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Chapter 1

Genetic and Biochemical Factors Related to the Risk and Disability Progression in Multiple Sclerosis

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Abstract

Sclerosis multiplex (multiple sclerosis, MS) is a chronic autoimmune inflammatory disease of the central nervous system. The immune regulatory defects lead to the process of inflammation and neurodegeneration that results in the deterioration of neurological functions. It is still unclear as to why MS is so devastating and rapidly progressive in one patient and less so in another. It is known that the etiopathogenesis of MS is very complex, and many factors can be involved in the risk and character of the disease and its progression. In this chapter, we discuss the general molecular and cellular mechanisms of action of genetic and biochemical factors that are related to immune system regulation and thus can be connected to the individually varying risk and disability progression of MS. We found that gene variants of the gene polymorphism rs6897932 in interleukin 7 receptor α chain gene rs10735810 in vitamin D receptor gene and HLA-DR and HLA-DQ genes as well as the serum level of vitamin D are associated with MS risk or disability progression in Central European Slovak population.

Keywords: multiple sclerosis, risk, disability progression, gene polymorphism, biochemical marker

1. Introduction

In triggering an autoimmune response in multiple sclerosis (MS), environmental factors have a strong effect and interact with complex risk-conferring genetic variants [1–3]. In this process, the myelin reactive T cells with altered functional characteristics are formed and...
activated [4]. The immune regulatory defects and increased migration of autoreactive lymphocytes within the brain, that are the typical traits in MS, lead to the process of inflammation, myelin sheath breakdown, demyelination, remyelination, neuronal and axonal degeneration, and subsequent deterioration of neurological functions [5]. Neurodegeneration, neuronal and axonal damage that correlate with the progression of the disease can be a process partly independent from inflammation and demyelination or even can be the cause of demyelination occurring from the disease onset. Axonal damage in MS is a result of many pathological processes [6, 7].

It is still unclear as to why MS is so devastating and rapidly progressive in one patient and less so in another. Because the etiopathogenesis of MS is very complex, disease development as well as the characteristics of disease progression is probably the consequence of multifactorial interaction. Our work is dedicated to genetic and biochemical markers that were chosen according to their possible role in the modulation of the immune response in MS patients and thus could be associated with MS risk and disability progression. In our work, we discuss the immune response-related genetic factors associated with MS that can be generally classified into HLA genes and non-HLA genes. Since vitamin D can have an important role in the pathogenesis of MS, great part of our work is dedicated to its metabolism, functions, mechanisms of action in MS and genetic factors that can modify these effects. In this work, we also present the results of our own analysis of genetic and biochemical markers that we found to be associated with MS risk or progression in the group consisting of MS patients with clinically diagnosed MS and healthy individuals from the region of Central Slovakia. To evaluate the disease progression rate, we used the widely accepted multiple sclerosis severity score (MSSS, score range 0.01–9.99) [8] that considers the neurological impairment of the functional systems (expanded disability status scale score) [9] together with disease duration. For the purpose of the association analysis of these markers with the rate of disease disability progression, we stratified MS patients by MSSS scores to three groups—slowly progressing MS (MSSS < 3), mid-rate progressing MS (MSSS 3–6) and rapidly progressing MS (MSSS > 6) [10].

2. Immune response-related genetic factors in the risk and progression of multiple sclerosis

MS is a typical gender-dependent disease; a higher risk of MS is observed in women than in men in all populations and races. A study conducted in Canada found female to male ratio in individuals affected by MS to be 3:1 [11]. The risk of MS development in siblings of an affected individual is estimated to be 5%, in children 2%, in monozygotic twins 25% [5]. However, it has been shown that genetic predisposition is not strong enough to induce disease development, and appropriate environmental triggers are necessary to start the disease process [1, 12]. In general, the MS-associated genes can be classified into genes of the HLA-complex and non-HLA genes [3].
3. Gene polymorphisms and haplotypes in the aetiology of multiple sclerosis

The single-nucleotide polymorphism (SNP) is a variation of a single nucleotide, which is present in the population in a frequency higher than 0.01. SNPs are the most common type of genetic variation and are usually caused by somatic or gametic mutations. Nucleotide change can cause the formation or loss of restriction sites for bacterial endonucleases that are able to cleave the specific DNA sequence. The identification of gene polymorphisms that are in correlation with other risk factors, including biochemical markers, can be useful in establishing the risk of MS development, prognosis, clinical course of the disease and response to therapy. Haplotype (haploid genotype) is a certain combination of alleles or SNPs in the sequence of the DNA, which is localized on one chromosome and is inherited together. When two alleles are in linkage disequilibrium, they are inherited together in a higher frequency than expected randomly [13]. The combination of more alleles, known as tagging SNPs, enables us to identify the other associated alleles. For example, the allele A of gene polymorphism rs3135388 corresponds to the incidence of allele HLA-DRB1*1501, which is the most common genetic risk factor for MS development [14–16].

4. HLA genes and MS

Antigen expression that is inducible by cytokines is different on the various immune cells. Major histocompatibility complex II (MHCII) antigens are transmembrane proteins localized on the immune cells, thus having an important role in the process of exogenous antigen presentation to T cells. MHCII molecules are coded by the gene of human leucocyte antigens D (HLA-D gene) that is localized on chromosome 6 and has three regions—HLA-DP, HLA-DQ and HLA-DR [17]. The susceptibility of the population to autoimmune diseases depends on the individual ability to express HLA-DQ and HLA-DR antigens. This expression can be induced by virus infection, most likely by EBV, influenza or paramyxovirus [18]. MHCII gene expression is regulated by vitamin D through its binding to the vitamin D–responsive elements (VDREs) that are localized in the promoter region of HLA-DRB1 gene. This fact can explain the interaction between vitamin D, that is an important factor modifying MS development and disease course, and genetic predisposition to MS represented mainly by a highly conservative allele HLA-DRB1*1501. HLA-DRB1*1501 allele is in general considered to be the most important susceptibility allele of MS [19–24]. This allele was found to be present in over 50% of MS cases [25, 26]. The increased frequencies of the DRB1*15 allele in MS patients have been described in Northern Europeans [23, 27], South and North Americans [20, 28], Mediterraneans [29, 30] and African Americans [21]. In Spanish cohorts, the DRB1*03 was the second most frequent allele associated with MS, but only after eliminating HLA-DRB1*15 [29]. The DRB1*03 allele has also been found to be significantly associated with the increased risk of MS in Scandinavians [27], Sardinians [31], and Australians [32]. Fernández et al. [33] found that the DRB1*13 allele is protective against MS development in Spaniards. A protective effect of the alleles DRB1*01, DRB1*07, DRB1*12 and DRB1*14 was confirmed in the recent meta-
analysis in Caucasians [24]. The allele DRB1*07 was found to be protective against MS also in Scandinavians [34]. The DRB1*13.03 allele was found to be the primary risk allele in MS patients of European descent [23]. The protective effect against MS has also been shown for the HLA-DRB1*11 allele [24, 29].

The DQB1*06:02 allele was found to be linked to the increased risk of MS with a proved tight linkage disequilibrium between DRB1*15 and DQB1*06 in Caucasians [35]. As the risk factor of MS, DQB1*06:02 allele has also been identified in a cohort of Afro-Brazilians [36] and Spaniards [33]. Kaushansky et al. [37] suggested that the role of the DRB1*15:01 and DQB1*06:02 alleles in MS depends on the heterogeneous interaction of target antigen, genotype, and phenotype. On the contrary, Isobe et al. [38] found none of the HLA-DQB1 alleles to be associated with MS in African Americans. According to the combinations of HLA-alleles, the association of HLA-DRB1*15/*15 genotype with MS was identified by several studies [32, 34, 39]. In multi-case MS families, Barcellos et al. [39] identified a high risk DRB1*15/*08 genotype and protective DRB1*15/*14 genotype. The study of Sawcer et al. [23] indicates that in all populations of North-European ancestry, a predisposition to MS is linked with the DRB1*15:01-DQB1*06:02 haplotype. Furthermore, Link et al. [34] in a Scandinavian cohort showed that risk haplotypes for MS are almost all DRB1*15 bearing haplotypes, while protective effect against MS development are HLA class I A*02 allele-bearing haplotypes. In Sardinian MS patients, Cocco et al. [40] confirmed a positive association of the haplotype HLA DRB1*03:01-DQB1*02:01 with MS.

In the study from our laboratory, we analysed the association of the HLA-DRB1/DQB1 genes, alleles and their combinations with susceptibility to MS in the population from central Slovakia. We found that the increased risk of MS is in individuals carrying alleles HLA-DRB1*15, DRB1*03 and DQB1*06, genotypes HLA-DRB1*15/*15 DQB1*06/*06 and haplotype DRB1*15-DQB1*06. In addition, we also found that HLA-DRB1/DQB1 class II alleles DRB1*07, DRB1*13, DQB1*03, genotypes DRB1*13/*11, DQB1*05/*03 and haplotypes DRB1*13-DQB1*06 and DRB1*11-DQB1*03 are associated with the protection against MS development. We cannot exclude that the proposed protective effects of the DRB1*11-DQB1*03 and DRB1*13-DQB1*06 haplotypes in our cohort could be, at least partially, due to the linkededisequilibrium with alleles in the HLA class I region which is primarily associated with MS [41].

5. Non-HLA genes and MS

Gene products of non-HLA genes can contribute to the genetic risk of MS by modulation of different processes. These genes are involved in the regulation of functions of T- and Bcells, dendritic cells, NKcells, cytokine signalization, metabolism of interferons, vitamin D metabolism, neuronal regeneration and many others [3]. It has been found that these genes can contribute not only to the increased inherited risk of MS development but also to the risk of other autoimmune diseases [42, 43]. The examples of the SNPs involved in the etiopathogenesis of MS are summarised in Table 1.
| Gene                  | Function                                      | Localization | SNP            | Alleles      |
|-----------------------|-----------------------------------------------|--------------|----------------|--------------|
| HLA-DRB1*1501         | Antigen presentation                          | 6p21         | rs3135388      | C/T          |
|                       |                                               |              | rs3135005      | C/T          |
| IL-7Ra                | Proliferation of memory T cells, T- and B-cell development | 5p13         | rs6897932      | C/T          |
| IL-2Ra                | Sensitization of T cells to IL-2, proliferation of T cells | 10p15        | rs2104286      | A/G          |
|                       |                                               |              | rs12722489     | A/G          |
|                       |                                               |              | rs11256369     | C/G          |
|                       |                                               |              | rs706103       | C/T          |
| CD58                  | Function of regulatory T cells                | 1p13         | rs2300747      | A/G          |
|                       |                                               |              | rs6677309      | A/C          |
|                       |                                               |              | rs12044852     | A/C          |
| CD6                   | Regulation, adhesion of T, B, and other T cells | 11q13        | rs17824933     | C/G          |
|                       |                                               |              | rs12288280     | G/T          |
| CLEC16A               | Cell receptor, induction of immune response   | 16p13        | rs6498169      | A/G          |
|                       |                                               |              | rs12708716     | A/G          |
| VAV1                  | Lymphocyte survival, differentiation and proliferation | 19p13        | rs2546133      | C/T          |
|                       |                                               |              | rs2617822      | A/G          |
| PRKCA                 | Regulation of IL-2 expression, receptor       | 17q22-q23    | Allele variants in introns 3, 8 |             |
| EVI 5 (ectopic viral integration site 5 protein homolog) | Nuclear protein, cell cycle regulation | 1p22        | rs66080578     | A/T          |
|                       |                                               |              | rs11808092     | A/C          |
|                       |                                               |              | rs11804321     | C/T          |
| IRF5                  | Regulation of cytokine activation             | 7q32         | rs4728142      | A/G          |
|                       |                                               |              | rs3807306      | A/C          |
| IRF8                  | Expression of interferon response genes, development of B cells | 16q24        | rs17445836     | A/G          |
| TYK2                  | Gene expression                               | 19p13        | rs34536443     | C/G          |
|                       |                                               |              | rs5562744      | C/T          |
| TNFRSF1A (tumour necrosis factor receptor superfamily member 1A) | Receptor for tumour necrosis factor 12p13 | 18q22        | rs1800693      | A/G          |
|                       |                                               |              | rs419584       | A/C/G        |
|                       |                                               |              | rs576455       | C/T          |
|                       |                                               |              | rs419577       | C/T          |
| CD40                  | Receptor for tumour necrosis factor           | 20q12-q13    | rs1883832      | C/T          |
| CD226                 | Activator of NK cells, lymphocyte adhesion, co-stimulator of T cells | 18q22        | rs763361       | C/T          |
| KIF1B (kinesin family member 1B) kinesin family member 1B) | Neuronal regeneration | 1p36        | rs10492972     | C/T          |
| GPC 5 (glypican 5)     | Neuronal growth and repairation               | 13q32        | rs9523762      | A/G          |

**Table 1.** Gene polymorphisms involved in the etiopathogenesis of MS [3, 44–48].
6. Genetic variants in interleukin 7 receptor α chain (IL-7Ra) gene

IL7 is a type 1 short-chain cytokine of the haematopoietin family involved in the modulation of T- and B-cell development and T-cell homeostasis. To perform the immune system functions, IL7 binds to the transmembrane receptor that is formed by heterodimerising the common cytokine gamma chain and IL7 receptor alpha chain (IL7Ra or CD127). IL7Ra is a membrane glycoprotein folded to bind and mediate the action of IL7 and other alpha helical cytokines. IL7Ra consists of an extracellular domain, transmembrane region and cytoplasmic tail, which uses kinases for signal transduction [49]. The localization of the IL7Ra gene is chromosome 5p13.3. An increased expression of IL7Ra in peripheral blood mononuclear cells was found in MS patients when compared to controls [50, 51]. The IL7Ra and IL7 mRNA increased expression was found also in the cerebrospinal fluid of MS patients, possibly suggesting an altered balance between the isoforms of IL7Ra and a higher signal-inducing immune cell proliferation and survival [52]. According to the alternative splicing of exon 6 in IL7Ra gene, membrane-bound or soluble isoforms of IL-7Ra are produced [53]. A significantly increased ratio of the membrane-bound to soluble isoforms of IL7Ra in MS patients can facilitate the aberrant activation of potentially auto-reactive T cells [54].

Single-nucleotide polymorphisms in IL7Ra gene are involved in the dysregulation of immune homeostasis and thus can be associated with susceptibility to MS [55]. A genome-wide study in a large group of subjects from the UK and USA identified the strong association between SNP rs6897932 in IL7Ra gene and the risk of MS [2]. The non-conservative amino acid change on position 244 (Ile→Thr) of IL7Ra is a result of the SNP rs6897932 (ATC → ACC) in exon 6 of IL7Ra gene [56]. This amino acid change has a functional effect on the product of expression of alternative spliced IL7Ra gene, which is manifested by changes in the proportion of the soluble versus membrane-bound isoforms of IL7Ra. The change of this ratio can be followed by a different regulation of the IL7 signal transduction pathway and directly associates the SNP rs6897932 with MS [57].

It has been found that allele C of SNP rs6897932 in IL7Ra gene contributes to the increased genetic risk of MS in groups of MS patients from the USA [57, 58], South Spain [59], Nordic countries—Denmark, Finland, Norway and Sweden [52], France [60], Netherlands [61] and Japan [62]. The homozygosity for C allele was identified as a risk genotype for MS susceptibility in Netherlands [61] and Spain [59]. A genotype association was also confirmed by the finding of increased counts of CC genotype of rs6897932 in MS patients compared to controls in the cohorts from the USA [58] and Japan [62]. Corresponding with the contribution of allele C to the risk of MS, the protective effect of allele T for MS risk in a Nordic case-control group has been reported by Lundmark et al. [52]. The protective effect of allele T has been reported also in Spain by Alcina et al. [59]. On the contrary, no association between SNP rs6897932 and MS was found in cohorts of MS patients from Northern Ireland [58], Germany [44] and Western Balkan countries—Serbia, Croatia and Slovenia [63].

Only a few studies addressed the question whether the rs6897932 in IL7Ra gene contributes only to the genetic risk of MS, or whether it can also affect the disease course and disability progression [44, 57, 61, 64]. Groups of patients with different forms of MS were compared in
several studies. In Northern European primary-progressive MS cases (PP MS), the underexpression of \(\text{IL7Ra}\) gene as well as different allele frequencies of \(\text{IL7Ra}\) promoter SNP was confirmed. Moreover, \(\text{IL7Ra}\) gene expression was found to be up-regulated in secondary-progressive MS (SP MS) patients [64]. In a study by Akkad et al. [44] in German MS patients, it was found that the soluble \(\text{IL7Ra}\) reduced the expression and allelic and genotypic association between rs6897932 and SP or PP MS but not with RR MS. In their study, a significantly higher frequency of allele C and genotype CC of rs6897932 in SP and PP MS patients was found, but not in RR MS patients compared to controls. The assessment of the severity of MS by MSSS did not show any association between rs6897932 genotype and disease severity in the USA [57]. Differences in allele frequencies in SP MS patients compared to healthy controls were reported in Dutch MS patients by Sombekke et al. [61]. In spite of that, no association between rs6897932 genotype and disease severity (MSSS, EDSS, other clinical tests) and disease activity (relapse rate and MRI markers) was found.

Results of our own work suggest the relevance of rs6897932 allele and gene variants in MS pathogenesis in Slovaks [10]. Our results have revealed that allele C is present in a higher frequency in MS patients (77.4%) as compared to the control group (72.3%), which indicates an increased risk of MS development (OR=1.314, 95% CI=1.004–1.720, \(p=0.047\)). Interestingly, allele T was manifested in MS patients in a significantly lower frequency representing only 22.6% as compared to 27.7% in controls. This suggests that allele T seems to be protective against MS development (OR = 0.761, 95% CI= 0.582–0.996, \(p = 0.047\)). The additive model fitted the best to assess association between genotypes and MS risk. Logistic regression analysis adjusted for sex and age revealed that there is a significant association between \(\text{IL7Ra}\) rs6897932 genotype and MS risk (OR = 0.764, 95% CI = 0.586–0.995, \(p_{\log} = 0.045\)). The genotype analysis showed that MS patients manifested a lower frequency of genotype CT when compared to controls (34.8% vs. 36.3) and genotype TT (5.2% vs. 9.9%) and a higher frequency of genotype CC (60.0% vs. 54.1%). When we used the additive genetic model, we found a significantly decreased risk of MS development in carriers of allele T with genotype CT (OR = 0.865, 95% CI = 0.609–1.228, \(p = 0.05\)) as well as with genotype TT (OR = 0.565, 95% CI = 0.282–1.132, \(p = 0.05\)).

After stratification of MS patients according to the disease disability progression rate, we found a significantly lower frequency of allele T in the subgroup of rapidly progressing MS patients (18.1%) as compared to 27.7% in controls. These results suggest that allele T is associated with protection against rapid disability progression of MS (OR = 0.576, 95% CI = 0.348–0.955, \(p = 0.031\)). An additive genetic model adjusted for sex and age fitted the best to assess the association between genotypes and the rate of disease disability progression. Linear logistic regression with disease disability rate as the dependent variable—MSSS (1, 2, 3 and 0 for controls) revealed that there is a significant association between \(\text{IL7Ra}\) rs6897932 genotype and disability progression of MS (\(p_{\log} = 0.034\)). Genotype analysis showed that the frequency of genotype TT is higher in controls (9.6%) and lower in MS patients with rapid disability progression (3.5%). Frequency of genotype CC was higher in rapidly progressing MS patients (67.2%) and lower in controls (54.1%). The data suggest that individuals carrying genotype TT are protected against rapid disease disability progression of MS.
We have shown for the first time in a Central European Slovak population that allele C of rs6897932 is associated with the risk of MS, and allele T has a protective additive effect against MS susceptibility. Moreover, we revealed that minor allele T and genotype TT of rs6897932 in the IL7Ra gene are protective against rapid disease disability progression in MS [65].

7. Vitamin D and its role in risk and progression of multiple sclerosis

In the past decades, much attention has been given to vitamin D and its role in MS and other autoimmune diseases. The following sections are dedicated to the metabolism and structure of vitamin D, its immunological effects, serum level and mechanisms of action of vitamin D in the prevention and treatment of MS. We also describe the genetic factors that can modulate the biological effects of vitamin D.

7.1. The structure and metabolism of vitamin D

Vitamin D in the human body undergoes a complex metabolism. Cholecalciferol (vitamin D₃), as a precursor of a hormonally active form, is produced in the skin from 7-dehydrocholesterol after sunlight exposure and can also be absorbed from the diet. Subsequently, cholecalciferol is hydroxylated in the liver forming 25-hydroxycholecalciferol, calcidiol. The hormonally active form of vitamin D, 1,25- dihydroxycholecalciferol, calcitriol, is produced by further hydroxylation especially in the kidneys and also in other tissues. The enzyme catalysing this hydroxylation is 25-hydroxyvitamin D-1α-hydroxylase, coded by CYP27B1 (cytochrome P450 family 27 subfamily B member 1) gene [66, 67]. In various cells, the bioactive form of vitamin D binds to the vitamin D receptor (VDR) providing its physiological functions by modulation of the target gene’s transcription [68]. The circulating serum level of vitamin D depends not only on environmental factors such as exposition to sunlight and vitamin D intake but also on genetic and epigenetic factors. The genetic factors can influence the effects of vitamin D through the variability of the genes participating in its activation and degradation, transport and receptor signalling [69].

7.2. The effect of vitamin D on the functions of immune cells

There is growing evidence that vitamin D not only regulates bone metabolism but also has large-scale immunomodulatory and anti-inflammatory effects. A linkage has been found between vitamin D deficiency and increased risk of autoimmune diseases [70]. The immunocompetent cells—macrophages, dendritic cells, Tcells and Bcells are able to produce calcitriol and express the VDR at the high rate. Through this, vitamin D modulates the synthesis of various cytokines and immunoglobulins and is involved in the regulation of innate and adaptive immune response. Autocrine and paracrine effects of vitamin D depend also on its serum level, and individuals with hypovitaminosis D are in a state of immune system dysfunction and are predisposed to the development of autoimmune diseases [71].

In Tcells, calcitriol inhibits the production of IL-12 and IFN-γ and subsequent differentiation of Th1 lymphocytes that are the key cells involved in the MS development. Calcitriol improves
the immunosuppressive functions of T<sub>REG</sub> cells and ameliorates the T<sub>H2</sub>-cell development by the activation of the promoter region of IL-4 gene [19, 72]. Vitamin D increases the expression of IL-4, IL-5 and IL-10 that are able to activate T<sub>H2</sub> cells, decreases the production of IFN-γ, blocks the formation of T<sub>H1</sub> cells after antigen stimulation and has positive effects on the T<sub>H1</sub>-mediated autoimmune diseases [73, 74]. B cells, which also participate in the demyelinating process and produce intrathecal immunoglobulins, express VDR and vitamin D hydroxylases. In B cells, calcitriol reduces the intracellular signal pathways of nuclear transcription factor NF-kappa B (NF-kB) and CD40 signalling [75]. Calcitriol inhibits the maturation and proliferation of B cells, induces apoptosis of B cells, inhibits the differentiation of plasma and memory cells and decreases the production of immunoglobulins IgG and IgM [76]. Immature B cells are more prone to regulation by calcitriol when compared to plasma cells. Calcitriol also decreases the expression of MHCII molecules and co-stimulatory molecules in B cells [19].

Calcitriol formed in macrophages inhibits the immune response by suppressing proliferation of T<sub>H1</sub>- and T<sub>H17</sub>-cells and promoting the functions of T<sub>H2</sub>- and T<sub>REG</sub> cells [71]. Calcitriol inhibits the secretion of IL-12 by antigen-presenting cells and monocytes [77]. Vitamin D blocks the differentiation of immature dendritic cells and the expression of co-stimulatory molecules CD40, CD80, CD86 and MHC II, thus decreasing the capacity of dendritic cells to activate autoreactive T cells. Vitamin D also ameliorates the spontaneous apoptosis of mature dendritic cells [73]. In macrophages, calcitriol suppresses intracellular oxidative burst and listericidal activity. It also suppresses the expression of Fc and TLR receptors induced by IFN-γ that are important for antigen recognition [78]. Vitamin D suppresses the proliferation of antigen-specific T cells and chemotaxis of dendritic cells by decreasing the expression of differentiation antigens CD80, CD86 and HLA-DR molecules [79].

7.3. The murine model of MS and vitamin D

In mice that lack the VDR gene or the gene of the enzyme catalysing vitamin D activation, an abnormal development and function of T<sub>H1</sub>-lymphocytes and deficiency of peripheral T-lymphocytes have been observed [80, 81]. Calcitriol treatment can prevent the induction and progression of autoimmune diseases including experimental autoimmune encephalomyelitis (EAE), a murine model of MS [82, 83]. Calcitriol can also decrease the severity of EAE symptoms, and its deficiency causes an increased susceptibility of animals to EAE induction [77, 82]. In mice with chronic EAE, vitamin D administration suppresses the proliferation of specific T<sub>H1</sub> cells, inhibits IL-12 dependent production of IFN-γ, prevents relapses and reduces perivascular infiltration, demyelination plaque formation and axonal degeneration in the brain and spinal cord [84].

7.4. Serum level of vitamin D and dose management

To reflect vitamin D status in the human body, calcidiol plasma level measurement is usually used. Calcidiol is the main circulating form of vitamin D in plasma, and its biological half-time is 19 days [85]. The recommended daily dose of vitamin D is approximately 10 times higher than in the past. The optimal serum level of vitamin D is 75–250 nmol/l (30–100 ng/ml). In
countries with less sunny climate, the necessary daily dose of vitamin D is 1000–4000 IU/day (1 μg = 40 IU) [86].

The risk of vitamin D overdosing is hypercalcaemia and subsequent organ and tissue damage. Whole body exposure to sunlight results in the production of around 10,000 IU of vitamin D, so it is not simple to cause vitamin D intoxication by its short-term peroral supplementation. The results of several studies suggest that even high-dose vitamin D₃ supplementation in MS patients is safe and clinically useful. Burton et al. [87] administered high peroral doses of vitamin D to healthy individuals and MS patients. The initial dose was 40,000 IU/day during 28 weeks, followed by 10,000 IU/day during 12 weeks; later it was gradually decreased to 0 IU/day, combined with 1.2 grams of calcium per day. During the period of 40,000 IU of vitamin D per day, the serum levels reached 413 nmol/l, which was higher than the conventional limit established for vitamin D toxicity (250 nmol/l). Calcidiol serum levels remained around this limit for 18 weeks without any observed negative effects. The serum level of calcium was in the physiological reference range during the whole study duration. Moreover, no cardiac rhythm abnormalities or impairment of hepatic or renal functions was observed. Kimball et al. [88] administered 4000–40,000 IU/day to patients in the active phase of MS together with 1.2 grams of calcium. Medium serum level of calcidiol was 78 ± 35 nmol/l and rose to 386 ± 157 nmol/l. Serum calcium level and urinary calcium to creatinine ratio did not exceed the physiological reference values. Vitamin D supplementation in this study did not cause any change in the serum level of hepatic enzymes, creatinine, electrolytes, proteins and parathormone. Although the serum level of calcidiol doubled the physiological upper range value, hypercalcaemia or hypercalciuria was not observed.

Although the significant toxicity of vitamin D₃ was not observed even in doses of 40,000 IU/day, its daily dose in healthy individuals should not exceed 2000 IU. The optimal daily dose of vitamin D₃ that should be routinely recommended to women during pregnancy and lactation is 1000 IU. Children born in families with MS history should be administered daily 200–400 IU of vitamin D₃ [66].

7.5. Vitamin D and the course, prevention and treatment of MS

The role of vitamin D in the prevention of MS development has been confirmed by many experimental, epidemiological, genetic and immunological studies. Vitamin D insufficiency during the whitematter development can alter the pathways of axonal differentiation and adhesion and increase the apoptosis of oligodendrocytes that express VDR. This results in local microenvironmental changes and altered regenerative and remyelinating capacity [66]. In individuals with an increased genetic risk of MS, it is possible to prevent the demyelination process by preventive vitamin D administration. This preventive strategy would be better than reparation of already developed myelin and axonal damage [12, 66].

High-dose peroral vitamin D intake has been found to be inversely associated with the risk of MS in a cohort of more than 90,000 women. Peroral vitamin D supplementation in a dose higher than 400 IU/day leads to the reduction of MS risk when compared to the individuals with no vitamin D intake (RR = 0.59, 95% CI = 0.38–0.91, p = 0.006) [89]. Also, calcidiol plasma levels are
inversely correlated with MS risk. This association is particularly obvious in whites, while among blacks and Hispanics with lower 25-hydroxyvitamin D levels than whites, there was no significant association between vitamin D and MS risk [90]. Vitamin D also has reparative effects for the nervous tissue, especially in patients in the early phases of the disease. In countries with low sun exposure, food supplementation of vitamin D could be a simple and cheap method of MS prevention [86]. The incidence of MS could be reduced by the administration of vitamin D to pregnant women, and all children living in mild climates should be more exposed to sunlight and should be on a vitamin D–rich diet [66].

Vitamin D is not only a factor modifying MS risk, but it can also have a role in the modulation of disease course. It has been observed that in relapsing remitting MS, calcidiol plasma levels are lower during relapses compared to the periods of remission [91]. In addition, there is evidence that lower calcidiol levels are associated with higher relapse rates and higher risk of exacerbation, as well as higher expanded disability status scale (EDSS) scores and progressive forms of MS [92–94]. Vitamin D can improve memory and cognitive impairments in patients with MS, Alzheimer disease and in patients after chemotherapeutical treatment [95]. High-dose peroral vitamin D supplementation has immunomodulatory effects and leads to reduction in the number of relapses and suppression of the inflammatory activity and proliferation of T cells [87], as well as the decrease in the number of gadolinium-enhancing lesions in brain [88].

In our study, we examined the serum levels of calcidiol in a group of MS patients from the Central-Northern part of Slovakia. We found that hypovitaminosis D is more frequent in MS patients, when compared to healthy individuals. Serum levels of calcidiol were significantly lower in MS patients when compared to controls (15.0 ± 6.1 ng/ml vs. 18.2 ± 8.3 ng/ml, \( p_{(K-W)} = 0.001 \)). Moreover, we found that there is an association of the serum level of vitamin D with the rate of MS disability progression (\( p(K-W) = 0.000 \)). We detected similar serum levels of calcidiol in slow progressing and mid-rate progressing MS patients (15.7 ± 5.0 ng/ml vs. 15.8 ± 6.6 ng/ml), but interestingly we noticed a marked decrease of calcidiol serum levels in rapidly progressing MS patients (12.8 ± 5.9 ng/ml). In addition, calcidiol level was significantly lower in all subgroups of MS patients when compared to controls (18.2 ± 8.3 ng/ml). Thus we can conclude that decreased serum level of calcidiol in MS patients can be one of the factors related to increased risk of MS development, as well as increased risk of rapid disease disability progression.

### 7.6. Genetic factors related to vitamin D effects in MS

Nucleotide exchange in DNA sequence can cause the production of protein products with different activities. Polymorphisms of the genes involved in the activation, transport, signaling and degradation of vitamin D can, together with other factors, modify the individual immune response and thus can be related to MS. Because of the beneficial effects of vitamin D, in individuals with genes predisposing to its higher serum levels, the risk of MS should be reduced [96]. The serum level of vitamin D can be modified by VDR gene polymorphisms [97–99]. The fact that serum levels of vitamin D are similar in twins, and especially when they are monozygotic twins, speaks in favour of a genetic regulation. Gene polymorphisms FokI in
VDR gene, rs4646536 and rs703842 in the CYP27B1 gene and rs10741657 in the CYP2R1 gene are the significant predictors of calcidiol serum level [99]. Hypovitaminosis D is common in higher latitudes because of the lack of sun exposure [100]. The fact that not all vitamin D–deficient individuals develop MS is probably the result of the complexity of the etiopathogenesis of MS and the interaction of many factors. The positive effects of vitamin D in MS can be dampened for example by the allele HLA-DRB1*15 [96]. In MS patients, it is necessary to find out the link between the genotype and the vitamin D serum level and also the genetic interactions among the genes CYP27B1, VDR and HLA [19]. The gene polymorphisms associated with vitamin D metabolism are summarized in Table 2.

| Gene                        | function     | Localization | SNP            | Allele  |
|-----------------------------|--------------|--------------|----------------|---------|
| CYP27B1 (cytochrome P450 family 27 subfamily B member 1, 25-hydroxyvitamin D_1 alpha-hydroxylase) | Hydroxylation | 12q13        | rs703842    | C/T     |
|                            |              |              | rs10877012    | G/C     |
|                            |              |              | rs4646536     | C/T     |
|                            |              |              | rs10877015    | A/G     |
|                            |              |              | rs11820409    | A/G     |
|                            |              |              | rs118204012   | A/G     |
|                            |              |              | rs118204011   | C/T     |
| CYP2R1 (cytochrome P450 family 2 subfamily R member 1, vitamin D_25-hydroxylase) | Hydroxylation | 11p15        | rs10741657  | A/G     |
|                            |              |              | rs10500804    | G/T     |
|                            |              |              | rs12794714    | A/G     |
| DBP (vitamin D binding protein) | Transport in plasma | 4q12       | rs7041      | G/T     |
|                            |              |              | rs4588       | A/C     |
| VDR (vitamin D receptor)    | Receptor     | 12q13        | rs1544410 (BsmI) | A/G (B/b) |
|                            |              |              | rs7975232 (ApaI) | T/C (A/a) |
|                            |              |              | rs731236 (TaqI) | T/C (T/t) |
|                            |              |              | rs10735810 (FokI) | C/T (F/F) |
|                            |              |              | rs11568820 (Cdx2) | G/A     |
|                            |              |              | rs2254210     | A/G     |
|                            |              |              | rs98784       | C/T     |
| CYP24A1 (cytochrome P450 family 24 subfamily A polypeptide, vitamin D 24-hydroxylase) | Deactivation  | 20q13        | rs2296241   | A/G     |

Table 2. The gene polymorphisms associated with vitamin D metabolism [19, 98, 99, 101].

7.6.1. Genetic variants in vitamin D receptor gene in MS

According to the effects of vitamin D in MS, the molecular mechanisms of vitamin D function should be considered. As mentioned earlier, vitamin D executes its physiological effect via
binding and activation of VDR. Interestingly, the activation of VDR by calcitriol can suppress the induction of EAE, while animals that lack VDR are not protected against EAE [102]. The gene for VDR is located on the 12q13 chromosomal region and consists of 11 exons. Non-coding exons 1A, 1B and 1C are located in the 5’ end of the VDR gene, and exons 2–9 encode the structural portion of the VDR protein [103]. VDR sequence is similar to that of the receptors for steroid hormones and hormones of the thyroid gland. VDR is a regulatory transcription factor and consists of highly conservative DNA-binding and ligand-binding domains. The signal pathways associated with the VDR regulate the transcription of genes involved in the regulation of bone metabolism, immune response and cancer [83]. The polymorphisms in the initiation codon of the VDR gene can cause the formation of transcription variants coding different proteins [104]. In the VDR gene, SNPs Apal (rs7975232), BsmI (rs1544410), FokI (rs10735810) and TaqI (rs731236) have functional biological effects and are mostly studied in MS as well as in other diseases. These gene polymorphisms can alter mRNA level, its stability and alternative splicing and also the stability of the final gene product, amount of protein isoforms and their interactions [105]. FokI gene polymorphism is located in exon 2 of the VDR gene, and its variants result in a change of protein structure. There are two possible allele variants, f (presence of a restriction site for FokI endonuclease) and F (absence of a restriction site for FokI endonuclease). It has been confirmed that the f (T) allele leads to the expression of a VDR protein, which is three amino acids longer (427 amino acids) than the F (C) allele (424 amino acids). The shorter isoform of the receptor is more transcriptionally potent through a more efficient interaction with transcription factor TFIIB [105, 106]. Near the 3’ end of the VDR gene, we can find the Apal and BsmI polymorphism in the intron between exon 8 and 9 and TaqI gene polymorphism in exon 9 [107]. The allele variants of these gene polymorphisms and their combinations regulate the functions of VDR through the modulation of mRNA stability. In Caucasians, TaqI, Apal and BsmI polymorphisms are in strong linkage disequilibrium and are present in five haplotype blocks. Haplotype 2 (t-A-B) probably results in a lower number of ‘A’ in polyA variable number of tandem repeats (VNTR), while haplotype 1 (T-A-b) is connected to a large number of ‘A’, thus modulating mRNA stability [106]. Morrison et al. [108] found that allele b (G) of the BsmI polymorphism causes a decreased expression of VDR mRNA.

Interestingly, several studies have found an association between VDR gene polymorphisms and the risk of MS. Differences in allele frequency of the BsmI polymorphism in the VDR gene were found in Japan by Fukazawa et al. [109], who for the first time pointed out the involvement of VDR gene polymorphisms in the pathogenesis of MS. The association of VDR gene polymorphisms with MS has been confirmed in cohorts of MS patients from Japan [110], the UK [111, 112], Australia [107] and the USA [98]. On the contrary, no association of VDR gene polymorphisms with the risk of MS was found by studies in MS patients from Canada [113], Netherlands [114], Greece [115], Spain [116, 117], Tasmania [118] and Iran [119]. The presence of specific haplotypes of the VDR gene can increase the risk of MS development, especially its progressive forms. Tajouri et al. [107] in Australia found haplotype A-t (T-C) of Apal and TaqI polymorphism to increase the risk of MS development, especially its progressive forms. The carriage of allele t (C) in their study increased MS risk twice. Fukazawa et al. [109] found allele b (G) and genotype bb (GG) of BsmI polymorphism to increase MS risk, but without any
association with the form and severity of MS (EDSS, magnetic resonance imaging (MRI)). Allele b (G) of BsmI polymorphism of VDR has been found to be associated with MS risk in combination with allele A (T) of ApaI polymorphism by Niino et al. [110]. However, in their study, they did not find any association of ApaI gene polymorphism with clinical form and severity of MS evaluated by the EDSS score, disease duration and MRI findings. Agliardi et al. [120] in Italy found that allele T (T) and genotype TT (TT) are protective against MS development, supported by the finding that the expression of VDR mRNA is increased four times by genotype Tt (TC) and eight times by genotype TT (TT) when compared to genotype tt (CC). The observed effect is present especially when the protective allele T is present in the combination with HLA-DRB1*15 allele.

The role of VDR gene polymorphisms is still not completely understood, and it seems to vary among different populations. For proper cell signalling to decrease the risk of MS, it is probably necessary to reach a certain level of the transcriptional activity of VDR that is also modified genetically. For proper immunoregulation, the individuals that have the genotype causing the decreased VDR protein activity can need a higher peroral vitamin D intake or higher level of sun exposure. Contrarily, in individuals with higher transcriptional activity of VDR, a lower sun exposure or vitamin D intake can be sufficient for proper immune system regulation.

The findings of our previous study in MS patients from the Central-Northern region of Slovakia have confirmed the association of FokI heterozygous genotype Ff with an increased risk of MS in women [10]. Although we found no statistically significant differences in the proportions of FokI genotypes or allele frequencies between total MS patient and the control group, we have observed significant differences in the FokI genotype distribution between women with MS and the female control group (p = 0.042). Our results have shown a significantly higher frequency of heterozygous Ff genotypes in FokI polymorphism in the female MS group (53.4%) as compared to 43.7% in the female control group (OR = 1.48, 95% CI = 1.01–2.16). In spite of this fact, when we compared the subgroup of rapidly progressing MS patients with the subgroup of slowly progressing MS patients, allele and genotype counts were not significantly different between them (allele f: 34.5 vs. 43.3%, allele F: 65.5 vs. 56.7%, genotype ff: 10.3 vs. 13.4%, genotype Ff: 48.3 vs. 59.8%). Since we have not shown any significant association between FokI VDR gene polymorphism and the rate of disease disability progression in our cohort of Slovak MS patients, we observed a trend of higher frequency of homozygotes FF to be 41.4% in MS patients with rapid progression of disease as compared to 26.8% in slow progressing MS patients (OR = 1.93, 95% CI=0.94–3.94) with a marginal level of significance (p = 0.071). From the results of our study, it seems that contributions from genetic and allelic variants of FokI VDR gene polymorphism have only a small impact in a disease as complex as MS, and its role in the etiopathogenesis of MS still remains controversial.

8. Conclusions

In summary, we can conclude that many genetic and biochemical factors can be involved in the etiopathogenesis of MS. These markers could be used to evaluate the risk of MS development and the risk of rapid disease disability progression.
The proposed markers that have been found to be associated with MS risk or disability progression in Central European Slovak population are summarized in Table 3. In our studies, we identified decreased serum level of vitamin D, allele C and genotype CC of polymorphism rs6897932 in the *IL7Ra* gene, genotype Ff of rs10735810 in the *VDR* gene (only in women); HLA-alleles DRB1*15, DRB1*03, DQB1*06; HLA-genotypes DRB1*15/*15, DQB1*06/*06 and HLA-haplotype DRB1*15-DQB1*06 as the main risk factors for MS development. On the contrary, allele T of rs6897932 in the *IL7Ra* gene (in individuals with genotype CT and TT); HLA-alleles DRB1*07, DRB1*13, DQB1*03; HLA-genotypes DRB1*13/*11, DQB1*05/*03 and HLA-haplotypes DRB1*13-DQB1*06 and DRB1*11-DQB1*03 displayed a protective effect against MS development. Genotype CC of rs6897932 in the *IL7Ra* gene and decreased serum level of vitamin D were identified as negative prognostic factors for rapid disability progression in MS, while minor allele T of rs6897932 in the *IL7Ra* gene (especially in individuals with TT genotype) was identified as a protective factor disability progression.

| **Risk factors** | **Rapid disease disability progression** |
|-----------------|----------------------------------------|
| Decreased serum vitamin D | Decreased serum vitamin D |
| rs6897932 in *IL7Ra* gene—allele C, genotype CC | rs6897932 in *IL7Ra* gene—genotype CC |
| HLA-alleles DRB1*15, DRB1*03, DQB1*06; HLA-genotypes DRB1*15/*15, DQB1*06/*06; HLA-haplotype DRB1*15-DQB1*06 | |
| rs10735810 in *VDR* gene | |
| Ff (only women) | |

| **Protective factors** | **Rapid disease disability progression** |
|-----------------------|----------------------------------------|
| rs6897932 in *IL7Ra* gene—allele T, genotype CT and TT | rs6897932 in *IL7Ra* gene—allele T, genotype TT |
| HLA-alleles DRB1*07, DRB1*13, DQB1*03; HLA-genotypes DRB1*13/*11, DQB1*05/*03; HLA-haplotypes DRB1*13-DQB1*06, DRB1*11-DQB1*03 | |

Table 3. The proposed markers associated with the MS risk or disability progression in Slovaks [10, 41, 65].

From the results of our study, we conclude that rs6897932 of the *IL7Ra* gene, rs10735810 in the *VDR* gene, HLA-DR and DQ genotypes, as well as serum level of vitamin D may be the important markers that could be used as part of a panel of markers to evaluate the risk of MS development and disability progression. The relevance of these markers identified in our study should be verified in larger groups of individuals not only in Slovakia but also in other different populations. The relevant positive or negative prognostic genetic or biochemical markers can
improve the diagnostic and therapeutic procedure and can help to minimize neurological damage in predisposed individuals.

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References

[1] Kakalacheva K, Lünemann JD. Environmental triggers of multiple sclerosis. FEBS Lett. 2011; 585(23):3724–9. doi: 10.1016/j.febslet.2011.04.006

[2] International Multiple Sclerosis Genetics Consortium, Hafler DA, Compston A, Sawcer S, Lander ES, Daly MJ, De Jager PL, de Bakker PI, Gabriel SB, Mirel DB, Ivinson AJ, Pericak-Vance MA, Gregory SG, Rioux JD, McCauley JL, Haines JL, Barcellos LF, Cree B, Oksenberg JR, Hauser SL. Risk alleles for multiple sclerosis identified by a genome-wide study. N Engl J Med. 2007;357(9):851-62. PMID: 17660530

[3] Nischwitz S, Müller-Myhsok B, Weber F. Risk conferring genes in multiple sclerosis.FEBS Lett. 2011; 585(23):3789–97. doi: 10.1016/j.febslet.2011.03.037
[4] Stinissen P, Hellings N. Activation of myelin reactive T cells in multiple sclerosis: a possible role for T cell degeneracy? Eur J Immunol. 2008; 38(5):1190–3. doi: 10.1002/eji.200838371

[5] Compston A, Coles A. Multiple sclerosis. The Lancet. 2008; 372(9648):1502–17. doi: 10.1016/S0140-6736(08)61620-7

[6] Levin MC, Douglas JN, Meyers L, Sangmin L, Yoojin S, Gardner LA. Neurodegeneration in multiple sclerosis involves multiple pathogenic mechanisms. Degen Neurol Neuromuscul Dis. 2014; 2014(4):49–63. doi: http://dx.doi.org/10.2147/DNND.S54391

[7] Kostic MS, Rajkovic JS, PoticFloranovic MS, Dimov ID, Pavlovic DD. Multiple sclerosis and oxidative stress—a clinical perspective. Neurochem J. 2013; 7(1):76–86. doi: 10.1134/S1819712412040083

[8] Roxburgh RH, Seaman SR, Masterman T, Hensiek AE, Sawcer SJ, Vukusic S, Achti I, Confavreux C, Costans M, le Page E, Edan G, McDonnell GV, Hawkins S, Trojan M, Liguori M, Cocco E, Marroso MG, Tesser F, Leone MA, Weber A, Zipp F, Miterski B, Epplen JT, Oturai A, Sorensen PS, Celius EG, Lara NT, Montalan X, Villoslada P, Silva AM, Marta M, Lete I, Dubois B, Rubio J, Butzkueven H, Kilpatrick T, Mycko MP, Selmaj KW, Rio ME, Sá M, Salemi G, Savettieri G, Hillert J, Compston DA. Multiple sclerosis severity score: using disability and disease duration to rate disease severity. Neurology. 2005; 64(7):1144–51. PMID: 15824338

[9] Kurtzke JF. Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). Neurology. 1983; 33(11):1444–52. PMID: 6685237

[10] Čierny D, Michalík J, Kurča E, Dobrota D, Lehótský J. FokI vitamin D receptor gene polymorphism in association with multiple sclerosis risk and disability progression in Slovaks. Neurol Res. 2015; 37(4):301–8. doi: 10.1179/1743132814Y.0000000459

[11] Orton SM, Herrera BM, Yee IM, Valdar W, Ramagopalan SV, Sadovnick AD, Ebers GC; Canadian Collaborative Study Group. Sex ratio of multiple sclerosis in Canada: a longitudinal study. Lancet Neurol. 2006; 5(11):932–6. PMID: 17052660

[12] Hayes CE. Vitamin D: a natural inhibitor of multiple sclerosis. Proc Nutr Soc. 2000; 59(4):531–5. PMID: 11115787

[13] Wall JD, Pritchard JK. Assessing the performance of the haplotype block model of linkage disequilibrium. Am J Hum Genet. 2003; 73(3):502–15. PMID: 12916017

[14] De Bakker PI, McVean G, Sabeti PC, Miretti MM, Green T, Marchini J, Ke X, Monsuur AJ, Whittaker P, Delgado M, Morrison J, Richardson A, Walsh EC, Gao X, Galver L, Hart J, Hafler DA, Pericak-Vance M, Todd JA, Daly MJ, Trowsdale J, Wijmenga C, Vyse TJ, Beck S, Murray SS, Carrington M, Gregory S, Deloukas P, Rioux JD. A high-resolution HLA and SNP haplotype map for disease association studies in the extended human MHC. Nat Genet. 2006; 38(10):1166–72. PMID: 16998491

[15] Sombekke MH, Lukas C, Crusius JB, Tejedor D, Killestein J, Artesa D, Martinez A, Uitdehaag BM, Knol DL, Peña AS, Geurts JJ, De Jager PL, Barkhof F, Vrenken H, Polman
CH. HLA-DRB1*1501 and spinal cord magnetic resonance imaging lesions in multiple sclerosis. Arch Neurol. 2009; 66(12):1531–6. doi: 10.1001/archneurol.2009.278

[16] Benešová Y, Vašků A, Stourač P, Hladíková M, Fiala A, Bednařík J. Association of HLA-DRB1*1501 tagging rs3135388 gene polymorphism with multiple sclerosis. J Neuroimmunol. 2013; 255(1–2):92–6. doi: 10.1016/j.jneuroim.2012.10.014

[17] Mehra NK, Kaur G. 2003. Gene map of the human leukocyte antigen (HLA) region. In Expert Rev Mol Med. 2003; 5:1. Available from: http://journals.cambridge.org/full-text_content/ERM/ERM5_07/S1462399403005957sup001.pdf

[18] Jedlička P. Onemocnění bíléhmotymozkomíšní. In: Jedlička P, Keller O. editors. Speciálníneurologie. 1st ed. Praha: Galén; 2005. p. 203–212. ISBN 80-7262-312-5.

[19] Sundqvist E, Bäärnhielm M, Alfredsson L, Hillert J, Olsson T, Kockum I. Confirmation of association between multiple sclerosis and CYP27B1. Eur J Hum Genet. 2010; 18(12):1349–52. doi: 10.1038/ejhg.2010.113

[20] Dyment DA, Herrera BM, Cader MZ, Willer CJ, Lincoln MR, Sadovnick AD, Risch N, Ebers GC. Complex interactions among MHC haplotypes in multiple sclerosis: susceptibility and resistance. Hum Mol Genet. 2005; 14(14):2019–26. PMID: 15930013

[21] Oksenberg JR, Barcellos LF, Cree BA, Baranzini SE, Bugawan TL, Khan O, et al. Mapping multiple sclerosis susceptibility to the HLA-DR locus in African Americans. Am J Hum Genet. 2004; 74(1):160–7.

[22] Lincoln MR, Montpetit A, Cader MZ, Saarela J, Dyment DA, Tiislar M, Ferretti V, Tienari PJ, Sadovnick AD, Peltonen L, Ebers GC, Hudson TJ. A predominant role for the HLA class II region in the association of the MHC region with multiple sclerosis. Nat Genet. 2005; 37(10):1108–12.

[23] Sawcer S, Hellenthal G, Pirinen M, Spencer CCA, Patsopoulos NA, Moutsianas L, et al. The International Multiple Sclerosis Genetics Consortium (IMSGC), Wellcome Trust Case Control Consortium 2 (WTCCC2). Genetic risk and a primary role for cell-mediated immune mechanisms in multiple sclerosis. Nature. 2011; 476(7359):214–9. doi: 10.1038/nature10251

[24] Zhang Q, Lin CY, Dong Q, Wang J, Wang W. Relationship between HLA-DRB1 polymorphism and susceptibility or resistance to multiple sclerosis in Caucasians: a meta-analysis of non-family-based studies. Autoimmun Rev. 2011; 10(8):474–81. doi: 10.1016/j.autrev.2011.03.003

[25] Schmidt H, Williamson D, Ashley-Koch A. HLA-DR15 haplotype and multiple sclerosis: a HuGE review. Am J Epidemiol. 2007; 165(10):1097–109. PMID: 17329717

[26] Ramagopalan SV, Morris AP, Dyment DA, Herrera BM, DeLuca GC, Lincoln MR, Orton SM, Chao JM, Sadovnick AD, Ebers GC. The inheritance of resistance alleles in multiple sclerosis. PLoS Genet. 2007; 3(9):e150. doi: 10.1371/journal.pgen.0030150
[27] Masterman T, Ligers A, Olsson T, Andersson M, Olerup O, Hillert J. HLA-DR15 is associated with lower age at onset in multiple sclerosis. Ann Neurol. 2000; 48(2):211–9. PMID: 10939572

[28] Kaimen-Maciel DR, Reiche EM, Borelli SD, Morimoto HK, Melo FC, Lopes J, Dorigon RF, Cavalet C, Yamaguchi EM, Silveira TL, Da Silva WV, Comini-Frota ER, Brum Souza DG, Donadi EA. HLA-DRB1* allele-associated genetic susceptibility and protection against multiple sclerosis in Brazilian patients. Mol Med Rep. 2009; 2(6):993–8. doi: 10.3892/mmr_0000204.

[29] Romero-Pinel L, Pujal JM, Martínez-Yélamos S, Guiberras L, Matas E, Bau L, Torrabadella M, Azqueta C, Arbizu T. HLA-DRB1: genetic susceptibility and disability progression in a Spanish multiple sclerosis population. Eur J Neurol. 2011; 18(2):337–42. doi: 10.1111/j.1468-1331.2010.03148.x

[30] Kouri I, Papakonstantinou S, Bempes V, Vasiliadis HS, Kyritsis AP, Pelidou HS.HLA associations with multiple sclerosis in Greece. J Neurol Sci. 2011; 308(1–2):28–31. doi: 10.1016/j.jns.2011.06.037

[31] Marrosu MG, Murru R, Murru MR, Costa G, Zavattari P, Whalen M, Cocco E, Mancosu C, Schirru L, Solla E, Fadda E, Melis C, Porru I, Rolesu M, Cucca F. Dissection of the HLA association with multiple sclerosis in the founder isolated population of Sardinia. Hum Mol Genet. 2001; 10(25):2907–16. PMID: 11741834

[32] Stankovich J, Butzkueven H, Marriott M, Chapman C, Tubridy N, Tait BD, Varney MD, Taylor BV, Foote SJ; ANZgene Consortium, Kilpatrick TJ, Rubio JP. HLA-DRB1 associations with disease susceptibility and clinical course in Australians with multiple sclerosis. Tissue Antigens. 2009; 74(1):17–21. doi: 10.1111/j.1399-0039.2009.01262.x

[33] Fernández O, Fernández V, Alonso A, Caballero A, Luque G, Bravo M, León A, Mayorga C, Leyva L, de Ramón E. DQB1*0602 allele shows a strong association with multiple sclerosis in patients in Malaga, Spain. J Neurol. 2004; 251(4):440–4. PMID: 15083289

[34] Link J, Kockum I, Lorentzen AR, Lie BA, Celius EG, Westerlind H, Schaffer M, Alfredsson L, Olsson T, Brynedal B, Harbo HF, Hillert J. Importance of human leukocyte antigen (HLA) class I and II alleles on the risk of multiple sclerosis. PLoS One. 2012; 7(5):e36779. doi: 10.1371/journal.pone.0036779

[35] Kaushansky N, Ben-Nun A. DQB1*06:02-associated pathogenic anti-myelin autoimmunity in multiple sclerosis-like disease: potential function of DQB1*06:02 as a disease-predisposing allele. Front Oncol. 2014; 4:280.doi: 10.3389/fonc.2014.00280

[36] Caballero A, Alves-Leon S, Papais-Alvarenga R, Fernandez O, Navarro G. Alonso, A. DQB1*0602 confers genetic susceptibility to multiple sclerosis in Afro-Brazilians. Tissue Antigens. 1999; 54(5):524–26. PMID: 10599893
Kaushansky N, Altmann DM, David CS, Lassmann H, Ben-Nun A. DQB1*0602 rather than DRB1*1501 confers susceptibility to multiple sclerosis-like disease induced by proteolipid protein (PLP). J Neuroinflammation. 2012; 9:29. doi: 10.1186/1742-2094-9-29

Isobe N, Gourraud PA, Harbo HF, Caillier SJ, Santaniello A, Khanhkanian P, Maiers M, Spellman S, Cereb N, Yang S, Pando MJ, Piccio L, Cross AH, De Jager PL, Cree BA, Hauser SL, Oksenberg JR. Genetic risk variants in African Americans with multiple sclerosis. Neurology. 2013; 81(3):219–27. doi: 10.1212/WNL.0b013e31829bfe2f

Barcellos LF, Sawcer S, Ramsay PP, Baranzini SE, Thomson G, Briggs F, et al. Heterogeneity at the HLA-DRB1 locus and risk for multiple sclerosis. Hum Mol Genet. 2006; 15(18):2813–24. PMID: 16905561

Cocco E, Murru R, Costa G, Kumar A, Pieroni E, Melis C, Barberini L, Sardu C, Lorefice L, Fenu G, Frau J, Coghe G, Carboni N, Marrosu MG. Interaction between HLA-DRB1-DQB1 Haplotypes in Sardinian multiple sclerosis population. PloS One. 2013; 8(4):e59790. doi: 10.1371/journal.pone.0059790

Michalik J, Čírnová E, Kantárová D, Juraj J, Párník Z, Kurča E, Dobrota D, Lehotský J. The association of HLA-DRB1 and HLA-DQB1 alleles with genetic susceptibility to multiple sclerosis in the Slovak population. Neurol Res. 2015; 37:1060–67 doi: 10.1080/01616412.2015.1115212.

D’Netto MJ, Ward H, Morrison KM, Ramagopalan SV, Dyment DA, Deluca GC, Handunnetthi L, Sadovnick AD, Ebers GC. Risk alleles for multiple sclerosis in multiplex families. Neurology. 2009; 72(23):1984–8. doi: 10.1212/WNL.0b013e3181a92c25

Hoffjan S, Akkad DA. The genetics of multiple sclerosis: an update 2010. Mol Cell Probes. 2010; 24(5):237–43. doi: 10.1016/j.mcp.2010.04.006

Akkad DA, Hoffjan S, Petrasch-Parwez E, Beygo J, Gold R, Epplen JT. Variation in the IL7RA and IL2RA genes in German multiple sclerosis patients. J Autoimmun. 2009; 32(2):110–5. doi: 10.1016/j.jaut.2009.01.002

Brynedal B, Bomfim LL, Olsson T, Duvefelt K, Hillert J. Differential expression, and genetic association, of CD58 in Swedish multiple sclerosis patients. ProcNatAcadSci U S A. 2009; 106(23):E58. doi: 10.1073/pnas.0904338106

De Jager PL, Jia X, Wang J, de Bakker PI, Ottoboni L, Aggarwal NT, Piccio L, Raychaudhuri S, Tran D, Aubin C, Briskin R, Romano S; International MS Genetics Consortium, Baranzini SE, McCauley JL, Pericak-Vance MA, Haines JL, Gibson RA, Naeglin Y, Utidehaag B, Matthews PM, Kappos L, Polman C, McArthuWL, Strachan DP, Evans D, Cross AH, Daly MJ, Compston A, Sawcer SJ, Weiner HL, Hauser SL, Hafler DA, Oksenberg JR. Meta-analysis of genome scans and replication identify CD6, IRF8 and TNRFSF1A as new multiple sclerosis susceptibility loci. Nat Genet. 2009; 41(7):776–82. doi: 10.1038/ng.401
[47] Dyment DA, Cader MZ, Chao MJ, Lincoln MR, Morrison KM, Disanto G, Morahan JM, De Luca GC, Sadovnick AD, Lepage P, Montpetit A, Ebers GC, Ramagopalan SV. Exome sequencing identifies a novel multiple sclerosis susceptibility variant in the TYK2 gene. Neurology. 2012; 79(5):406–11. doi: 10.1212/WNL.0b013e3182616fc4

[48] Park TJ, Kim HJ, Kim JH, Bae JS, Cheong HS, Park BL, Shin HD. Associations of CD6, TNFRSF1A and IRF8 polymorphisms with risk of inflammatory demyelinating diseases. Neuropathol Appl Neurobiol. 2013; 39(5):519–30. doi: 10.1111/j.1365-2990.2012.01304.x

[49] Fry TJ, Mackall CL. 2002. Interleukin-7: from bench to clinic. Blood. 2002; 99(11):3892–904

[50] Bomprezzi R, Ringnér M, Kim S, Bittner ML, Khan J, Chen Y, Elkahloun A, Yu A, Bielekova B, Meltzer PS, Martin R, McFarland HF, Trent JM. Gene expression profile in multiple sclerosis patients and healthy controls: identifying pathways relevant to disease. Hum Mol Genet. 2003; 12(17):2191–9. PMID: 12915464

[51] Ramanathan M, Weinstock-Guttman B, Nguyen LT, Badgett D, Miller C, Patrick K, Brownscheidel C, Jacobs L. In vivo gene expression revealed by cDNA arrays: the pattern in relapsing–remitting multiple sclerosis patients compared with normal subjects. J Neuroimmunol. 2001; 116(2):213–9. PMID: 11438176

[52] Lundmark F, Duvefelt K, Iacobaeus E, Kockum I, Wallström E, Khademi M, Oturai A, Ryder LP, Saarelä J, Harbo HF, Celius EG, Salter H, Olsson T, Hillert J. Variation in interleukin 7 receptor alpha chain (IL7R) influences risk of multiple sclerosis. Nat Genet. 2007; 39(9):1108-13. PMID: 17660816

[53] Goodwin RG, Friend D, Ziegler SF, Jerzy R, Falk BA, Gimpel S, Cosman D, Dower SK, March CJ, Namen AE, et al. Cloning of the human and murine interleukin-7 receptors: demonstration of a soluble form and homology to a new receptor superfamily. Cell. 1990; 60(6):941–51. PMID: 2317865

[54] Kreft KL, Verbraak E, Wierenga-Wolf AF, van Meurs M, Oostra BA, Laman JD, Hintzen RQ. Decreased systemic IL-7 and soluble IL-7Rα in multiple sclerosis patients. Genes Immun. 2012; 13(7):587–92. doi: 10.1038/gen.2012.34

[55] Zhang Z, Duvefelt K, Svensson F, Masterman T, Jonasdottir G, Salter H, Emahazion T, Hellgren D, Falk G, Olsson T, Hillert J, Anvret M. Two genes encoding immune-regulatory molecules (LAG3 and IL7R) confer susceptibility to multiple sclerosis. Genes Immun. 2005; 6(2):145–52. PMID: 15674389

[56] Teutsch SM, Booth DR, Bennetts BH, Heard RN, Stewart GJ. Identification of 11 novel and common single nucleotide polymorphisms in the interleukin-7 receptor-alpha gene and their associations with multiple sclerosis. Eur J Hum Genet. 2003; 11(7):509–15. PMID: 12825072

[57] Gregory SG, Schmidt S, Seth P, Oksenberg JR, Hart J, Prokop A, Caillier SJ, Ban M, Goris A, Barcellos LF, Lincoln R, McCauley JL, Sawcer SJ, Compston DA, Dubois B, Hauser
SL, Garcia-Blanco MA, Pericak-Vance MA, Haines JL; Multiple Sclerosis Genetics Group. Interleukin 7 receptor alpha chain (IL7R) shows allelic and functional association with multiple sclerosis. Nat Genet. 2007; 39(9):1083–91. PMID: 17660817

[58] O’Doherty C, Kantarci O, Vandenbroeck K. IL7RA polymorphisms and susceptibility to multiple sclerosis. N Engl J Med. 2008; 358(7):753–4. doi: 10.1056/NEJMc0707553

[59] Alcina A, Fedetz M, Ndagire D, Fernández O, Leyva L, Guerrero M, Arnal C, Delgado C, Matesanz F. The T244I variant of the interleukin-7 receptor-alpha gene and multiple sclerosis. Tissue Antigens. 2008; 72(2):158–61. doi: 10.1111/j.1399-0039.2008.01075.x

[60] Weber F, Fontaine B, Cournu-Rebeix I, Kroner A, Knop M, Lutz S, Müller-Sarnowski F, Uhr M, Bettecken T, Kohli M, Ripke S, Ising M, Rieckmann P, Brassat D, Semana G, Babron MC, Mrejen S, Gout C, Lyon-Caen O, Yaoaung J, Edan G, Clanet M, Holsboer F, Clerget-Darpoux F, Müller-Myhsok B. IL2RA and IL7RA genes confer susceptibility for multiple sclerosis in two independent European populations. Genes Immun. 2008; 9(3):259–63. doi: 10.1038/gene.2008.14

[61] Sombekke MH, van der Voort LF, Kragt JJ, Nielsen JM, Guzel H, Visser A, Oudejans CB, Crusius JB, Peña AS, Vrenken H, Polman CH, Killestein J. Relevance of IL7R genotype and mRNA expression in Dutch patients with multiple sclerosis. MultScler. 2011; 17(8):922–30. doi: 10.1177/1352458511402411

[62] Fang L, Isobe N, Yoshimura S, Yonekawa T, Matsushita T, Masaki K, Doi H, Ochi K, Miyamoto K, Kawano Y, Kira J; South Japan Multiple Sclerosis Genetics Consortium. Interleukin-7 receptor alpha gene polymorphism influences multiple sclerosis risk in Asians. Neurology. 2011; 76(24):2125–7. doi: 10.1212/WNL.0b013e3181f646c

[63] Stanković A, Dincić E, Ristić S, Lovrecić L, Starcević-Cizmarević N, Djurić T, Sepić J, Kapović M, Raicević R, Peterlin B, Alavantić D, Zivković M. Interleukin 7 receptor alpha polymorphism rs6897932 and susceptibility to multiple sclerosis in the Western Balkans. MultScler. 2010; 16(5):533–6. doi: 10.1177/1352458509360548

[64] Booth DR, Arthur AT, Teutsch SM, Bye C, Rubio J, Armati PJ, Pollard JD, Heard RN, Stewart GJ; Southern MS Genetics Consortium. Gene expression and genotyping studies implicate the interleukin 7 receptor in the pathogenesis of primary progressive multiple sclerosis. J Mol Med. 2005; 83(10):822–30. PMID: 16075257

[65] Čierny D, Hányašová S, Michalik J, Kantorová É, Kurča E, Škereňová M, Lehotský J. Genetic variants in interleukin 7 receptor α chain (IL-7Ra) are associated with multiple sclerosis risk and disability progression in Central European Slovak population. J Neuroimmunol. 2015; 282:80–4. doi: 10.1016/j.jneuroim.2015.03.010

[66] Chaudhuri A. Why we should offer routine vitamin D supplementation in pregnancy and childhood to prevent multiple sclerosis. Med Hypotheses. 2005; 64(3):608–18. PMID: 15617877
[67] Saccone D, Asani F, Bornman L. Regulation of the vitamin D receptor gene by environment, genetics and epigenetics. Gene. 2015; 561(2):171–80. doi: 10.1016/j.gene.2015.02.024

[68] Norman AW. Minireview: vitamin D receptor: new assignments for an already busy receptor. Endocrinology. 2006; 147(12):5542–8. PMID: 16946007

[69] Ascherio A, Munger KL, Simon KC. Vitamin D and multiple sclerosis. Lancet Neurol. 2010; 9(6):599–612. doi: 10.1016/S1474-4422(10)70086-7

[70] Adorini L, Penna G. Control of autoimmune diseases by the vitamin D endocrine system. Nat Clin Pract Rheumatol. 2008; 4(8):404–12. doi: 10.1038/ncprheum0855

[71] Bikle DD. Vitamin D regulation of immune function. Vitam Horm. 2011; 86:1–21. doi: 10.1016/B978-0-12-386960-9.00001-0

[72] Smolders J, Thewissen M, Peelen E, Menheere P, Tervaert JW, Damaoiseaux J, Hupperts R. Vitamin D status is positively correlated with regulatory T cell function in patients with multiple sclerosis. PLoS One. 2009; 4(8):e6635. doi: 10.1371/journal.pone.0006635.

[73] Penna G, Adorini L. 1 Alpha,25-dihydroxyvitamin D3 inhibits differentiation, maturation, activation, and survival of dendritic cells leading to impaired alloreactive T cell activation. J Immunol. 2000; 164(5):2405–11. PMID: 10679076

[74] Boonstra A, Barrat FJ, Crain C, Heath VL, Savelkoul HF, O’Garra A. 2001. 1α,25-Dihydroxyvitamin D3 has a direct effect on naïve CD4+ T cells to enhance the development of Th2 cells. J Immunol. 2001; 167(9):4974–80. PMID: 11673504

[75] Geldmeyer-Hilt K, Heine G, Hartmann B, Baumgrass R, Radbruch A, Worm M. 1,25-dihydroxyvitamin D3 impairs NF-kB activation in human naïve B cells. Biochem Biophys Res Commun. 2011; 407(4):699–702. doi: 10.1016/j.bbrc.2011.03.078

[76] Chen S, Sims GP, Chen XX, Gu YY, Chen S, Lipsky PE. Modulatory effects of 1,25-dihydroxyvitamin D3 on human B cell differentiation. J Immunol. 2007; 179(3):1634–47. PMID: 17641030

[77] Lemire JM, Archer DC, Beck L, Spiegelberg HL. Immunosuppressive actions of 1,25-dihydroxyvitamin D3: preferential inhibition of Th1 functions. J Nutr. 1995; 125(6 Suppl):1704–8S. PMID: 7782931

[78] Helming L, Böse J, Ehrchen J, Schiebe S, Frahm T, Geffers R, Probst-Kepper M, Balling R, Lengeling A. 1alpha,25-Dihydroxyvitamin D3 is a potent suppressor of interferon gamma-mediated macrophage activation. Blood. 2005; 106(13):4351–8. PMID: 16118315

[79] Bartels LE, Hvas CL, Agnholt J, Dahlerup JF, Agger R. Human dendritic cell antigen presentation and chemotaxis are inhibited by intrinsic 25-hydroxy vitamin D activation. Int Immunopharmacol. 2010;10(8):922–8. doi: 10.1016/j.intimp.2010.05.003
[80] O’Kelly J, Hisatake J, Hisatake Y, Bishop J, Norman A, Koeffler HP. Normal myelo‐poiesis but abnormal T lymphocyte responses in vitamin D receptor knockout mice. J Clin Invest. 2002; 109(8):1091–9. PMID: 11956247

[81] Panda DK, Miao D, Tremblay ML, Sirookhi R, Hendy GN, Goltzman D. Targeted ablation of the 25-hydroxyvitamin D 1alpha-hydroxylase enzyme: evidence for skeletal, reproductive, and immune dysfunction. Proc Natl Acad Sci U S A. 2001; 98(13):7498–503. PMID: 11416220

[82] Cantorna MT, Hayes CE, DeLuca HF. 1,25-Dihydroxyvitamin D3 reversibly blocks the progression of relapsing encephalomyelitis, a model of multiple sclerosis. Proc Natl Acad Sci U S A. 1996; 93(15):7861–4. PMCID: PMC38839

[83] Kamen DL, Tangpricha V. Vitamin D and molecular actions on the immune system: modulation of innate and autoimmunity. J Mol Med (Berl). 2010; 88(5):441–450. doi: 10.1007/s00109-010-0590-9

[84] Mattner F, Smiroldo S, Galbiati F, Muller M, Di Lucia P, Poliani PL, Martino G, Panin Bordignon P, Adorini L. Inhibition of Th1 development and treatment of chronic-relapsing experimental allergic encephalomyelitis by a non-hypercalcemic analogue of 1,25-dihydroxyvitamin D(3). Eur J Immunol. 2000; 30(2):498–508. PMID: 10671205

[85] Haynes RC Jr. Vitamin D. In Gilman AG, Rall TW, Nies AR, Taylor P. editors. Goodman and Gilman’s the pharmacological basis of therapeutics. 8th ed. New York: McGraw Hill; 1992. p. 1515

[86] Pierrot-Deseilligny C, Souberbielle JC. Widespread vitamin D insufficiency: a new challenge for primary prevention, with particular reference to multiple sclerosis. Presse Med. 2011; 40(4 Pt 1):349–56. doi: 10.1016/j.pmed.2011.01.003

[87] Burton JM, Kimball S, Vieth R, Bar-Or A, Dosch HM, Cheung R, Gagne D, D’Souza C, Ursell M, O’Connor P. A phase I/II dose-escalation trial of vitamin D3 and calcium in multiple sclerosis. Neurology. 2010; 74(23):1852–9. doi:10.1212/WNL.0b013e3181e1ec2

[88] Kimball SM, Ursell MR, O’Connor P, Vieth R. Safety of vitamin D3 in adults with multiple sclerosis. Am J Clin Nutr. 2007; 86(3):645–51. PMID: 17823429

[89] Munger KL, Zhang SM, O’Reilly E, Hernán MA, Olek MJ, Willett WC, Ascherio A. Vitamin D intake and incidence of multiple sclerosis. Neurology. 2004; 62(1):60–5. PMID: 14718698

[90] Munger KL, Levin LI, Hollis BW, Howard NS, Ascherio A. Serum 25-hydroxyvitamin D levels and risk of multiple sclerosis. JAMA. 2006; 296(23):32–8. PMID: 17179460

[91] Soilu-Hänninen M, Airas L, Mononen I, Heikkilä A, Viljanen M, Hänninen A. 25-Hydroxyvitamin D levels in serum at the onset of multiple sclerosis. Mult Scler. 2005; 11(3):266–71.
[92] Ozgocmen S, Bulut S, Ilhan N, Gulkesen A, Ardicoglu O, Ozkan Y. Vitamin D deficiency and reduced bone mineral density in multiple sclerosis: effect of ambulatory status and functional capacity. J Bone Miner Metabol. 2005; 23(4):309–13. PMID: 15981027

[93] Smolders J, Menheere P, Kessels A, Damoiseaux J, Hupperts R. Association of vitamin D metabolite levels with relapse rate and disability in multiple sclerosis. Mult Scler. 2008; 14(9):1220–4. doi: 10.1177/1352458508094399

[94] Runia TF, Hop WC, de Rijke YB, Buljevac D, Hintzen RQ. Lower serum vitamin D levels are associated with a higher relapse risk in multiple sclerosis. Neurology. 2012; 79(3): 261–6. doi: 10.1212/WNL.0b013e31825fdec7

[95] Cadden MH, Koven NS, Ross MK. Neuroprotective effects of vitamin D in multiple sclerosis. Neurosci Med. 2011; 2(3):198–207. doi: 10.4236/nm.2011.23027

[96] Simon KC, Munger KL, Kraft P, Hunter DJ, De Jager PL, Ascherio A. Genetic predictors of 25-hydroxyvitamin D levels and risk of multiple sclerosis. J Neurol. 2011; 258(9): 1676–82. doi: 10.1007/s00415-011-6001-5

[97] Smolders J, Damoiseaux J, Menheere P, Tervaert JW, Hupperts R. Fok-I vitamin D receptor gene polymorphism (rs10735810) and vitamin D metabolism in multiple sclerosis. J Neuroimmunol. 2009; 207(1–2):117–21. doi: 10.1016/j.jneuroim.2008.12.011

[98] Simon KC, Munger KL, Xing Yang, Ascherio A. Polymorphisms in vitamin D metabolism related genes and risk of multiple sclerosis. Mult Scler. 2010; 16(2):133–8. doi: 10.1177/1352458509355069

[99] Orton SM, Morris AP, Herrera BM, Ramagopalan SV, Lincoln MR, Chao MJ, Vieth R, Sadovnick AD, Ebers GC. Evidence for genetic regulation of vitamin D status in twins with multiple sclerosis. Am J Clin Nutr. 2008; 88(2):441–7. PMID: 18689381

[100] Holick MF, Chen TC. Vitamin D deficiency: a worldwide problem with health consequences. Am J Clin Nutr. 2008; 87(4):1080S–6S. PMID: 18400738

[101] Ramagopalan SV, Dyment DA, Cader MZ, Morrison KM, Disanto G, Morahan JM, Berlanga-Taylor AJ, Handel A, De Luca GC, Sadovnick AD, Lepage P, Montpetit A, Ebers GC. Rare variants in the CYP27B1 gene are associated with multiple sclerosis. Ann Neurol. 2011; 70(6):881–6. doi: 10.1002/ana.22678

[102] Meehan TF, DeLuca HF. The vitamin D receptor is necessary for Ialpha,25-dihydroxyvitamin D(3) to suppress experimental autoimmune encephalomyelitis in mice. Arch Biochem Biophys. 2002; 408(2):200–4. PMID: 12464272

[103] Miyamoto K, Kesterson RA, Yama moto H, Taketani Y, Nishiwaki E, Tatsumi S, Inoue Y, Morita K, Takeda E, Pike JW. Structural organization of the human vitamin D receptor chromosomal gene and its promoter. Mol Endocrinol. 1997; 11(8):1165–79. PMID: 9212063

[104] Wang TT, Tavera-Mendoza LE, Laperriere D, Libby E, MacLeod NB, Nagai Y, Bourdeau V, Konstorum A, Lallemant B, Zhang R, Mader S, White JH. Large-scale in silico
and microarray-based identification of direct 1,25-dihydroxyvitamin D3 target genes. MolEndocrinol. 2005; 19(11):2685–95. PMID: 16002434

[105] Uitterlinden AG, Fang Y, Van Meurs JB, Pols HA, Van Leeuwen JP. Genetics and biology of vitamin D receptor polymorphisms. Gene. 2004; 338(2):143–56. PMID: 15315818

[106] Jurutka PW, Remus LS, Whitfield GK, Thompson PD, Hsieh JC, Zitzer H, Tavakkoli P, Galligan MA, Dang HT, Haussler CA, Haussler MR. The polymorphic N terminus in human vitamin D receptor isoforms influences transcriptional activity by modulating interaction with transcription factor IIB. MolEndocrinol. 2000; 14(3):401–20. PMID: 10707958

[107] Tajouri L, Ovcaric M, Curtain R, Johnson MP, Griffiths LR, Csurhes P, Pender MP, Lea RA. Variation in the vitamin D receptor gene is associated with multiple sclerosis in an Australian population. J Neurogenet. 2005; 19(1):25–38. PMID: 16076630

[108] Morrison NA, Qi JC, Tokita A, Kelly PJ, Crofts L, Nguyen TV, Sambrook PN, Eisman JA. Prediction of bone density from vitamin D receptor alleles. Nature. 1994; 367(6460): 284–7. PMID: 8161378

[109] Fukazawa T, Yabe I, Kikuchi S, Sasaki H, Hamada T, Miyasaka K, Tashiro K. Association of vitamin D receptor gene polymorphism with multiple sclerosis in Japanese. J Neurol Sci. 1999; 166(1):47–52. PMID: 10465499

[110] Niino M, Fukazawa T, Yabe I, Kikuchi S, Sasaki H, Tashiro K. Vitamin D receptor gene polymorphism in multiple sclerosis and the association with HLA class II alleles. J Neurol Sci. 2000; 177(1):65–71. PMID: 10967184

[111] Partridge JM, Weatherby SJ, Woolmore JA, Highland DJ, Fryer AA, Mann CL, Boggild MD, Ollier WE, Strange RC, Hawkins CP. Susceptibility and outcome in MS: associations with polymorphisms in pigmentation-related genes. Neurology. 2004; 62(12): 2323–5. PMID: 15210908

[112] Cox MB, Ban M, Bowden NA, Baker A, Scott RJ, Lechner-Scott J. Potential association of vitamin D receptor polymorphism Taq1 with multiple sclerosis. MultScler. 2012; 18(1):16–22. doi: 10.1177/1352458511415562

[113] Steckley JL, Dyment DA, Sadovnick AD, Risch N, Hayes C, Ebers GC. Genetic analysis of vitamin D related genes in Canadian multiple sclerosis patients. Canadian Collaborative Study Group. Neurology. 2000; 54(3):729–32. PMID: 10680811

[114] Smolders J, Damoiseaux J, Menheere P, Tervaert JW, Hupperts R. Association study on two vitamin D receptor gene polymorphisms and vitamin D metabolites in multiple sclerosis. Ann N Y Acad Sci. 2009; 1173:515–20. doi: 10.1111/j.1749-6632.2009.04656.x

[115] Sioka C, Papakonstantinou S, Markoula S, Gkartziou F, Georgiou A, Georgiou I, Pelidou SH, Kyritsis AP, Fotopoulos A. Vitamin D receptor gene polymorphisms in
multiple sclerosis patients in northwest Greece. J Negat Results Biomed. 2011; 10:3. doi: 10.1186/1477-5751-10-3

[116] Irizar H, Muñoz-Culla M, Zuriarrain O, Goyenechea E, Castillo-Triviño T, Prada A, Saenz-Cuesta M, De Juan D, Lopez de Munain A, Olascoaga J, Otaegui D. HLA-DRB1*15:01 and multiple sclerosis: a female association? MultScler. 2012; 18(5):569–77. doi: 10.1177/1352458511426813

[117] García-Martín E, Agúndez J. A. G., Martínez C, Benito-León J, Millán-Pascual J, Calleja P, Díaz-Sánchez M, Pisa D, Turpin-Fenoll L, Alonso-Navarro H, Ayuso-Peralta L, Torrecillas D, Plaza-Nieto JF, Jiménez-Jiménez FJ. Vitamin D3 Receptor (VDR) Gene rs228570 (Fok1) and rs731236 (Taq1) variants are not associated with the risk for multiple sclerosis: results of a new study and a meta-analysis. PLoS One. 2013; 8(6): e65487. doi: 10.1371/journal.pone.0065487

[118] Dickinson JL, Perera DI, van der Mei AF, Ponsonby AL, Polanowski AM, Thomson RJ, Taylor BV, McKay JD, Stankovich J, Dwyer T. Past environmental sun exposure and risk of multiple sclerosis: a role for the Cdx-2 vitamin D receptor variant in this interaction. MultScler. 2009; 5(5):563–70. doi: 10.1177/1352458509102459

[119] Niksresht AR, Dadkhah B, Kamali-Sarvestani E. The association of vitamin D receptor gene BsmI polymorphism with multiple sclerosis in Iranian patients. J Kerman Univ MedSci Health Serv. 2009; 16(2):116–23. doi: http://jkmu.kmu.ac.ir/en/index.php/kmus/article/view/245

[120] Agliardi C, Guerini FR, Saresella M, Caputo D, Leone MA, Zanzottera M, Bolognesi E, Marventano I, Barizzone N, Fasano ME, Al-Daghri N, Clerici M. Vitamin D receptor (VDR) gene SNPs influence VDR expression and modulate protection from multiple sclerosis in HLA-DRB1*15-positive individuals. Brain Behav Immun. 2011; 25(7):1460–7. doi: 10.1016/j.bbi.2011.05.015
