Journal of Medicine in Scientific Research

Volume 5 | Issue 2 Article 4

Subject Area: Clinical and Chemical Pathology

Association between vitamin D receptor gene (BsmI-rs1544410 and FokI-rs2228570) polymorphisms and type 1 diabetes mellitus in Egyptian children

Abeer M. Mohyi
Cairo University

Dina M. El-Abd
Cairo University

Eman A. Rashed
National Institute of Diabetes and Endocrinology

Ghada A. Omar
National Institute of Diabetes and Endocrinology, ghadaomar32@yahoo.com

Mona M. A. AbdulLatif
Cairo University

Recommended Citation

Mohyi, Abeer M.; El-Abd, Dina M.; Rashed, Eman A.; Omar, Ghada A.; and A. AbdulLatif, Mona M. (2022) "Association between vitamin D receptor gene (BsmI-rs1544410 and FokI-rs2228570) polymorphisms and type 1 diabetes mellitus in Egyptian children," *Journal of Medicine in Scientific Research*: Vol. 5: Iss. 2, Article 4.
DOI: https://doi.org/10.4103/jmisr.jmisr_4_22

Association between vitamin D receptor gene (BsmI-rs1544410 and FokI-rs2228570) polymorphisms and type 1 diabetes mellitus in Egyptian children

Eman A. Rashed[a], Abeer M. Mohyi[b], Mona M.A. AbdulLatif[b], Dina M. El-Abd[b], Ghada A. Omar[a]

[a]Department of Clinical and Chemical Pathology, National Institute of Diabetes and Endocrinology and [b]Department of Clinical Pathology, Faculty of Medicine, Cairo University, Cairo, Egypt

Abstract

Introduction

Type 1 diabetes mellitus (T1DM) is an autoimmune disorder triggered by both environmental and genetic risk factors. Although some studies have reported that single-nucleotide polymorphisms of vitamin D receptor (VDR) gene play a role in T1DM susceptibility, their associations are inconclusive and remain controversial in different ethnic groups. This case–control study aimed to evaluate the association between VDR (*BsmI*-rs1544410 and *FokI*-rs2228570) single-nucleotide polymorphisms and T1DM in Egyptian children.

A total of 100 participants, 50 children with T1DM (cases) and 50 nondiabetic, age-matched and sex-matched patients (controls), were enrolled in the study in which *BsmI*-rs1544410 and *FokI*-rs2228570 variants were genotyped using TaqMan real-time PCR technology.

Results

VDR gene (*BsmI*-rs1544410 and *FokI*-rs2228570) variants are not associated with T1DM in Egyptian children.

Conclusions

VDR (*BsmI*-rs1544410 and *FokI*-rs2228570) variants did not differ significantly between T1DM cases and nondiabetic controls among Egyptian children.

Keywords: *BsmI*-rs1544410 and *FokI*-rs2228570, type 1 diabetes mellitus, vitamin D receptor gene

Introduction

A chronic pattern of autoimmune destruction of the beta cells in the pancreas, resulting in the body's inability to produce insulin, with subsequent elevation of blood glucose is defined as type 1 diabetes mellitus (T1DM). It constitutes a rough average of 5–15% of all diabetes cases all over the world [1]. For Egypt, in 1996, 2006, and 2011, in the Nile Delta region, children aged 0–18 years showed a T1DM prevalence rate of 1.9, 15.5, and 26.8/10^5/year, respectively, and an incidence rate in the same years of 0.7, 2.0, and 3.1/10^5/year, respectively [2] while in 2019, the prevalence rate among school-age children in Menoufia Governorate was of 3.75/1000 [3]. The extensive increase in the incidence and prevalence of T1DM, with its major unfavorable consequences, have forced researchers to focus on detecting the associated risk factors to allow early preclinical intervention and prevention.

The association between vitamin D deficiency and T1DM is well established. The biological actions of vitamin D are triggered by its binding to specific receptors that are found in most human tissues [4] including several insulin-responsive metabolic tissues that are concerned with glucose homeostasis, such as the liver, skeletal muscle, adipose tissue, and the

Access this article online

Quick Response Code:

Website: www.jmsr.eg.net

DOI: 10.4103/jmisr.jmisr_4_22

Correspondence to: Ghada A. Omar, MD, Department of Clinical and Chemical Pathology, National Institute of Diabetes and Endocrinology, 49 Manial Street, Cairo 11553, Egypt.
Tel: +20 122 461 0448;
E-mail: ghadaomar32@yahoo.com

Submitted: 20-Jan-2022 **Revised:** 31-Jan-2022 **Accepted:** 09-Feb-2022 **Published:** 09-Aug-2022

How to cite this article: Rashed EA, Mohyi AM, AbdulLatif MM, El-Abd DM, Omar GA. Association between vitamin D receptor gene (BsmI-rs1544410 and FokI-rs2228570) polymorphisms and type 1 diabetes mellitus in Egyptian children. J Med Sci Res 2022;5:93-7.

pancreatic islet cells [5]. These receptors, nominated vitamin D receptors (VDR), belong to the transcription receptor family and are encoded by the VDR gene on chromosome 12q13.11 [6]. Two of the common single-nucleotide polymorphisms (SNPs) of the VDR gene (*BsmI*-rs1544410 C>T resulting in a silent mutation with increased VDR mRNA stability) [7] and [*FokI*-rs2228570 G>A transcribing a shorter protein (424 amino acids) which is more active than the long-form [427 amino acids], were suggested to have variable associations with T1DM susceptibility among different ethnic-specific groups [8]. Depending on the aforementioned findings, the aim of this study was to detect the association of VDR (*BsmI*-rs1544410 and *FokI*-rs2228570) variants and T1DM in Egyptian children.

Patients and Methods

This is a case–control study of 100 Egyptian children [50 T1DM patients (cases) and 50 nondiabetic patients (controls)] who were selected from the Pediatrics Outpatient Clinic of the National Institute of Diabetes and Endocrinology, Cairo, Egypt. We obtained the approval of the Ethics Committee of the General Organization of Teaching Hospitals and Institutes, assent from participating children aged over 7 years, and informed written consent from the parents of all the recruited children. Inclusion criteria of the cases included T1DM, which was diagnosed according to the guidelines of the American Diabetes Association [9] with an age range of 6–12 years, while the exclusion criteria of controls, who were age-matched and sex-matched patients, included no family history of T1DM or any other type of autoimmune disease. Controls were recruited from children attending the Pediatrics Outpatient Clinic for general checkup. The study was performed across a span of 5 months from September 2020 to February 2021.

Sample collection and assay: venous blood samples were collected in three vacutainer tubes. The first was an EDTA tube for real-time PCR analysis. The second was another EDTA tube for the measurement of glycated hemoglobin (A1C) using high-performance liquid chromatography of the Bio-Rad D-10 A1C Testing System (Bio-Rad Laboratories Inc., Hercules, California, USA). The last one was a serum separation vacutainer tube for random blood glucose, lipid profile (total cholesterol, triglycerides, and high-density lipoprotein cholesterol), and creatinine analysis which were performed by the Architect Clinical Chemistry System (Abbott, Abbott Park, Illinois, USA). Low-density lipoprotein cholesterol concentration was calculated according to the traditional Friedewald formula [10].

DNA genotyping: DNA extraction and purification was done from 200 µl peripheral blood samples using a QIAamp DNA blood Mini kit (Qiagen, catalog no. 51104) (Qiagen GmbH, Hilden, Germany) according to the manufacturer's protocol (QIAamp DNA Mini Kit Handbook 06/2012) and applied on fully automated QIAcube platform at the facility of Kasr Aini Clinical and Chemical Pathology Department, Faculty of Medicine, Cairo University, Egypt. When the process was completed, 100 µl of extracted DNA was stored frozen at − 20°C till the time of genotyping. Real-time PCR allelic discrimination was performed on StepOne Real-Time PCR System version 2.3 Applied Biosystem (Thermo Fisher Scientific, Applied Biosystems, Waltham, Massachusetts, USA), using TaqMan SNP genotyping assays and TaqMan Universal PCR Master Mix (Applied Biosystem, Foster City, California, USA). Targeted sequences of *BsmI*-rs1544410 and *FokI*-rs2228570 were analyzed by being amplified from the extracted DNA. Using sequence-specific primers, the targeted DNA was amplified by ampliTaq Gold DNA polymerase from the TaqMan Universal PCR Master Mix II of Applied Biosystems, Part No: 4427964 (Thermo Fisher Scientific, Applied Biosystems). TaqMan MGB (minor groove binder) probes from the SNP genotyping assay distinguished between the two alleles by providing a fluorescence signal for the amplification of each allele. A measure of 2 µl of the DNA was added to 18 µl of the prepared reaction mix. The thermal cycling conditions were specified and the PCR was performed. A substantial increase in VIC-dye fluorescence only indicates homozygosis for allele 1 (wild allele); in FAM-dye fluorescence only indicates homozygosis for allele 2 (mutant allele); and in both VIC-dye and FAM-dye fluorescence indicates allele 1–allele 2 heterozygosis. After PCR amplification, an endpoint plate read was performed on 7500 real-time PCR Systems (Applied Biosystems). Sequence Detection System Software used the fluorescence measurements made during the plate read to plot fluorescence (Rn) values based on the signals from each well. The plotted fluorescence signals indicated which alleles were in each sample.

Statistical analysis

Statistical data were analyzed using SPSS, version 13 for Windows (SPSS Inc. 233 South Wacker Drive, Chicago city, IL 60606-6412, Illinois, USA). Quantitative data were reported in terms of mean ± SD or median interquartile range. Comparative statistical analysis was carried out using Student's *t* test while Mann–Whitney *U* test was used for non-normal distribution. Using χ^2 goodness-of-fit test, all frequencies in cases and controls were in accordance with frequencies predicted by the Hardy–Weinberg equilibrium with a *P* value more than 0.05. Categorical data association was tested using the χ^2 test for independence. (Monte Carlo method is used in case of computational limits and Fisher's exact test is used when sample sizes are small.) A *P* value of less than or equal to 0.05 was considered as statistically significant.

Results

This present case–control study comprised 100 patients with an age range from 6 to 12 years. They were divided into cases (50 T1DM patients) and controls (50 nondiabetic, age-matched and sex-matched patients). On comparing the demographic and laboratory data between cases and controls, a statistically significant increase is observed in cases over controls regarding random blood glucose, A1C, total cholesterol, high-density

lipoprotein cholesterol, and serum creatinine ($P < 0.001$), while there are no statistically significant differences between the two studied groups as regards age ($P = 0.691$), triglyceride ($P = 0.107$) and low-density lipoprotein cholesterol ($P = 0.230$) (Table 1).

Distribution of genotype and allele frequencies of VDR, *BsmI* (rs1544410 C>T) and *FokI* (rs2228570 G>A) variants did not differ significantly between cases and controls ($P > 0.05$). Also, they did not differ significantly in either recessive or dominant gene models of inheritance ($P > 0.05$) (Tables 2 and 3 respectively).

There is no statistically significant association of the biallelic haplotypes (*BsmI* and *FokI*) when compared between cases and controls ($P = 0.688$) (Table 4).

Table 1: Demographic and laboratory data of cases and controls

Demographic and Laboratory data	Cases ($n=50$)	Controls ($n=50$)	*P*
Age (years)	11.0 (9.0-12.0)	11.0 (8.0-12.0)	0.691‡
RBG (mg/dl)	230.68±91.52	89.88±9.33	<0.001*†
A1C (%)	9.98±1.75	5.23±0.21	<0.001*†
TC (mg/dl)	193.62±41.20	170.38±25.22	<0.001*†
TG (mg/dl)	85.5 (66.75-133.75)	88.0 (54.0-103.25)	0.107‡
HDL-C (mg/dl)	55.0 (48.0-68.75)	48.0 (45.0-53.0)	<0.001*‡
LDL-C (mg/dl)	114.62±33.36	107.58±24.28	0.230†
Creat (mg/dl)	0.62±0.09	0.54±0.13	<0.001*†

A1C, glycated hemoglobin; Creat, creatinine; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; RBG, random blood glucose; TC, total cholesterol; TG, triglycerides. †Student's *t* test for mean±SD. ‡Mann–Whitney *U* test for median (interquartile range). **P* value is statistically significant at *P* value less than or equal to 0.05.

Table 2: Distribution of *BsmI* variant genotypes and alleles in cases and controls

BsmI (1544410 C>T)	Cases ($n=50$) [*n* (%)]	Controls ($n=50$) [*n* (%)]	*P* value (χ^2)
Genotypes			
CC	12 (24.0)	16 (32.0)	0.672
CT	28 (56.0)	25 (50.0)	
TT	10 (20.0)	9 (18.0)	
Recessive gene model TT vs (CT + CC)			
TT	10 (20.0)	9 (18.0)	0.799
CT+CC	40 (80.0)	41 (82.0)	
Dominant gene model (TT + CT) vs. CC			
TT+CT	38 (76.0)	34 (68.0)	0.373
CC	12 (24.0)	16 (32.0)	
Allele			
C	52 (52.0)	57 (57.0)	0.478
T	48 (48.0)	43 (43.0)	

χ^2, χ^2 test. *P* value is statistically significant at *P* value less than or equal to 0.05.

Discussion

T1DM is one of the most common chronic, global public health problems with a progressively increasing incidence and contributes to about 5–15% of all cases of diabetes with extensive economic consequences. Nowadays, it is likely that genetics is the most prominent efficient tool detecting those at risk of developing T1DM before it is triggered by environmental factors [11]. Over the course of the past years, several studies had been investigating not only the effects of the association of each of the VDR gene variants individually, but also the outcome of their combined associations, with the development of T1DM in different ethnic populations. However, the findings of these studies were conflicting and inconclusive [8,12,13]. Therefore, we proceeded with this case–control study to detect the association between VDR (*BsmI*-rs1544410 and *FokI*-rs2228570) variants and T1DM in Egyptian children.

The findings of this current study revealed no statistically significant differences concerning the distribution of the frequencies of *VDR* gene (*BsmI*-rs1544410 and *FokI*-rs2228570) polymorphisms between cases and controls in the recruited Egyptian children, stating no significant association with T1DM. The same results were detected in the Portuguese population [14] and in the overall African and American population [8]. In disagreement, the *VDR* gene (*BsmI*-rs1544410 and *FokI*-rs2228570) polymorphisms were associated with the development of T1DM and differed significantly between patients and controls in southern European population (Greek) [15], Egyptian children [16], and the subgroup analysis of the African and American population [8].

For *BsmI*-rs1544410, agreeing with our results, there was no association between it and T1DM in Kuwaiti children [17]; but on the contrary, it was associated with T1DM in Egyptian [18] and Saudi patients [19]. All the aforementioned findings were contradicted by other studies stating that this *BsmI* polymorphism distribution was more frequent in the Brazilian nondiabetic individuals [20] and Korean controls with a suggestion that these alleles are protective against developing T1DM [21].

For *FokI*-rs2228570, several studies in different ethnic groups were in accordance with our findings as in Brazilian individuals [20] in a meta-analysis according to a subgroup analysis by ethnicity [13], Egyptian [22], Danish [23], and Pakistani populations [24]. In contrast to our study, *FokI* polymorphism differed significantly between diabetics and nondiabetics and had a significant association with the development of T1DM in Egyptians [25,26] and Brazilians [27].

It is worthy to note that the distribution of VDR (*BsmI* and *FokI*) polymorphisms and their association with the development of T1DM were not only controversial in the different populations but even in the same one. In an Asian study, the regional

Table 3: Distribution of *FokI* variant genotypes and alleles in cases and controls

FokI (228570 G>A)	Cases (n=50) [n (%)]	Controls (n=50) [n (%)]	P value (χ^2)
Genotypes			
GG	27 (54.0)	20 (40.0)	^{MC}P=0.244
AG	20 (40.0)	28 (56.0)	
AA	3 (6.0)	2 (4.0)	
Recessive gene model AA vs. (AG + GG)			
AA	3 (6.0)	2 (4.0)	^{FE}P=0.495
AG + GG	47 (94.0)	48 (96.0)	
Dominant gene model (AA + AG) vs. GG			
AA + AG	23 (46.0)	30 (60.0)	0.161
GG	27 (54.0)	20 (40.0)	
Allele			
G	74 (74.0)	68 (68.0)	0.350
A	26 (26.0)	32 (32.0)	

χ^2, χ^2 test, ^{MC}P, Monte Carlo exact test, ^{FE}P, Fisher's exact test. P value is statistically significant at P value less than or equal to 0.05.

Table 4: Association of biallelic haplotypes (*BsmI* and *FokI*) in cases and controls

Biallelic haplotypes	Cases (n=100) [n (%)]	Controls (n=100) [n (%)]	P value (χ^2)
AC	18 (18.0)	25 (25.0)	
AT	8 (8.0)	7 (7.0)	0.688
GC	34 (34.0)	32 (32.0)	
GT	40 (40.0)	36 (36.0)	

χ^2, χ^2 test. P value is statistically significant at P value less than or equal to 0.05.

stratification analysis showed that the *BsmI* variant conferred an increased T1DM risk in East Asian population, while the *FokI* variant was increased in the West Asian one [28]. Also, published results concerned with the *FokI* variant were disputed in the Egyptian population where it was associated with T1DM susceptibility in two studies [16,25] while it was not associated in a third one [22]. Such a discrepancy in the results of similar studies in different populations might be explained by ethnic-specific genetic polymorphisms, the possibility of population stratification bias when analyzing the samples, variable gene expressions, different and small sample sizes, using different analytical methods for genotyping, different clinical inclusion and exclusion criteria, and variations in the geo-epidemiological and environmental factors affecting these populations as the exposure to ultraviolet irradiation and diet.

The current study suggests that the combined effect of the biallelic haplotypes of *BsmI*-rs1544410 and *FokI*-rs2228570 does not contribute significantly to the development of T1DM as the association of these biallelic haplotypes showed no statistically significant difference when compared between cases and controls. A meta-analysis [29] and a study of Colombian families [30] were in accordance with this finding. Disagreeing with this finding, interaction of *Bsm*I and *Fok*I *VDR* haplotypes with each other contributed significantly to disease susceptibility [31].

Conclusion

This study suggested no association between *VDR* (*BsmI*-rs1544410 and *FokI*-rs2228570) variants and T1DM in Egyptian children.

Further studies involving larger numbers of participants with the estimation of vitamin D deficiency and combining genetic information from numerous risk loci can assist the prediction of T1DM and are required to better understand ethnic-specific genetic associations with T1DM and allow early intervention in genetically at-risk infants.

Acknowledgements

The contributors in this study acknowledge the general support and the facility to use the equipment of Faculty of Medicine, Cairo University and the National Institute of Diabetes and Endocrinology, Cairo, Egypt.

Source(s) of support: patients were selected from the Pediatrics Outpatient Clinic of the National Institute of Diabetes and Endocrinology, Cairo, Egypt. All the laboratory investigations were conducted by the Clinical and Chemical Pathology Department of the National Institute of Diabetes and Endocrinology, Cairo, Egypt; equipment facility except the genotyping was conducted at the facility of Clinical Pathology Departments, Faculty of Medicine, Cairo University, Egypt.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

References

1. Mobasseri M, Shirmohammadi M, Amiri T, Vahed N, Hosseini Fard H, Ghojazadeh M. Prevalence and incidence of type 1 diabetes in the world: a systematic review and meta-analysis. Health Promot Perspect 2020; 10:98–115.
2. El-Ziny MA, Salem NA, El-Hawary AK, Chalaby NM, Elsharkawy AA. Epidemiology of childhood type 1 diabetes mellitus in Nile Delta, northern Egypt – a retrospective study. J Clin Res Pediatr Endocrinol

2014; 6:9–15.
3. Hassan FM, Khatab AA, Abo El-Fotoh WM, Ganh IN. Prevalence of diabetes among school-age children. Menoufia Med J 2019; 32:305–310.
4. Rak K, Bronkowska M. Immunomodulatory effect of vitamin D and its potential role in the prevention and treatment of type 1 diabetes mellitus—a narrative review. Molecules 2019; 24:53.
5. Maestro MA, Molnár F, Mouriño A, Carlberg C. Vitamin D receptor 2016: novel ligands and structural insights. Expert Opin Ther Patents 2016; 26:1291–1306.
6. Elfasakhany FM, Al-Qahtani ON, Badri AM, Abuelela HA. Analysis of vitamin D receptor gene polymorphism in chronic periodontitis among Saudi population. Eur Dent Res Biomater J 2021; 2:12–16.
7. Makoui MH, Imani D, Motallebinezhad M, Azimi M, Razi B. Vitamin D receptor gene polymorphism and susceptibility to asthma: meta-analysis based on 17 case-control studies. Ann Allergy Asthma Immunol 2020; 124:57–69.
8. Zhai N, Bidares R, Makoui MH, Aslani S, Mohammadi P, Razi B, *et al*. Vitamin D receptor gene polymorphisms and the risk of the type 1 diabetes: a meta-regression and updated meta-analysis. BMC Endocr Disord 2020; 20:121.
9. American Diabetes Association, 2. Classification and Diagnosis of Diabetes: *Standards of Medical Care in Diabetes—2018*. Diabetes Care 2018; 41 (Supplement_1):S13–S27.
10. Rifai N, Warnick GR. Measurement of lipids, lipoproteins, and apolipoproteins. In: Burtis CA, Ashwood ER, Bruns DE, editors. *Tietz textbook of clinical chemistry and molecular diagnosis*. 4th ed. Louis, Missouri: Elsevier Saunders; 2006:938-952.
11. Winkler C, Haupt F, Heigermoser M, Zapardiel-Gonzalo J, Ohli J, Faure T, *et al*. Identification of infants with increased type 1 diabetes genetic risk for enrollment into primary prevention trials-GPPAD-02 study design and first results. Pediatr Diabetes 2019; 20:720–727.
12. Motohashi Y, Yamada S, Yanagawa T, Maruyama T, Suzuki R, Niino M, *et al*. Vitamin D receptor gene polymorphism affects onset pattern of type 1 diabetes. J Clin Endocrinol Metab 2003; 88:3137–3140.
13. Zhang J, Li W, Liu J, Wu W, Ouyang H, Zhang Q, *et al*. Polymorphisms in the vitamin D receptor gene and type 1 diabetes mellitus risk: an update by meta-analysis. Mol Cell Endocrinol 2012; 355:135–142.
14. Lemos M, Fagulha A, Coutinho E, Gomes L, Bastos M, Barros L, *et al*. Lack of association of vitamin D receptor gene polymorphisms with susceptibility to type 1 diabetes mellitus in the Portuguese population. Hum Immunol 2008; 69. 134–138.
15. Panierakis C, Goulielmos G, Mamoulakis D, Petraki E, Papavasiliou E, Galanakis E. Vitamin D receptor gene polymorphisms and susceptibility to type 1 diabetes in Crete, Greece. Clin Immunol 2009; 133:276–281.
16. Abd-Allah SH, Pasha HF, Hagrass HA, Alghobashy AA. Vitamin D status and vitamin D receptor gene polymorphisms and susceptibility to type 1 diabetes in Egyptian children. Gene 2014; 536:430–434.
17. Rasoul MA, Haider MZ, Al-Mahdi M, Al-Kandari H, Dhaunsi GS. Relationship of four vitamin D receptor gene polymorphisms with type 1 diabetes mellitus susceptibility in Kuwaiti children. BMC Pediatr 2019; 19:71.
18. Ahmed AE, Sakhr HM, Hassan MH, El-Amir MI, Ameen HH. Vitamin D receptor rs7975232, rs731236 and rs1544410 single nucleotide polymorphisms, and 25-hydroxyvitamin D levels in Egyptian children with type 1 diabetes mellitus: effect of vitamin D co-therapy. Diabetes Metab Syndr Obes 2019; 12:703–716.
19. Iyer A, Lanham-New S, Khoja S, Al-Ghamdi M, Al Doghaither H. Relationship between vitamin D receptor gene polymorphisms and type 1 diabetes mellitus in Saudi patients. Int J Pharmacol 2017; 13:1092–1097.
20. Mory DB, Rocco ER, Miranda WL, Kasamatsu T, Crispim F, Dib SA. Prevalence of vitamin D receptor gene polymorphisms FokI and BsmI in Brazilian individuals with type 1 diabetes and their relation to beta-cell autoimmunity and to remaining beta-cell function. Hum Immunol 2009; 70:447–451.
21. Cheon CK, Nam HK, Lee KH, Kim SY, Song JS, Kim C. Vitamin D receptor gene polymorphisms and type 1 diabetes mellitus in a Korean population. Pediatr Int 2015; 57:870–874.
22. Hamed EO, Abdel-Aal AM, Din AK, Atia MM. Vitamin D level and Fok-I vitamin D receptor gene polymorphism in Egyptian patients with type-1 diabetes. Egypt J Immunol 2013; 20:1–10.
23. Thorsen SU, Mortensen HB, Carstensen B, Fenger M, Thuesen BH, Husemoen L, *et al*. No association between type 1 diabetes and genetic variation in vitamin D metabolism genes: a Danish study. Pediatr Diabetes 2014; 15:416–421.
24. Nasreen M, Lone KP, Khaliq S, Khaliq S. Serum vitamin D levels and gene polymorphisms (Fok1 and Apa1) in children with type I diabetes and healthy controls. J Pak Med Assoc 2016; 66:1215–1220.
25. El Kafoury A, Haroun M, Embaby A, Dawoods A. The association of polymorphic sites in some genes with type 1 diabetes mellitus in a sample of Egyptian children. Egypt J Med Human Genet 2014; 15.
26. Mohamed E, Lamia AM, Hany AM, Azza AK, Nahla I, Abdallah SA, Nora M. Vitamin D receptor gene (Fok-I) polymorphisms in type 1 diabetic children; case study in Zagazig University Hospitals. Egypt J Hosp Med 2021; 85:4223–4226.
27. Mory DB, Gabbay MA, Rocco ER, Kasamatsu T, Crispim F, Miranda WL, Dib SA. High frequency of vitamin D receptor gene polymorphism FokI in Brazilian type 1 diabetes mellitus patients with clinical autoimmune thyroid disease. Diabetol Metab Syndr 2016; 8:29.
28. Wang G, Zhang Q, Xu N, Xu K, Wang J, He W, Yang T. Associations between two polymorphisms (FokI and BsmI) of vitamin D receptor gene and type 1 diabetes mellitus in Asian population: a meta-analysis. PLoS ONE 2014; 9:e89325.
29. Guo SW, Magnuson VL, Schiller JJ, Wang X, Wu Y, Ghosh S. Meta-analysis of vitamin D receptor polymorphisms and type 1 diabetes: a HuGE review of genetic association studies. Am J Epidemiol 2006; 164:711–724.
30. Natalia GL, Alejandra R, Martin T, Maria VL, Alejandra V, *et.al*. VDR gene haplotypes associate with type 1 diabetes and suggest interaction with HLA, IL2RA and CTLA4 loci in Colombian families. Curr Res Diab Obes J 2018; 6:555688.
31. Tizaoui K, Kaabachi W, Hamzaoui A, Hamzaoui K. Contribution of VDR polymorphisms to type 1 diabetes susceptibility: systematic review of case-control studies and meta-analysis. J Steroid Biochem Mol Biol 2014; 143:240–249.