

VDR FokI rs2228570 SNP in Autoimmune Thyroiditis: Literature Review

Shaikh Parveen Aleena, Khurana Rajneesh, Megi Inaishvili, Nameera Parveen Shaikh, Salome Glonti, Nino Kedelidze, Irina Nakashidze¹

Abstract

Autoimmune thyroiditis disease (AITD) is a widespread autoimmune pathology. Notably, Genetics factors have a key contribution to the clinico-pathogenesis of AITD. It's suggested that several immune-related genes have been implicated in genetic susceptibility to AIT. Vitamin D receptor (VDR) and its system is involved in an orchestrated mechanism of inflammatory response that leads to autoimmunity. In this review, we summarize together the VDR FokI rs2228570 SNP in autoimmune thyroiditis. Based on the literature review, we have concluded that rs2228570 SNP may have susceptibility and predisposition to develop AITD. (TCM-GMJ December 2022; 7 (4):P10-P13)

Keywords: Autoimmune Thyroiditis, Vitamin D polymorphism, FokI, rs2228570.

Introduction

Autoimmune thyroiditis (AIT) is a widespread autoimmune pathology. Notably, Genetics factors have a key contribution to the clinico-pathogenesis of AITD. It's suggested that several immune-related genes have been implicated in genetic susceptibility to AIT.

Vitamin D receptor (VDR) and its system is involved in an orchestrated mechanism of inflammatory response that leads to autoimmunity. Among Autoimmune thyroid Diseases (AITDs), Hashimoto's Thyroiditis (HT) also known as Autoimmune Thyroiditis (AIT) is the most common manifestation. The etiology of the AIT is complex and unknown, although the interaction between [environmental factors](#) and genetic predisposition plays an important role. Environmental factors such as drugs, iodine intake, irradiation, viral infections, and hormonal effects can directly affect thyrocytes and may have adverse immunomodulatory and toxic effects.

Genetic factors predominate about 70-80% in the development of the disease, and as with many other autoimmune diseases; females are more affected. Vitamin D is now known to have pleiotropic effects. Several novel roles have been identified in the last years including Autoimmune diseases, COVID-19, Depression, Fibrosis etc. Vitamin D has been investigated in affecting thyroid diseases incidence AITD as well. (1)

Vitamin D receptor (VDR) polymorphism is extensively studied in its association with many autoimmune disorders such as Dia-

betes Mellitus 1, Systemic Lupus Erythematosus, Multiple Sclerosis, Rheumatoid Arthritis, and Tuberculosis. One such influence can be seen in Autoimmune Thyroiditis. However, the effect of VDR SNPs on susceptibility to autoimmunity differs depending on distinct populations and ethnicity.

VDR gene role in AIT. Vitamin D receptor (VDR) is an intranuclear receptor. VDR gene is located on chromosome 12q13.1 which consists of 11 exons. (23) Genetic variants in regulatory parts of the VDR gene are studied under VDR polymorphism. Vitamin D suppresses the immune system from thyroid autoimmunity by binding to the VDR, expressed in immune cells (including, monocytes, macrophages, dendritic cells, and T and B lymphocytes and etc). The binding of its receptor (VDR) and its ligand leads to an anti-inflammatory response to innate immunity contributing to the development of immunosuppressive and regulatory effects on the adaptive immune system. (2) There are about sixty identified VDR single nucleotide polymorphisms (SNPs), Fok I rs2228570, Apa I rs7975232, Taq I rs731236, and Bsm I rs1544410, which are linked with an increased risk of AITD. It has been demonstrated that patients with lower serum vitamin D had disease manifestations of AITDs. (3) Vitamin D as an immunomodulator is involved in the onset and progression of AITD. In patients with AITD, low levels of vitamin D were observed; additionally, levels of antithyroid antibodies and thyroid volume were related to vitamin D deficiency influencing the duration of HT. (4)

VDR belongs to the thyroid/steroid receptor family, an intracellular nuclear receptor. A Vitamin D/VDR complex is formed after binding to its receptor, which initiates a chain of events. Following complex formation, it is translocated to the nucleus and forms a heterodimer with retinoid X receptor (RXR) and upon recognition it binds with transcription factor IIB (TFIIB) to Vitamin D response element (VDRE). This finally leads to transcription activation or suppression of Vita-

From the ¹Batumi Shota Rustaveli State University, Batumi, Georgia.
Received December 08, 2022; accepted December 20, 2022.
Address requests to: Aleena Parveen Shaikh

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min D response genes. VDR gene polymorphism contributes to altered activation and modified effects on immune cell interactions.

Autoimmune thyroiditis (AIT) is a widespread autoimmune pathology. Notably, Genetics factors have a key contribution to the clinico-pathogenesis of AITD. It's suggested that several immune-related genes have been implicated in genetic susceptibility to AIT.

VDR SNPs and AIT. SNPs such as VDR rs1544410 (BsmI), rs7975232(ApaI), and rs731236 (TaqI) polymorphisms have shown susceptibility to Hashimoto thyroiditis development. Research done on 145 Croatian HT patients and 145 age-, sex- and ethnically matched healthy euthyroid controls were genotyped for VDR polymorphism and further studied. The results demonstrated that the BsmI-TaqI bT haplotype was significantly frequent in individuals with diagnosed HT, whereas the BsmI-TaqI BT haplotype was more often seen in healthy controls. Thus, demonstrating various protective and susceptible haplotypes. (5)

Distinct races and environments have shown variable GD susceptibility with VDR gene polymorphism. For instance, VDR rs7975232 polymorphism was associated with an increased risk of GD risk in Chinese populations (30.34% vs. 25.42% in controls; $P = 0.041$). (6) Similarly, association in Apa I has been observed in Southwest Chinese Han population. A case control cohort study including a total of 650 Chinese GD patients and 1209 controls were selected. When investigating the susceptibility, results indicated that AA genotype and A allele of VDR/Apa I were significantly associated; in contrast, other polymorphisms such as FokI, TaqI, and BsmI showed no substantial relations. As a result, it can be suggested that the development of GD is affected by VDR mRNA expression and levels of secreted cytokines. (7) In an evaluation based on ethnicity from a meta-analysis, it can be established that VDR rs1544410 polymorphism has shown an increased risk in the Asian population while the decreased risk of AITD in African and European populations. On the other hand, VDR rs731236 polymorphism in the Asian and African populations is susceptible to increased risk of AITD like Hashimoto's thyroiditis and Graves, while no relation has been found in the European population. This concludes variation with ethnicity also varies with VDR polymorphism (8).

FokI (rs2228570) SNPs and AIT. FokI (rs2228570) located on the start codon of the VDR gene. It is suggested that the Rs2228570 is a functional SNP. This section reviews the role of FokI SNP in autoimmune Thyroiditis. In research on the Serbian population, 44 female patients with the diagnosis of Hashimoto thyroiditis and decreased or deficient levels of Vitamin D were selected, and 32 age-matched, sex-matched healthy controls from the same geographic were selected. A significant difference was noted when VDR-FokI polymorphisms were compared between HT and control patients (p value < 0.05). Increased risk of disease development can be seen when comparing patients with mutation to those without mutation (odds ratio: 4.472). (9) It has been established by Djurovic et al. that FokI polymorphism significantly influences disease occurrence in the Serbian population and suggested that the female population with FF genotype may have a higher risk of HT occurrence. (9)

In the Asian population, when frequencies of the CC genotype and C allele for the VDR rs2228570 polymorphism were compared in the Japanese population, results showed higher polymorphisms in Hashimoto thyroiditis patients with low levels of serum Vitamin D levels than in control subjects (P values = 0.0174 and 0.0458, respectively). (10) The CC genotype is implicated in directly causing autoimmune thyroid destruction while C allele is involved in increased interleukin production of inter-

leukin 12 (IL-12), resulting in cytotoxic T cell and Th 1 cell-mediated thyroid destruction explaining the pathogenesis. This data suggests that in patients with HT, immune regulation by VDR is thus suppressed.

Similar VDR-FokI genotype distribution was noted between Chinese and Japanese populations. Lin et al. demonstrated that in the Chinese population, 36.7% CC genotypes were present in HT patients compared to controls (23.3%), and a comparable significant difference in VDR SNP genotypes was also noted (P value: 0.0458). (11)

These data are consistent with the study of Zarrin et al. in north-west Iran. (12) 121 adult patients suffering from autoimmune Thyroiditis and Graves disease (GD) and 117 healthy controls age and sex matched in the same population were compared. The results demonstrated that patients with FokI CC and CT genotype showed an increased risk of AITDs and precisely, genotype CC showed increased disease risk occurrence of Hashimoto thyroiditis ($p = 0.04$; OR= 3.38). (12)

Yazici et al. also observed that 111 patients were compared with 159 healthy controls in studying their VDR genes such as FokI, BsmI, and ApaI by polymerase chain reaction (PCR) -based restriction analysis method. Results demonstrated an increased risk of Hashimoto thyroiditis incident was seen in patients carrying FF FokI genotype and TT TaqI genotype; on the contrary heterogeneous genotypes BbAaTtFf offered protection for HT in the population. Implying that VDR rs731236 and rs2228570 polymorphisms were significantly linked with susceptibility risk in the Turkish population. (13) These findings were also consistent in the Egyptian population. A Study done by Hanna et al., 2021 included 112 HT and 48 hypothyroid patients as control. Results were consistently confirmed FokI polymorphism in HT patients (11.4%) compared to controls with zero occurrences. Furthermore, among the general Egyptian population there was a low frequency of FF genotype in non-HT and hypothyroid (control patients), about 2.6% and 6%, respectively. This proves the association between VDR FokI occurrence and autoimmunity in the population (14).

The presence of dominant or recessive genes affects risk assessment. The presence of homozygous genotype (FF) was higher in patients of Hashimoto's Thyroiditis (p value= 0.0002 and OR= 2.22) while heterozygous genotype (Ff) (OR=0.63; $p=0.029$) and homozygous recessive genotype (ff) (OR=0.40; $p=0.017$) were lower; signifying that patients carrying dominant genotype show two times higher risk of Hashimoto's Thyroiditis while heterozygous and recessive genotype were protective in disease incidence in Iraqi population (15). This result agrees with studies of Yazici et al., which also observed that heterozygous genotypes showed a decreased risk of development of AITDs in the Turkish population. (13) A meta-analysis by Wang et al. also demonstrates that the dominant F allele of the VDR- FokI gene has a higher risk of HT (OR=1.44, $p=0.010$) when compared with the combined Ff and ff genotype. (3)

Recent studies show an association between low serum levels of Vitamin D and autoimmune thyroid pathologies such as HT. One such example is a study done in a west-Ukrainian population consisting of 153 patients of various thyroid pathologies. Results showed that patients suffering with hypothyroidism and carrying AA genotype had significantly low levels of Vitamin D by 18.8% irrespective of their initial cause (postoperative or autoimmune) compared to AIT patients without hypothyroidism suggesting that low levels of vitamin D intensify thyroid insufficiency. Different pathologies also showed differences in vitamin D levels. Significant decrease in Vitamin D levels about 1.89 and 2.05 times, were seen in patients suffering from postoperative hypothyroidism and AIT-induced hypothyroidism respectively

when compared to control group. (16)

In the Taiwanese Chinese population, increased frequency of CC genotype and C allele of VDR FokI polymorphism were studied in GD patients. Thus, drawing conclusions that VDR FokI T/C polymorphism could probably be used as a genetic biomarker for predicting Graves disease progression and incidence. (17)

The Different population shows variation in susceptibility to disease progression. Zhou et al., 2009 demonstrated that FokI (OR 1.68 (95% CI: 1.28-2.20, P = 0.0002), ApaI and BsmI polymorphism showed an increased risk of development of GD in the Asian population while on contradictory TaqI, FokI, ApaI and BsmI polymorphism showed no correlation in disease development in the Caucasian population. (18) Ramos-Lopez et al., 2005 report such association wherein three populations have differentially distributed VDR polymorphism. 789 Graves patients and 823 healthy controls were analyzed for VDR polymorphism. German and polish populations but not the Serbian population demonstrated an increased association between polymorphism and Graves disease incidence. Variant f (p= 0.0024) of VDR FokI polymorphism was found to be associated in the German population while variant F (p= 0.0049) in the Polish population. (19) Concluding that different populations with the same VDR polymorphism show variation in disease occurrence. VDR gene SNPs are associated with numerous disease predisposition. Studies have shown predisposition to Pancreatic and colon cancers. (24-26) FokI polymorphism is also associated with an increased risk of cancer susceptibility as well. Multiple studies have shown increased risk for ovarian, prostate and skin cancer with FokI polymorphism. In addition, FokI T/T genotype was suggested to be associated with increased risk of head and neck squamous cell carcinoma (20). To understand the relationship of FokI VDR gene polymorphism and clinical significance of Papillary Thyroid Cancer (PTC) development, a case-controlled study was conducted in the Turkish population consisting of 165 patients with PTC and 172 controls. The Frequency of VDR FokI polymorphism increased in patients suffering from PTC as compared to controls. Significant differences were linked to different allele distributions in susceptibility to PTC. The following results were observed, patients with FokI CT/TT or TT genotype compared to CC genotype had an increased risk of T3 and T4 PTC (TT vs CC: OR = 2.71, 95% CI = 1.13–6.53, p = 0.003) and extra thyroid invasions development. Moreover, they were more expected to develop Stage III and IV PTC and multifocal tumors (TT vs CC: OR = 5.20, 95% CI = 1.93–11.94; p = 0.001) compared to CC genotype. It was observed that patients having tumor size more than or equal to 10mm compared to <10mm carried TT genotype. In conclusion, patients with VDR gene FokI were not only associated with an increased risk of PTC but also adverse pathologic and prognostic factors and hence can be used as poor biomarkers in PTC patients (21). It is important to understand that Hashimoto's thyroiditis and PTC are relatively common diseases with interlinked etiologies and progression into one another.

Efficacy of VDR against therapy which is used to decrease viable thyroid cancer cell count, has been found to vary on FF or ff genotype of FokI VDR polymorphism. FF FokI variant is associated with greatest resistance while ff FokI variant has been associated with a good response to therapy with a decrease in viable cell count. This observation can be interpreted as that FF FokI polymorphism is associated with aggressive cancer progression. (22)

However, some populations failed to be consistent with this association. For example, in a study performed in patients of Caucasian polish origins, 223 adult patients suffering from AIT

were studied and compared to 130 unrelated controls of the same origin. The study failed to find a strong association in rs2228570 FokI polymorphism affecting the risk of disease occurrence between AIT and controls at least in the population mentioned above. (1) Similarly, Meng et al. failed to demonstrate such an association among the Chinese population. (6) Such contradictory outcomes were also observed by Feng et al., 2013, which suggested that TaqI and BsmI polymorphism are associated with AITD susceptibility rather than FokI and ApaI polymorphism. (16) The inconsistency in results shows the association is influenced due to distinct ethnicities, different geographical regions, and lifestyle causes such as diet, and sunlight exposure.

Conclusion

rs2228570 Polymorphism is associated with autoimmune thyroid susceptibility. Thus, based on the existing references, VDR FokI rs2228570 has shown a strong correlation in AITD progression and susceptibility.

Funding: This research was funded by Batumi Shota Rustaveli State University, the grant №02-12.57.

References

- Maciejewski A, Kowalczyk MJ, Herman W, Czyżyk A, Kowalska M, Żaba R, Łączka K. Vitamin D receptor gene polymorphisms and autoimmune thyroiditis: are they associated with disease occurrence and its features?. *BioMed research international*. 2019 Aug 21;2019.
- Bizzaro G, Antico A, Fortunato A, Bizzaro N. Vitamin D and autoimmune diseases: is vitamin D receptor (VDR) polymorphism the culprit. *Isr Med Assoc J*. 2017 Jul 1;19(7):438-43.
- Wang J, Lv S, Chen G, Gao C, He J, Zhong H, Xu Y. Meta-analysis of the association between vitamin D and autoimmune thyroid disease. *Nutrients*. 2015 Apr 3;7(4):2485-98.
- Bizzaro G, Shoenfeld Y. Vitamin D and autoimmune thyroid diseases: facts and unresolved questions. *Immunologic research*. 2015 Feb;61(1):46-52.
- Štefanić M, Papić S, Suver M, Glavaš-Obrovac L, Karner I. Association of vitamin D receptor gene 3'-variants with Hashimoto's thyroiditis in the Croatian population. *International Journal of Immunogenetics*. 2008 Apr;35(2):125-31.
- Meng S, He ST, Jiang WJ, Xiao L, Li DF, Xu J, Shi XH, Zhang JA. Genetic susceptibility to autoimmune thyroid diseases in a Chinese Han population: Role of vitamin D receptor gene polymorphisms. In *Annales d'endocrinologie* 2015 Dec 1 (Vol. 76, No. 6, pp. 684-689). Elsevier Masson.
- Zhou F, Liang Z, Wang X, Tan G, Wei W, Zheng G, Ma X, Tian D, Li H, Yu H. The VDR gene confers a genetic predisposition to Graves' disease and Graves' ophthalmopathy in the Southwest Chinese Han population. *Gene*. 2021 Aug 15;793:145750.
- Gao XR, Yu YG. Meta-analysis of the association between vitamin D receptor polymorphisms and the risk of autoimmune thyroid disease. *International journal of endocrinology*. 2018 Mar 22;2018.
- Djurovic J, Stojkovic O, Ozdemir O, Silan F, Akurut C, Todorovic J, Savic K, Stamenkovic G. Association between FokI, ApaI and TaqI RFLP polymorphisms in VDR gene and Hashimoto's thyroiditis: preliminary data from female patients in Serbia. *International Journal of Immunogenetics*. 2015 Jun;42(3):190-4.
- Inoue N, Watanabe M, Ishido N, Katsumata Y, Kagawa T, Hidaka Y, Iwatani Y. The functional polymorphisms of VDR, GC and CYP2R1 are involved in the pathogenesis of autoimmune thyroid diseases. *Clinical & Experimental Immunology*. 2014 Nov;178(2):262-9.
- Lin WY, Wan L, Tsai CH, Chen RH, Lee CC, Tsai FJ. Vitamin D receptor gene polymorphisms are associated with risk of Hashimoto's thyroiditis in Chinese patients in Taiwan. *Journal of clinical laboratory analysis*. 2006;20(3):109-12.
- Zarrin R, Bagheri M, Mehdizadeh A, Ayremlou P, Faghfour AH. The association of FokI and ApaI polymorphisms in vitamin D receptor gene with autoimmune thyroid diseases in the northwest of Iran. *Medical journal of the Islamic Republic of Iran*. 2018;32:4.
- Yazici D, Yavuz D, Tarcin O, Sancak S, Deyneli O, Akalin S. Vitamin D receptor gene ApaI, TaqI, FokI and BsmI polymorphisms in a group of Turkish patients with Hashimoto's thyroiditis. *Minerva Endocrinologica*. 2013;38:195-201.
- Hanna HW, Rizzo C, Halim RM, El Haddad HE, Salam R, Abou-Youssef HE. Vitamin D status in Hashimoto's thyroiditis and its association with vitamin D receptor genetic variants. *The Journal of Steroid Biochemistry and Molecular Biology*. 2021 Sep 1;212:105922.

15. Majid WJ, Algenabi AH, Alwadees AA. Vitamin-D Receptor (VDR) Gene Polymorphisms (FokI and TaqI) in Patients with Hashimoto's Thyroiditis of Iraqi Population. *European Journal of Molecular & Clinical Medicine*. 2020 Nov 1;7(2):64-8.
16. Kamyshna II, Pavlovych LB, Malyk IV, Kamyshnyi AM. 25-OH Vitamin D blood serum linkage with VDR gene polymorphism (rs2228570) in thyroid pathology patients in the West-Ukrainian population. *Journal of Medicine and Life*. 2021 Jul;14(4):549.
17. Chen R, Chang C, Chen H, Chen W, Tsai C, Tsai F. Association between vitamin-D receptor gene FokI polymorphism and Graves' disease among Taiwanese Chinese. *Journal of Clinical Laboratory Analysis*. 2007;21(3):173-177.
18. Zhou H, Xu C, Gu M. Vitamin D receptor (VDR) gene polymorphisms and Graves' disease: a meta-analysis. *Clinical endocrinology*. 2009 Jun;70(6):938-45.
19. Ramos-Lopez E, Kurylowicz A, Bednarczuk T, Paunkovic J, Seidl C, Badenhop K. Vitamin D receptor polymorphisms are associated with Graves' disease in German and Polish but not in Serbian patients. *Thyroid*. 2005 Oct 1;15(10):1125-30.
20. Hama T, Norizoe C, Suga H, Mimura T, Kato T, Moriyama H, Urashima M. Prognostic significance of vitamin D receptor polymorphisms in head and neck squamous cell carcinoma. *PLoS One*. 2011 Dec 29;6(12):e29634.
21. Beysel S, Eyerci N, Pinarli FA, Apaydin M, Kizilgul M, Caliskan M, Ozcelik O, Kan S, Cakal E. VDR gene FokI polymorphism as a poor prognostic factor for papillary thyroid cancer. *Tumor Biology*. 2018 Nov;40(11):1010428318811766.
22. Sharma V, Fretwell D, Crees Z, Kerege A, Klopper JP. Thyroid cancer resistance to vitamin D receptor activation is associated with 24-hydroxylase levels but not the ff FokI polymorphism. *Thyroid*. 2010 Oct 1;20(10):1103-11.
23. Apaydin M, Beysel S, Eyerci N, Pinarli FA, Ulubay M, Kizilgul M, Ozdemir O, Caliskan M, Cakal E. The VDR gene FokI polymorphism is associated with gestational diabetes mellitus in Turkish women. *BMC Medical Genetics*. 2019 Dec;20(1):1-6.
24. Nakashidze I, Dariya B, Peshkova T, Beridze S. The Genetic Polymorphisms in Colon Cancer. *Critical Reviews™ in Oncogenesis*. 2020;25(4).
25. Nakashidze I, Ahmad S. Genetic predisposition for pancreatic cancer. In: *Theranostic Approach for Pancreatic Cancer* 2019 Jan 1 (pp. 153-169). Academic Press.
26. Nakashidze I, Petrović N, Kedelidze N, Dariya B. Clinical Significance of Genetic Variants in Colon Cancer. In: *Colon Cancer Diagnosis and Therapy* Vol. 3 2022 (pp. 69-91). Springer, Cham.