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ORIGINAL RESEARCH

Vitamin D Receptor Gene Polymorphisms and Association with Vitiligo in Indonesian Population

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Introduction: Vitiligo is an acquired depigmenting skin disorder due to the loss of melanocyte function in the epidermis and hair follicles. The pathogenesis of vitiligo is multifactorial, with genetics being a predisposing factor. Previous studies had varying results regarding whether or not polymorphisms of vitamin D receptor (*VDR*) gene are associated with the risk of vitiligo in specific populations. This study investigated the association between three frequently analyzed *VDR* gene polymorphisms (*ApaI, BsmI, TaqI*) and susceptibility to vitiligo in Indonesian population.

Methods: Thirty-four vitiligo patients and 34 age- and sex-matched healthy subjects aged ≥18 years old were recruited in the Dermatology and Venereology Outpatient Clinic of Dr. Hasan Sadikin General Hospital, Bandung, Indonesia. Genomic deoxyribonucleic acid (DNA) was extracted from the peripheral blood using a DNA isolation kit. *VDR* gene polymorphisms (*ApaI, BsmI, and TaqI*) were investigated using the polymerase chain reaction-restriction-fragment length polymorphism method. The differences of genotype distributions and allele frequencies were statistically compared between case and control groups using Chi-square test.

Results: *VDR* gene polymorphisms were identified in 68 participants, consisting of Aa (n = 14), aa (n = 20), Bb (n = 15), bb (n = 19), and TT (n = 34) genotypes in the case group. In the control group, Aa (n = 6), aa (n = 28), Bb (n = 17), bb (n = 17), and TT (n = 34) genotypes were identified. However, only subjects with *ApaI* Aa genotype polymorphism had a 3.267-fold increased risk of developing vitiligo.

Conclusion: This study showed that *ApaI* Aa genotype polymorphism of the *VDR* gene increases the risk of vitiligo in Indonesian population.

Keywords: Indonesia, polymorphism, vitamin D receptor, vitiligo

Introduction

Vitiligo is an acquired depigmenting skin disorder due to the loss of melanocytes in the epidermis and hair follicles, affecting 0.5–1% of the population worldwide.[1–4] Many factors have been proposed to play a role in the pathogenesis of vitiligo, including autoimmune dysfunction, neural theory, and oxidative stress.[1,5] Discoveries in association studies as well family studies revealed genetic as a known predisposing factor of the depigmentation process in vitiligo, with 30–40% of the cases tend to have a positive family history.[6–8]

Nowadays, vitamin D analogs have been used as a treatment for vitiligo.[1,6] Vitamin D is a lipophilic hormone with protective and restorative effects on melanocytes.[9] It also inhibits specific cell activation and several proinflammatory cytokines, and reduces autoimmune damage on melanocytes.[2,9] The active form of vitamin D exerts its effects through the vitamin D receptors (VDRs) on target cells,[9] including melanocytes and lymphocytes.[2] The *VDR* is encoded by the *VDR* gene, located in chromosome 12q12–14.[2,9] More than 25 single-nucleotide polymorphisms (SNPs) have been described at the *VDR* gene locus.[2] The three most frequently analyzed adjacent polymorphisms of the *VDR* gene, namely *ApaI* (rs 7975232), *BsmI* (rs1544410), and *TaqI* (rs731236), are located in intron 8/exon 9.[2,10] These polymorphisms have been reported to be associated with several diseases, including vitiligo.[2] However, most studies evaluating the association

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of *VDR* gene polymorphisms and vitiligo susceptibility in specific populations, such as the Caucasian, Chinese, Pakistan, Northwestern Mexico, and Egyptian populations, showed varying results.[2,9–14] Therefore, this study aimed to analyze whether the three most common *VDR* gene polymorphisms might be associated with vitiligo susceptibility in the Indonesian population.

Subject and Methods

Subjects

This case-control study was conducted between April 2021 and December 2021. Blood samples were collected in the Dermatology and Venereology Outpatient Clinic of Dr. Hasan Sadikin General Hospital Bandung, Indonesia. The study was approved by the Research Ethics Committee of Dr. Hasan Sadikin General Hospital (Ethical Clearance number: LB.02.01/X.6.5/100/2021). Written informed consent was obtained from all participants.

A total of 68 participants, consisted of 34 vitiligo patients and 34 age- and sex-matched healthy subjects, were enrolled in this study. The inclusion criteria for the vitiligo group were vitiligo patients aged ≥18 years old who regularly visited the hospital during the study. For the control group, the inclusion criteria were vitiligo-free healthy individuals aged ≥18 years old who had no family history of vitiligo. Exclusion criteria for both groups were participants with a history and clinical examination results indicating multiple sclerosis, inflammatory bowel disease, Graves' disease, diabetes mellitus, Behçet disease, melanoma, colorectal carcinoma, and breast cancer.

Demographic and clinical characteristics were documented for all vitiligo patients, including age, sex, occupation, how long has the patient had this condition, age of onset, types of vitiligo, family history of vitiligo, stability of vitiligo, presence of leukotrichia, history of precipitating factors, and serum 25 hydroxyvitamin D [25(OH)D] level. Vitiligo Area Severity Index (VASI) and body surface area (BSA) were also measured. Vitiligo patients were classified into three major types according to the 2012 Vitiligo Global Issues Consensus Conference, namely: non-segmental vitiligo (NSV), segmental vitiligo (SV), and unclassified/undetermined vitiligo. The disease course was considered stable if the lesions had no increased activity within 12 months.

Deoxyribonucleic Acid Isolation and Genotyping

Three milliliters of peripheral blood sample was collected from the antecubital vein using evacuated ethylenediaminetetraacetic acid (EDTA) tubes (BD Vacutainer® EDTA tubes), followed by deoxyribonucleic acid (DNA) isolation using Wizard® Genomic DNA Isolation Kit (Promega, Madison, WI, USA). To detect the *ApaI, BsmI*, and *TaqI* polymorphisms of the VDR gene, polymerase chain reaction and restriction fragment length polymorphism (PCR-RFLP) methods were applied. Regions of the VDR gene flanking the studied SNPs were amplified by PCR, using a previously published primer[15,16] listed in Table 1. Genomic DNA was amplified using PCR. Initial denaturation was at 94°C for 4 minutes, followed by 30 cycles of denaturation at 94°C, annealing temperature at 60°C and 72°C for 1 minute each, and final extension at 72°C for 5 minutes. The PCR product was digested by ApaI, BsmI, and TaqI restriction enzymes (Thermo Scientific™, FastDigest FD1414, Mva12961, and FastDigest FD0674 Waltham, MA, USA) used to determine the genotypes of VDR gene polymorphisms. Digested samples were separated by 2% agarose gel electrophoresis, stained by red gel, and visualized using a gel documentation system.

Table 1 The Primer Sequences for *VDR ApaI, BsmI*, and *TaqI*

Single-Nucleotide Polymorphisms	Primers (5'–3')	Base Variation	Annealing Temperature (°C)
***ApaI* (rs7975232)**	F: 5'-CAG AGC ATG GAC AGG GAG CAA-3 R: 5'-GCA ACT CCT CAT GGC TGA GGT CTC-3'	T>G	60
***BsmI* (rs1544410)**	F: 5'-CAA CCA AGA CTA CAA GTA CCG CGT CAG TGA-3' R: 5'-AAC CAG CGG GAA GAG GTC AAG GG-3'	T>C	60
***TaqI* (rs731236)**	F: 5'-CAG AGC ATG GAC AGG GAG CAA-3 R: 5'-GCA ACT CCT CAT GGC TGA GGT CTC-3'	T>C	60

The amplified 740 base pairs (bp) PCR fragments was digested with *ApaI* restriction enzyme. Genotypes were identified as homozygous AA (one fragment of 740 bp), homozygous aa (2 fragments of 530 bp and 210 bp), and heterozygous Aa (3 fragments of 740 bp, 530 bp, and 210 bp).

Digestion of the 825 bp PCR fragment with *BsmI* restriction enzyme resulted in 3 different genotypes, namely homozygous BB (one fragment of 825 bp), homozygous bb (2 fragments of 650 bp and 175 bp), and heterozygous Bb (3 fragments of 825 bp, 650 bp, 175 bp).

Digestion of the 740 bp PCR fragment with *TaqI* restriction enzyme resulted in 3 genotypes, namely homozygous TT (2 fragments of 495 bp and 245 bp), homozygous tt (3 fragments of 290 bp, 245 bp, 205 bp), and heterozygous Tt (4 fragments of 495 bp, 290 bp, 245 bp, 205 bp).

Statistical Analysis

All data collected were statistically analyzed using the Statistical Package for Social Sciences (SPSS)® version 17. Categorical data were presented as proportion, while numerical variables were presented as mean ± standard deviation (SD) or (min-max). Differences in genotype distribution and allele frequencies in the case and control groups were statistically compared using Chi-square test. Test for deviations from Hardy-Weinberg Equilibrium (HWE) was performed using Chi-square distribution among controls for each SNP. Odds ratios (OR) with 95% confidence intervals (CI) were used to investigate the association between alleles, genotypes, and the risk of vitiligo in case and control groups. Statistical significance was established at $p < 0.05$.

Result

The demographic and clinical characteristics of the vitiligo patients are presented in Table 2. The mean age of the vitiligo patients was 31.09 ± 9.133 years old, while the mean duration of vitiligo, VASI score, and BSA was 5.97 ± 5.515, 0.97 ± 1.235, and 1.11 ± 1.375 years, respectively. The majority of vitiligo patients in this study were female with age of onset at ≤30 years old, had non-segmental type of vitiligo with emotional stress as precipitation factor, and had insufficient level of 25(OH)D. The majority ethnic group included in this study was Sundanese, followed by Javanese, Batak,

Table 2 Demographic and Clinical Characteristics of the Vitiligo Patients

Clinical Data	n=34
Age (years)	
Mean ± SD	31.09±9.133
Range	18.00–52.00
Gender	
Female	25 (73.5%)
Male	9 (26.5%)
Duration (years)	
Mean ± SD	5.97±5.515
Range	1.00–28.00
Age of onset	
>30-year-old	8 (23.5%)
≤30-year-old	26 (76.5%)
Type of vitiligo	
Segmental	4 (11.8%)
Nonsegmental	27 (79.4%)
Unclassified/indeterminate	3 (8.8%)
Family history of vitiligo	1 (2.9%)
Stability within the past year	8 (23.5%)
Leukotrichia	14 (41.2%)

(*Continued*)

Table 2 (Continued).

Clinical Data	n=34
Precipitation factor	
Emotional stress	20 (58.8%)
Emotional stress and mechanical trauma	5 (14.7%)
Sunburn	2 (6%)
Emotional stress and sunburn	2 (6%)
Mechanical trauma	1 (2.9%)
Chemical material	1 (2/9%)
Chemical material and sunburn	1 (2.9%)
Emotional stress, mechanical trauma, and sunburn	1 (2/9%)
Unknown	1 (2.9%)
VASI score	
Mean ± SD	0.97±1.235
Range	0.03–6.42
BSA	
Mean ± SD	1.11±1.375
Range	0.06–7.12
25-hydroxyvitamin D level	
Sufficient (30–100 ng/mL)	1 (2.9%)
Insufficient (10–<30 ng/mL)	21 (61.8%)
Deficient (<10 ng/mL)	12 (35,3%)

Abbreviations: SD, standard deviation; VASI, Vitiligo Area Score Index, BSA, body surface area.

Minangkabau, and Serawai. The allele and genotype frequencies of the *VDR* SNPs among cases and controls, as well as their associations with vitiligo susceptibility, are summarized in Table 3. In this study, the frequencies of genotype among the case group were 14, 20, 15, 19, and 34 for Aa, aa, Bb, bb, and TT, respectively. Among the control group, the frequencies of genotype were 6, 28, 17, 11, and 34 for Aa, aa, Bb, bb, and TT, respectively. The observed *ApaI* and *BsmI* genotype frequency distributions among the control group were consistent with HWE (p = 0.572 and 0.052, respectively). Meanwhile, the observed *TaqI* genotype frequencies deviated from HWE (p = not applicable). Therefore, no

Table 3 Allele and Genotype Frequencies of Selected *VDR* Polymorphisms Among Cases and Controls and the Association with Vitiligo Risk

	Vitiligo	Control	OR (95% CI)	p value
	n (%)	n (%)		
***Apal* (rs7975232)**				
A allele	14 (20.6%)	6 (8.8%)	2.679 (0.963–7.456)	0.053
a allele	54 (79.4%)	62 (91.2%)		
AA	0 (0.0%)	0 (0.0%)		
Aa	14 (41.2%)	6 (17.6%)	3.267 (1.071–9.965)	0.033*
aa	20 (58.8%)	28 (82.4%)		
***Bsml* (rs1544410)**				
B allele	15 (22.1%)	17 (25.0%)	0.849 (0.384–1.878)	0.686
b allele	53 (77.9%)	51 (75.0%)		
BB	0 (0.0%)	0 (0.0%)		
Bb	15 (44.1%)	17 (50.0%)	0.789 (0.304–2.050)	0.627
bb	19 (55.9%)	17 (50.0%)		

(*Continued*)

Table 3 (Continued).

	Vitiligo	Control	OR (95% CI)	p value
	n (%)	n (%)		
***TaqI* (rs731236)**				
T allele	68 (100.0%)	68 (100.0%)		N/A
t allele	0 (0.0%)	0 (0.0%)		
TT	34 (100.0%)	34 (100.0%)		N/A
Tt	0 (0.0%)	0 (0.0%)		
tt	0 (0.0%)	0 (0.0%)		

Notes: Chi-square test. *p < 0.05 is significant.
Abbreviations: CI, confidence interval; n, number; N/A, not assessed; OR, odds ratio.

further analysis for *TaqI* polymorphism was conducted in this study. This study showed that the Aa genotype was associated with a significantly-increased risk of developing vitiligo (odds ratio [OR] = 3.267, 95% confidence interval [CI] = 1.071–9.965; p = 0.033) compared to the aa genotype. However, there was no statistically significant difference between case and control groups in terms of allele frequency of *ApaI*, as well as both genotypes and allele frequencies of *BsmI* polymorphisms (p > 0.05). The representative gels for determining *ApaI, BsmI*, and *TaqI* genotypes are shown in Figure 1.

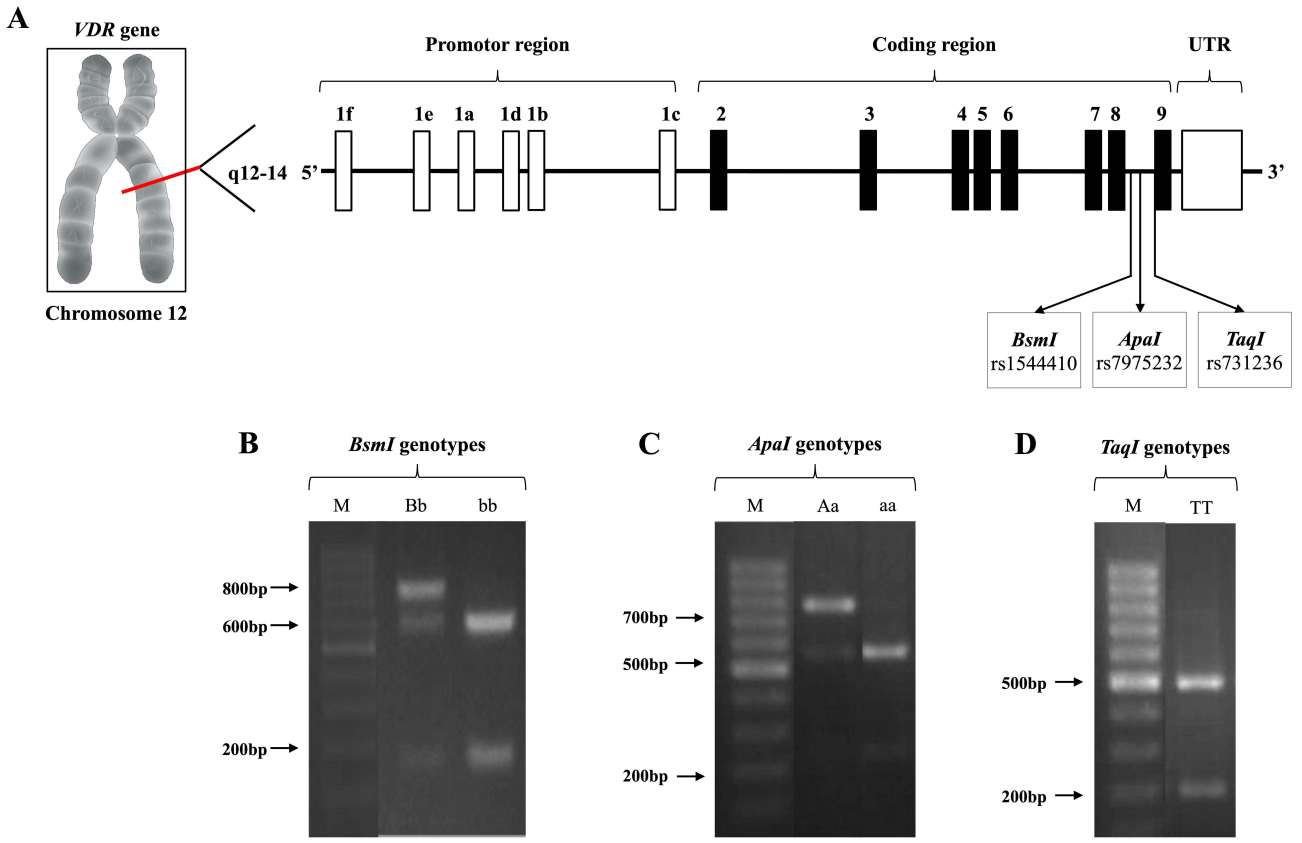

Figure 1 Structure of the genomic region of the *VDR* and the location of *BsmI* and *ApaI* (intron 8) and *TaqI* (exon 9) SNPs that are located near the 3' *untranslated region* (**A**). Representative gels for the determination of genotypes in cases are shown. Molecular weight DNA ladder (M) for size estimation of the DNA fragments. Digestion of 825 bp PCR product with *BsmI* restriction enzyme revealed heterozygous Bb (3 fragments of 825 bp, 650 bp, 175 bp) and homozygous bb (2 fragments of 650 bp and 175 bp) (**B**). Digestion of 740 bp PCR product with *ApaI* restriction enzyme revealed heterozygous Aa (3 fragments of 740 bp, 530 bp, 210 bp) and homozygous aa (2 fragments of 530 bp, 210 bp) (**C**). Digestion of 740 bp PCR with *TaqI* restriction enzyme revealed homozygous TT (2 fragments of 495 bp and 245 bp) (**D**).

Discussion

In vitiligo, despite playing a role in modulating T-cell activation as an autoimmune process, active vitamin D also protects epidermal melanin units and restores melanocytes, by way of controlling the activation, proliferation, and migration of melanocytes, and controlling pigmentation pathways.[17] Vitamin D exerts most of its physiological actions through its nuclear VDR on target cells.[2,11] VDR expression and function are thought to be influenced by *VDR* gene polymorphisms.[2]

The *VDR* gene is one of the best-studied genes in many autoimmune diseases.[9] The gene comprises five promoter regions and eleven exons, with exons 2–8 encoding proteins.[18] *ApaI* and *BsmI* polymorphisms are located in intron 8, while *TaqI* is in exon 9.[18] Polymorphisms in the intron do not change the protein's amino acid sequence,[19] but they can affect gene expression by disrupting the stability of messenger ribonucleic acid (mRNA),[18] altering intron regulatory elements, or influencing the splicing processes that control mRNA transcription.[2] Polymorphisms in this region are therefore silent genetic variants involved in the degradation of mRNA, further reducing receptor density.[2] *TaqI* polymorphism in the exon has a similar effect, ie, modification of splicing without changing the protein.[18,19] Due to the fact that nuclear VDR mediates most of the genomic effects on target cells, such as lymphocytes and melanocytes, *VDR* may represent a susceptibility gene for vitiligo.[2,10] However, the functional impact of *VDR* gene polymorphisms remains largely unknown.[2] Recent genetic studies have been conducted to determine the role of *VDR* gene polymorphisms in vitiligo, but the results varied. Some studies have linked *VDR* gene polymorphisms with risk of developing vitiligo, while conversely protecting against vitiligo in specific populations, indicating ethnicity as a potential source of heterogeneity.[2,9–14] Moreover, the heterogeneity of the study results may also be due to variations in the sample size, types of vitiligo, and genotyping methods.[6,20]

We conducted the first study investigating the association between the three frequently analyzed SNPs in the *VDR* gene and the risk of vitiligo in the Indonesian population. According to our results, the *ApaI* Aa genotype polymorphism is associated with a 3.267-fold increased risk of developing vitiligo. The result of this study was in accordance with a Pakistani study reporting that the *ApaI* Aa genotype polymorphism was associated with an increased risk of vitiligo (OR = 1.46; 95% CI = 1.01–2.13; p = 0.046).[11] A meta-analysis by Zhang et al[20] using data from six studies showed that *VDR ApaI* polymorphisms increased vitiligo susceptibility. Li et al[6] conducted a meta-analysis on the association of *ApaI* and *BsmI* polymorphisms with vitiligo risk from six independent case-control studies, and showed that East Asian populations who carry *ApaI*-a allele might be susceptible to vitiligo. They also concluded that the *ApaI* polymorphisms could be a potential biomarker for early detection of vitiligo.[6]

The current study also found that neither the allele nor the genotype of *BsmI* polymorphism had a significant association with vitiligo ($p > 0.05$). A similar study by Ochoa-Ramírez et al[14] in Mexico concluded that there was no association between *VDR BsmI* polymorphisms and vitiligo. A study conducted by Katsarou et al[10] in Southeastern European Caucasian population had a similar result. The study reported no association between *VDR BsmI* gene polymorphism and vitiligo.[10]

Limitations to this study might include the relatively small sample size for gene polymorphism study (34 cases and controls equally) and the fact that direct sequencing was not performed as a quality control for genotyping. This study also included data only from some Indonesian ethnicities (Sundanese, Javanese, Batak, Minangkabau, and Serawai), thus making the results apply only to these ethnicities. Therefore, further research with larger sample size with different Indonesian ethnicities is needed to validate our results and elucidate whether the same associations can also apply for vitiligo patients of differing ethnicities.

Conclusion

The *VDR ApaI* Aa genotype polymorphism may be considered a novel risk factor for vitiligo in Indonesian population. This finding strengthens the theory that *VDR* gene polymorphisms play a role in the etiopathogenesis of vitiligo.

Ethics Approval

This study complied with the Declaration of Helsinki, Good Clinical Practices, and local regulatory requirements. It was approved by the Medical Ethics Committee of Hasan Sadikin General Hospital Bandung (approval number: LB.02.01/X.6.5/100/2021).

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Disclosure

The authors declare no conflicts of interest.

References

1. Colucci R, Lotti T, Moretti S. Vitiligo: an update on current pharmacotherapy and future directions. *Expert Opin Pharmacother*. 2012;13(13):1885–1899.
2. Aydingöz İE, Bingűl İ, Doğru-Abbasoğlu S, Vural P, Uysal M. Analysis of vitamin D receptor gene polymorphisms in vitiligo. *Dermatology*. 2012;224(4):361–368. doi:10.1159/000339340
3. Kaur I, Singal A. Definition, epidemiology, and prevalence. In: Relhan V, Garg VK, Ghunawat S, Mahajat K, editors. *Comprehensive Textbook on Vitiligo*. 1st ed. London: CRC Press; 2020:7–8.
4. Bregvist C, Ezzedine K. Vitiligo: a review. *Dermatology*. 2020;236(6):571–592. doi:10.1159/000506103
5. Allam M, Riad H. Concise review of recent studies in vitiligo. *Qatar Med J*. 2013;2013(2):1–19. doi:10.5339/qmj.2013.10
6. Li L, Wu Y, Li L, et al. Association of ApaI and BsmI polymorphisms with vitiligo risk: a meta-analysis. *Clin Exp Dermatol*. 2015;40(7):794–803. doi:10.1111/ced.12633
7. Didona D, Didona B, Paolino G, Caro R. Etiopathogenesis. In: Relhan V, Garg VK, Ghunawat S, Mahajat K, editors. *Comprehensive Textbook on Vitiligo*. 1st ed. London: CRC Press; 2020:21–28.
8. Shen C, Gao J, Sheng Y, et al. Genetic susceptibility to vitiligo: GWAS approaches for identifying vitiligo susceptibility genes and loci. *Front Genet*. 2016;7:3–15. doi:10.3389/fgene.2016.00003
9. Li K, Shi Q, Yang L, et al. The association of vitamin D receptor gene polymorphisms and serum 25-hydroxyvitamin D levels with generalized vitiligo. *Br J Dermatol*. 2012;167(4):815–821. doi:10.1111/j.1365-2133.2012.11132.x
10. Katsarou MS, Sidiropoulou P, Leronymaki D, et al. Impact of vitamin D receptor gene polymorphisms on vitiligo susceptibility and clinical features in Southeastern European Caucasian population. *Int J Mol Med*. 2020;46(5):1899–1907. doi:10.3892/ijmm.2020.4732
11. Sajjad S, Munir S, Ber-Rahman S, Saba N, Rehman S. VDR polymorphisms *ApaI* (rs7975232), *TaqI* (rs731236), and *FokI* (rs2228570) in Pakistani vitiligo patients and controls. *J Pak Assoc Dermatol*. 2017;27(2):102–109.
12. Sobeih S, Mashaly HM, Gawdat H, Amr K, Hamid MF, Shaalan E. Evaluation of the correlation between serum levels of vitamin D and vitamin D receptor gene polymorphisms in an Egyptian population. *Int J Dermatol*. 2016;55(12):1329–1335. doi:10.1111/ijd.13363
13. Birlea SA, Fain PR, Spritz RA. A Romanian population isolate with high frequency of vitiligo and associated autoimmune diseases. *Arch Dermatol*. 2008;144(3):310–316. doi:10.1001/archderm.144.3.310
14. Ochoa-Ramírez LA, Díaz-Camacho SP, Becera-Loaiza DS, et al. Catalase but not *vitamin D receptor* gene polymorphisms are associated with nonsegmental vitiligo in Northwestern Mexicans. *Int J Dermatol*. 2019;58(11):1264–1269. doi:10.1111/ijd.14508
15. Pani MA, Knapp M, Donner H, et al. Vitamin D receptor allele combinations influence genetic susceptibility to type I diabetes in Germans. *Diabetes*. 2000;49(3):504–507. doi:10.2337/diabetes.49.3.504
16. Colombini A, Brayd-Bruno M, Lombardi G, et al. BsmI, ApaI, and TaqI polymorphisms in the vitamin D receptor gene (VDR) and association with lumbar spine pathologies: an Italian case-control study. *PLoS One*. 2016;11(5):e0155004. doi:10.1371/journal.pone.0155004
17. Polat M, Uzun O. Topical vitamin D analogs. In: Relhan V, Garg VK, Ghunawat S, Mahajan K, editors. *Comprehensive Textbook on Vitiligo*. 1st ed. London: CRC Press; 2020:119–122.
18. Khan MI, Bielecka ZF, Najm MZ, et al. Vitamin D receptor gene polymorphisms in breast and renal cancer: current state and future approaches (review). *Int J Oncol*. 2014;44(2):349–363. doi:10.3892/ijo.2013.2204
19. Triantos C, Aggeletopoulou I, Kalafateli M, et al. Prognostic significance of vitamin D receptor (VDR) gene polymorphisms in liver cirrhosis. *Sci Rep*. 2018;8(1):14065. doi:10.1038/s41598-018-32482-3
20. Zhang JZ, Wang M, Ding Y, et al. Vitamin D receptor gene polymorphism, serum 25-hydroxyvitamin D levels, and risk of vitiligo A meta-analysis. *Medicine*. 2018;97(29):1–8.