Research Article

Relation between serum levels of Vitamin D and Interleukin 6 in type 1 Diabetes in an Egyptian Population

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Abstract
Type 1 diabetes is an inflammatory disease of the pancreatic islets due interaction of activated T-cells and proinflammatory cytokines in the immune infiltrate**.** Vitamin D has vital immunomodulatory properties that help in prevention of occurrence of T1DM. IL- 6 is a multifunctional cytokine which has role in the pathogeneses of type 1 diabetes. This study was conducted to evaluate levels of vitamin D and IL6 in type 1 diabetic children and their relation to RBS, glycemic control and c-peptide level.
keywords: Type I diabetes, vitamin D, Il-6 , immunomodulatory process

Introduction
Diabetes mellitus is mainly an error of carbohydrate metabolism associated with hyperglycemia (and disturbance of protein and fat metabolism(1). Type I diabetes (T1DM) defined as a complete or near-complete insulin deficiency due to an immune-mediated selective destruction of the insulin producing-cells in the islets of Langerhans. Type 1 diabetes can be considered an inflammatory disease of the pancreatic islets in which a process of programmed cell death (apoptosis) is elicited in the-cells by interaction of activated T-cells and proinflammatory cytokines in the immune infiltrate (2)

Vitamin D has vital immunomodulatory properties that help in prevention of occurrence of T1DM animal models[3]. It activates human macrophages, antigen-presenting cell maturation and inhibits dendritic cell differentiation as well as affects cytokine production by interacting with most immune cells [4, 5], one of these cytokines is interleukin 6 (IL- 6).

IL- 6, a multifunctional cytokine is secreted by T cells and macrophages to stimulate immune response during inflammation and infection. Numerous epidemiological, genetic, rodent, and human in vivo and in vitro studies have investigated the putative role of action/lack of action of IL-6 in the pathogeneses underlying obesity, insulin resistance, cell destruction, type 1 diabetes, and type 2 diabetes(6). These studies suggest both protective and pathogenetic actions of IL-6 in diabetes. In this review, we briefly evaluate vitamin D level and IL-6 level among T1DM patients and investigate the association between concentration of vit D and IL6 in relation to FBG, C-peptide, glucose control among type 1 diabetic patients.

Subjects and methods
This study was conducted on 105 children, divided in to 2 groups, control group 35 apparently healthy children selected from outpatient pediatric clinic. Another 70 children with T1DM (**according to ADA criteria 2019)** were selected from pediatric endocrinology clinic (**Maternity and Children Minia university hospital, Minia, Egypt**). Both groups were matched in age and sex. About 8 ml of venous blood was collected from each patient by sterile venipuncture under complete aseptic conditions. This sample was divided as follow: Two ml in sterile ethylene diamine tetra acetic acid (EDTA) containing tube for HbA1c and six ml into one plain tubes. Blood was left to clot in the incubator then centrifuged. The expressed serum was used for measurement of serum levels of RBG,

C-peptide, 25(OH) vitamin D and IL-6. Vitamin D was determined by detecting the level of 25(OH) vitamin D using ELFA technique (Enzyme linked fluorescent Assay) by (Mini Vidas, Biomerieux, France), IL-6 was determined by enzyme linked immunosorbent assay (EIA) (Human IL6 EIA Bioassay technology laboratory, China).

Results

Comparison between studied groups regarding laboratory data:

Laboratory data of type 1 diabetic children found that statistically significant elevation in IL-6 level among T1D patients when compared to healthy subjects. The mean ±SD of IL6 was 10.9±1.5 in control group, 165.1±111.4 in diabetic group. Regarding serum vitamin D level the study subjects were categorized into three subsets based on their detected vitamin D: Study subjects with vitamin D levels <20 ng/ml were labeled as having deficiency", subjects with vitamin D levels between 21-29 ng/ml were classified as having "vitamin D insuffi-ciency" and the ones with vitamin D levels 30-100 ng/ml were considered as having "vitamin D sufficiency". Vitamin D defiency was found in 87.1% in diabetic patients and 14.3% in control group, there was significant difference between both groups. 11.4% of diabetic patient had insufficient level of vitamin D and 14,3% of control group were suffient, only 1.4% of diabetic patients had sufficent vitamin D level while 71.4% of control group had sufficient level (table1) (figure 1),(figure 2).

Correlation between Vitamin D and different parameters in type 1 diabetic patients

There was moderate positive correlation between vitamin D level and c-peptide level, while there was strong negative correlation between vitamin D with both RBS and HbA1c (Table 2).

Correlation between IL 6 and different parameters in type 1 diabetic patients

There was strong positive correlation between IL6 with both RBS and HbA1c while there was strong negative correlation between Il6 and c-peptide level. There was moderate negative correlation between IL6 and vitamin D (Table 3)

Table (1): Comparison between studied groups regarding laboratory data:

Variable	Cases N=70	Control No=35	p value
Blood glucose			
Range (mg/dl)	200-580	70-122	0.001*
Mean± SD	323.1±79.5	92.1±14.1	
HbA_1C			
Range (%)	7.5-14.1	4-5.4	0.001*
Mean± SD	10.6±1.8	4.6±0.3	
C-peptide			
Range (ng/ml)	0.006-0.45	1.1-3	0.001*
Mean± SD	0.24±0.14	2.1±0.5	
IL6			
Range (pg/ml)	46.8-455	8-14	0.001*
Mean± SD	165.1±111.4	10.9±1.5	
Vitamin D			
Sufficient (30-100 ng/ml)	1(1.4%)	25(71.4%)	0.001*
Insuffient (21-29 ng/ml)	8(11.4%)	5(14.3%)	
Deficient (< 20ng/ml)	61(87.1%)	5(14.3%)	

Table (2): Correlation between Vitamin D and different parameters in type 1 diabetic patients

	Vitamin D	
	R	**P**
RBS	-0.75	0.001*
C-Peptide	0.56	0.001*
HbA_1c	-0.82	0.001*

Table (3): Correlation between IL-6 and different parameters in type 1 diabetic patients cases

	IL-6	
	R	**P**
RBS	0.89	0.001*
C-Peptide	-0.82	0.001*
HbA_1c	0.90	0.001*
Vitamin D	-0.71	0.001*

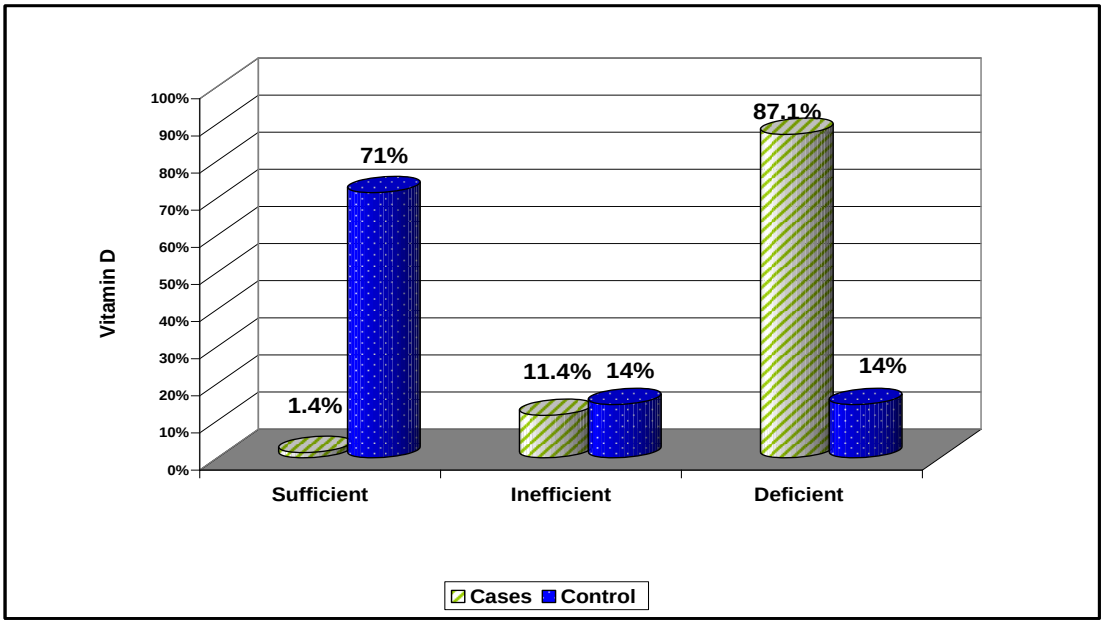

Figure (1): Vitamin D level in diabetic patients and control group

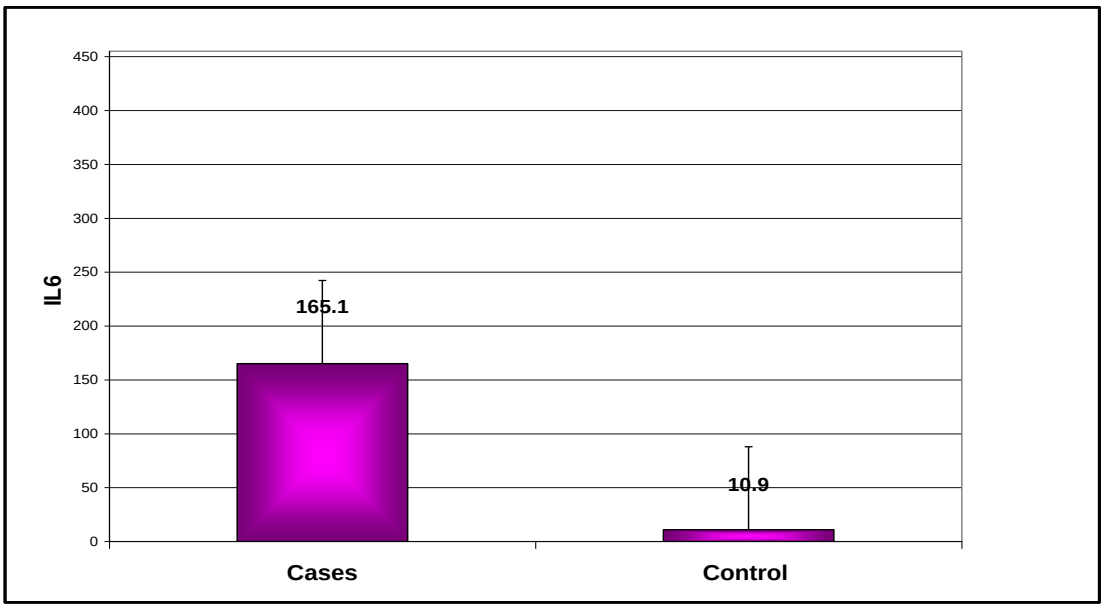

Figure (2): IL6 level in diabetic patients and control group.

Discussion

T1DM is a T-cell mediated disease that completely destroys the body's ability to synthesize and secrete insulin[8],[9] Vitamin D plays an immune-modulatory effects in T1DM prevention[9] by decreasing the proinflammatory cytokines expression involved in T1DM pathogenesis, thus making pancreatic β cells less liable to inflammation with subsequent reduction in T-cell recruitment and infiltration so suppression of the autoimmune process [10].

Serum vitamin D level in our study among T1DM revealed that 98.5% of cases have abnormally low vitamin D status (vitamin D deficiency and insufficiency). This was in line with a study done in Egypt [11] , which found that most of diabetic patients had vitamin D insufficiency. Also, another study done among Egyptian children with T1DM by Abd-Allah et al., [12] reported that 75% of T1DM exhibited vitamin D deficiency or insufficiency. Liu et al., [13] in their meta-analysis study suggested that low vitamin D level is associated with T1DM children. Also, Rasoul et al.,[14] reported significant frequency of vitamin D insufficiency and deficiency among T1DM children and concluded that serum vitamin D status is a major contributor in T1DM prevalence among Kuwaiti children. However some studies disagree with our study as a study done in Pakistan[15] that showed non-significant difference between diabetic and control group regarding vit D. Another research conducted in Chile by Garcia, et al., R (2007) found no difference in 25(OH) D level in type 1DM and healthy control[16]. Contrary to some reports[17] .The findings of the current study revealed more significant decreased vitamin D levels among those having higher RBS and HbA1c level. Additionally, C-peptide level was more significantly lower in T1DM children with lower serum level of vit. D. These findings can be explained that the vitamin D has strong role in improving β-cell function, inhibiting β-cell apoptosis and increasing β-cell replication thus influencing insulin secretion and increasing insulin sensitivity thus improving glycemic control [18]**.** Similar results were obtained in study done by Ahmed et al., (2019) who reported inverse correlation between serum 25(OH)D and HbA1c in T1DM patients with poor glycemic control among diabetic patients having vitamin D deficiency [19].

Regarding IL-6, current study showed significantly increased serum level of IL-6 among T1DM children, these results were consistent with meta-analysis done by (Chen yi et al.,) (2016) where level of IL-6 among diabetic patients where significantly higher than control subjects[20], another

study done among Egyptian T1DM children[21] revealed serum IL-6 concentrations where significantly higher in diabetic children however other studies reported no difference[22] or even decreased[23] IL-6 level in type 1 diabetic patients. Observed positive correlation between IL-6 with both RBS level and HbA1c in T1DM patients in our study agrees with several studies[24,25] this can be explained by the fact, that persistent hyperglycemia contributes to the formation of advanced glycation end products which has important role in the development of chronic inflammation. However study done among Egyptian type1 diabetic children shows no correlation between HbA1c and IL6 [21] Regarding correlation between Vit. D level and IL-6 our study revealed moderate negative correlation which may indicate an association with vitamin D deficiency and inflammatory state, represented by elevated circulating IL-6, these results agrees with study done by Shih et al., 2014[26]

Disclosure:
The authors report no conflicts of interest in this work.

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Ethical clearance:
Taken from faculty of medicine – Minia University Committee.

Conclusion:
The current study support the important role of vitamin D level in type I diabetes and its different level of deficiency with the level of RBG and HbA_1c. Also, our study show the correlation between IL-6 with both RBS level and HbA1c in T1DM patients. This suggest the role of IL-6 as inflammatory marker in T1DM.

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