conferred a higher risk of T2DM. Rs11568820 was also associated with reduced indices of β-cell insulin secretion. They demonstrated for the first time that VDR gene AA carriers have higher risk of T2DM and impaired insulin secretion [41]. Sahin *et al.* in 2017 investigated the association between FokI, ApaI, TaqI and BsmI polymorphisms of VDR gene and type 1 diabetes (T1DM) in children. They found a total of 9 studies comprising 1053 patients and 1017 controls. Their results showed that BsmIBB, BsmIBb and TaqItt polymorphisms were associated with an increased risk of T1DM, whereas BsmIbb and TaqITT had a protective function for T1DM in children [40].

Yu *et al.* in 2015 aimed to quantify the association between polymorphisms of BsmI and FokI in the VDR gene and T2DM risk through literature review. 23 articles with 30 case–control studies were included in their study. The association between BsmI polymorphism and T2DM was weak in two genetic models (Bb vs bb and BB+Bb vs bb). On the other hand, a strong association between FokI polymorphism and T2DM indicated that this gene polymorphism could potentially be a risk factor for T2DM, especially in Chinese populations. They concluded that reliable conclusions about any links between Vitamin D receptor genetic polymorphisms and T2DM will depend on studies that have a larger sample size [27].

Miettinen *et al.* investigated whether single nucleotide polymorphisms (SNPs) associated with $1,25(OH)_2D_3$ concentration in the vitamin D metabolic pathway show different genotype distributions between families with a child diagnosed with T1DM (cases) and families with a healthy child (controls). A total of 31 SNPs in eight genes were studied in case and control mothers and family. They found that SNPs in VDR had different genotype distributions between the case and control mothers (rs1544410, rs731236, rs4516035). They proposed that maternal genotypes of SNPs in VDR may influence the in-utero environment and thus contribute to the development of T1DM in the fetus [27].

Al-Daghri *et al.* assessed the link between VDR polymorphisms and genetic susceptibility to metabolic syndrome, T2DM and vitamin D deficiency in the Saudi Arabian population. Five-hundred-seventy Saudi individuals (285 Metabolic Syndrome and 285 controls) were enrolled in this cross-sectional study. TaqI, BsmI, ApaI and FokI single nucleotide polymorphisms of the VDR gene were genotyped. The CT genotype and the dominant model CT + TT of BsmI were associated with an increased risk of diabetes. In contrast, the CT and CT + CC genotypes of FokI showed a reduced risk of diabetes. They also found that the prevalence of vitamin D deficiency was lower in subjects with the AC genotype of ApaI. They concluded that components of Metabolic Syndrome for example obesity, low HDL and T2DM were

associated with the VDR gene [1].

Another study investigated the role of TaqI, BsmI, ApaI and FokI VDR polymorphisms in T1DM found a non-significant association of TaqI polymorphism with T1DM in the recessive model and BsmI polymorphism was not associated with T1DM in the dominant model. Furthermore, ApaI polymorphism was not associated with T1DM risk in the homozygous model and the FokI polymorphism was not associated with T1DM risk in the dominant model. Meta-analysis on haplotypes revealed that BAT was a significant protective factor for T1DM. The conclusion was made that while individual VDR polymorphisms seemed not to be associated with T1DM risk, haplotypes contributed significantly to disease susceptibility [47].

Frederiksen *et al.* studied the association between variants in VDR gene and protein tyrosine phosphatase, non-receptor type 2 gene *(PTPN2).* They also investigated whether there was an interaction between *VDR* and *PTPN2* and the risk of islet autoimmunity (IA) and progression to T1DM. They examined data from the Diabetes Autoimmunity Study in the Young (DAISY). Development of T1DM in IA positive children was associated with the *VDR* rs2228570 GG genotype and there was an interaction between *VDR* rs1544410 and *PTPN2* rs1893217. On the contrary, children with the *PTPN2* rs1893217 AA genotype, the *VDR* rs1544410 AA/AG genotype was associated with a decreased risk of T1DM. They concluded that the interaction between VDR and PTPN2 polymorphisms in the risk of T1DM may provide some information regarding the role of vitamin D in the etiology of T1DM [12].

Type 1 diabetes mellitus

Many investigations support hypothesis that low vitamin D may increase the chances for developing diabetes. Franchi *et al.* studied the vitamin D levels in children at the onset of T1DM compared with children with other diseases. They also tested the correlation between low vitamin D and the development of diabetes. They compared 58 children with T1DM and 166 children hospitalised for other diseases and found that the median $1,25(OH)_2D_3$ was significantly lower in the diabetic children (36.2 nmol/l) than in the other children (48.7 nmol/l). Interestingly, they found that low $1,25(OH)_2D_3$ levels showed an increase in the odds for developing T1DM. They also detected median $1,25(OH)_2D_3$ level was significantly lower in patients admitted with diabetic ketoacidosis (30.2 nmol/l) than in patients without ketoacidosis (40.7 nmol/l) [11].An association was found between low UVB irradiance, the primary source of circulating Vitamin D, and high incidence rates of type 1 childhood diabetes after controlling for per capita health expenditure. Incidence rates of T1DM approached zero in regions worldwide with high UVB irradiance [30].

A study by Hyppönen *et al.* researched the connection between dietary Vitamin D supplementation and decreased risk of developing T1DM in infancy. They performed a birth cohort study which included 12,055 pregnant women in northern Finland where the UVB prevalence is low. Data was collected in the first year of the newborns life about frequency and dose of Vitamin D supplementation, as well as any presence of suspected Rickets. By the end of the first year, the primary outcome measure was diagnosis of T1DM. Of the 10 366 children who were analyzed – followed up at age 1, only 81 were diagnosed with diabetes during the study [17].

Vitamin D levels are lower in children with multiple islet autoantibodies and in children with T1DM than in autoantibody-negative children. Raab *et al.* aimed to assess the prevalence of vitamin D deficiency in prediabetes and investigate whether or not progression to T1DM is faster in children with vitamin D deficiency and multiple islet autoantibodies. However, vitamin D deficiency was not associated with faster progression to T1DM in children with multiple islet autoantibodies [37].

In sub-tropical region with abundant sunlight children with T1DM have lower

serum vitamin D than children without T1DM. Greer *et al.* conducted a case–control study with 56 children with T1DM and 46 controls. They found that serum $1,25(OH)_2D_3$ was significantly lower in children with T1DM (mean 78.7 nmol/L) than in controls (mean 91.4 nmol/L) [16].

A case-control study among US active-duty military personnel with serum in the US Department of Defense Serum Repository identified 310 T1DM cases diagnosed between 1997 and 2009 and 613 controls. They found that in non-Hispanic whites, those with average $1,25(OH)_2D_3$ levels of ≥100 nmol/L had a 44% lower risk of developing T1DM than those with average $1,25(OH)_2D_3$ levels <75 nmol/L. However, there was no association between $1,25(OH)_2D_3$ levels and risk of T1DM among non-Hispanic blacks or Hispanics [32].

Type 2 diabetes mellitus and Insulin resistance

Many authors established the impact of Vitamin D deficiency on insulin resistance and abnormal glucose homeostasis in obesity. Borges *et al.* found that Vitamin D deficiency in the diet-induced obese mice increased hyperinsulinemia, hyperleptinemia and insulin resistance and islet changes, including alpha and β-cell disarray. In the insulin signalling pathway, insulin receptor substrate 2 expression was upregulated in the C/VitD- group and downregulated in the high fat/VitD- group. They concluded that Vitamin D deficiency exacerbated the adverse structural and physiological remodelling of pancreatic islets due to obesity, contributing to abnormal glucose homeostasis [6].

Serum $1,25(OH)_2D_3$ had a statistically significant inverse correlation with insulin resistance and positive correlation with insulin sensitivity [10]. Dutta *et al.* studied 157 individuals with prediabetes. They found Vitamin-D deficiency in 115 of these individuals. Those subjects who had the lowest vitamin-D levels (<10 ng/ml) had the highest insulin resistance (HOMA2-IR: 2.04 ± 0.67) [10]. In another study conducted on 426 prepubertal children who were assessed before and after puberty-onset serum $1,25(OH)_2D_3$, adiposity (BMI and waist circumference) and IR indicators (HOMA-IR). Authors found that children with higher adiposity and suboptimal-$1,25(OH)_2D_3$ before puberty-onset had higher HOMA-IR compared with their counterparts [7]. In the investigation of the associations between serum $1,25(OH)_2D_3$ concentrations and insulin resistance from adolescence to young adulthood, serum 25(OH)D concentrations and HOMA-IR were measured at the 17 and 20 year follow-ups of the West Australian Pregnancy Cohort (Raine) Study. The authors established that serum $1,25(OH)_2D_3$ concentrations were inversely associated with insulin resistance [5].

Evidence of the importance of Vitamin D for insulin resistance in obese subjects was investigated by *Bilge et al.* Their results demonstrated a difference in HOMA IR and $1,25(OH)_2D_3$ values between obesity and control groups and negative correlation between HOMA IR and $1,25(OH)_2D_3$ [4].

Supplementation of vitamin D significantly improves insulin resistance and glycemic status. Some authors determined that the increased circulating concentrations of $1,25(OH)_2D_3$ in obese adolescents are associated with improved markers of insulin sensitivity and resistance and reduced inflammation. After 6 months participants supplemented with vitamin D_3 had increases in serum $1,25(OH)_2D_3$ concentrations, fasting insulin, HOMA-IR and leptin-to-adiponectin ratio [3]. Gao *et al.* investigated the relationship between serum $1,25(OH)_2D_3$ and insulin sensitivity and β-cell function in 395 newly diagnosed T2DM patients. Patients were divided into three groups according to tertiles of $1,25(OH)_2D_3$ concentration. They found that HOMA-IR, Matsuda ISI, and INSR were significantly different among three groups in female patients [15]. Strobel *et al.* investigated the influence of a 6-month vitamin D supplementation in patients with T2DM. During this time, patients received Vigantol oil once a week (daily dose of 1904 IU) or placebo oil. After 6 months of therapy,

some patients achieved a $1,25(OH)_2D_3$ concentration of >20 ng/ml. Their HbA1c was significantly lower at baseline and after therapy than in patients with $1,25(OH)_2D_3$ below 20ng/ml. C-Peptide, insulin, and HOMA-index did not change significantly but fasting insulin was positively correlated with $1,25(OH)_2D_3$ concentrations after 6 months of therapy in both groups [44]. Nasri *et al.* investigated the effect of adding vitamin D to the therapeutic regimen of T2DM patients compared to placebo on regulating the blood glucose and glycemic parameters. Their study included 60 T2DM patients. Patients were given weekly vitamin D supplementation (50000 units) for 12 weeks. They found that HbA1c in male interventional group was significantly less than that of control group [33].

Krul-Poel *et al.* systematically reviewed the effect of Vitamin D supplementation on glycemic control in patients with T2DM. Twenty-three RCTs (Randomized controlled trials) examining the effect of Vitamin D supplementation on glycemic control in patients with T2DM were included in this systematic review representing a total of 1797 patients with T2DM. A significant effect of Vitamin D supplementation was seen on fasting glucose in a subgroup of studies with a mean baseline HbA1c ≥ 8% (64 mmol/mol). They, therefore, found that in patients with poorly controlled diabetes, a favourable effect of Vitamin D is seen on fasting glucose [23].

The decrease in the level of vitamin D is a risk factor for onset of diabetes and correlates with the progression of T2DM. Yang *et al.* evaluated serum $1,25(OH)_2D_3$ in newly diagnosed T2DM patients and explored the associations of $1,25(OH)_2D_3$ with insulin resistance and β-cell function. Their results showed that serum $1,25(OH)_2D_3$ was much lower in patients with newly diagnosed type 2 diabetes and the prevalence of hypovitaminosis $1,25(OH)_2D_3$ was 62.9% in diabetic patients. Among the diabetic patients, patients with hypovitaminosis $1,25(OH)_2D_3$ showed higher glycosylated haemoglobin as well as lower HOMA-β and Δ I30/ΔG30 [48]. Lima-Martinez *et al.* In 2017 assessed the relationship between $1,25(OH)_2D_3$ blood concentrations in subjects with obesity and T2DM risk according to the Finnish Diabetes Risk Score (FINDRISC) modified for Latin America (LA-FINDRISC). They detected a significantly higher LA-FINDRISC in the insufficient/low $1,25(OH)_2D_3$ group compared to normal $1,25(OH)_2D_3$ levels group. They also found that LA-FINDRISC was negatively correlated with plasma $1,25(OH)_2D_3$ levels and positively correlated with the HOMA-IR index [25]. Further to this, Nikooyeh *et al.* conducted a trial in subjects with T2DM to evaluate the possible effects of regular intake of calcium and/or Vitamin D fortified dairy product. They found that patients assigned to drink Vitamin D–fortified yogurt drink and Vitamin D + calcium–fortified yogurt drink had improved Vitamin D status accompanied by a significant increase in serum $1,25(OH)_2D_3$ concentrations. There was also a significant decrease in serum glucose, insulin, insulin resistance, and glycated haemoglobin in these patients compared to patients who were given plain yoghurt drinks [34]. Mitri *et al.* performed a systematic review of longitudinal observational studies of Vitamin D status and trials of Vitamin D supplementation on glycemic outcomes. They found that intake of >511 international units (IU)/day of Vitamin D was associated with a 27% lower risk of developing T2DM compared with an intake of <159 IU/day. Women who reported consumption of >800 IU/day of Vitamin D had a 23% lower risk of developing incident T2DM compared with women who reported consumption of <200 IU/da and men and women who self-reported higher consumption of both Vitamin D (>400 IU/day) and calcium (~600 to 800 mg/day) had ~40% lower risk of T2DM [29]. Talaei *et al.* added Vitamin D3 to the medication of patients with T2DM for eight weeks. Their results showed that Vitamin D supplementation significantly decreased serum fasting plasma glucose, insulin and insulin resistance in these patients [45].

The findings obtained in some experimental studies strongly imply that increasing

Vitamin D and Ca intakes may contribute to the prevention of obesity, T2DM, and bone disorders associated with these diseases [42]. Authors examined the effects of increased intake of Vitamin D and Ca in a mouse model of high fat diet-induced obesity on development of adiposity and diabetes. Incidentally, mice fed a high fat diet with high Vitamin D_3 and increased Ca content demonstrated a decreased weight of white adipose tissue depots and improved blood markers related to adiposity, T2DM, and Vitamin D status. Fasting plasma glucose and insulin concentrations in these mice were significantly decreased, approaching levels found in normal-weight, non-obese control. Increasing Vitamin D and Ca intakes in obesity also activates Ca^{2+}-mediated apoptotic pathway in adipocytes resulting in a decreased mass of fat tissue and improved markers of adiposity [42].

Plasma Vitamin D has a prognostic value in predicting increased risk of cardiovascular mortality as well as in initiation and/or progression of diabetic kidney disease in T2DM patients. Joergensen *et al.* found that very low levels of plasma Vitamin D to be a strong and independent predictor of all-cause mortality in T2DM patients [9]. Another study by Kayaniyil *et al.* included 712 subjects free of diabetes as confirmed by oral glucose tolerance tests (OGTTs). They established that Serum $1,25(OH)_2D_3$ was a significant independent predictor of insulin sensitivity and β-cell function across all models [19]. Lim *et al.* investigated 1080 nondiabetic Korean subjects at high risk of diabetes development and showed that the participants with $1,25(OH)_2D_3$ deficiency had an incidence of T2DM development 3.4 times that in those with sufficient levels, even after adjustment for obesity, dynamic measure of insulin resistance and pancreatic β cell function, and other known risk factors for T2DM [24]. Furthermore, Gagnon *et al.* examined the relationship between serum $1,25(OH)_2D_3$, dietary calcium, and risk of developing T2DM. In this nationally representative cohort of Australian adults, they showed that higher serum $1,25(OH)_2D_3$ concentrations were associated with a reduced risk of developing T2DM at 5 years [14].

Dalgård *et al.* examined serum $1,25(OH)_2D_3$ concentrations and various markers of insulin resistance and glucose metabolism in a population-based sample of elderly residents from the Faroe Islands located between Norway and Iceland. They found that subjects with $1,25(OH)_2D_3$ concentration below 50 nmol/L had a doubled risk of newly diagnosed diabetes compared with those having higher serum concentrations [9]. A study by Zhang *et al.* explored the relationship between insulin resistance and serum $1,25(OH)_2D_3$ concentrations in 117 Chinese patients with T2DM. They found that there was a negative correlation between $1,25(OH)_2D_3$ and HOMA-IR ($\beta = -0.314$,) and came to the conclusion that better Vitamin D status may be protective of glucose homeostasis since $1,25(OH)_2D_3$ was negatively associated with insulin resistance in Chinese patients with T2DM [50].

Conclusions

After reviewing the literature surrounding Vitamin D and diabetes, we can arrive at the conclusion that Vitamin D deficiency may predispose individuals to type 1 and type 2 diabetes. Vitamin D deficiency therefore could be involved in the pathogenesis of diabetes and an enhanced understanding of the mechanisms involved could lead to the development of preventive strategies. Vitamin D supplementation indicates exciting prospects and potential benefits for prevention of diabetes. However, adequately powered, randomised controlled trials with long periods of follow-up, experimental studies with animal models and/or cell cultures are needed to establish causality and the best formulation, dose, duration and period of supplementation.

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