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Research Article

Medicine for novel therapy

Bioinformatics Evolution of Gene Biomarkers in Multiple Sclerosis

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Abstract: Multiple sclerosis (MS) is an autoimmune disease in whsich a person's immune system destroys the myelin around nerve cells in the central nervous system (CNS), yet the peripheral nervous system remains intact. The aim of this study is to investigate the bioinformatics of gene biomarkers in multiple sclerosis. In this study, after reviewing the texts and searching for the bioinformatics databases of NCBI, Gencards, Swiss-prot, Diseasome, etc. the genes involved in the disease based on at, least one of the methods in-vivo, in-vitro, and in-silico has been suggested to be extracted will be considered as candidate genes. In order to compare the results in case and control groups, the expression data obtained from each group was standardized compared to the control group. Then, the connection network of expression data of candidate genes in patients and healthy people was drawn separately with the help of MATLAB software (Version 9.1), and the correctness of these networks and determined biomarkers was checked using the rectome and diseasome database. All statistical calculations were done using R and Matlab software. In the present study, using 5 central criteria including: maximum neighborhood component, degree, closeness, radiality and betweeness, the set of essential genes of MS disease was identified. Based on the results of the central criteria method, TNF, CD40, IL2, IL2RA, IL 7 genes had the most repetitions. According to the identification of the most effective genes related to MS disease in the present study, it is suggested that further studies be designed at the in vitro and clinical levels on the identified effective genes as diagnostic biomarkers of MS disease.

Keywords: Autoimmune Disease, Multiple Sclerosis, Gene Biomarkers, Central Nervous System.

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1. INTRODUCTION

Multiple sclerosis (MS) is an autoimmune disease in which a person's immune system destroys the myelin around nerve cells in the central nervous system (CNS), yet the peripheral nervous system remains intact[1,2]. This damage impairs the ability of parts of the nervous system to transmit signals, and as a result, creates a wide range of physical, mental and sometimes psychiatric symptoms and problems[3,4]. Specific symptoms of MS include diplopia, blindness in one eye, feeling weak in muscles and coordination disorder[5]. Susceptibility to MS is multi-gene and each gene accounts for a relatively small amount of the overall risk. The strongest susceptibility signal in extensive genomic studies is related to the HLA-DRB1 gene in the MHC II region, accounting for approximately 10% of the risk of the disease. Most of the genes associated with MS play a role in the acquired immune system, and some affect the susceptibility to other autoimmune diseases[6-8](Table 1). Viral infections or other disease-initiating factors facilitate the entry of T cells and antibodies into the CNS by disrupting the blood-brain barrier. This increases the supply of cell-adherent molecules, matrix metalloproteinases and pro-inflammatory cytokines that call more immune cells to the site, and activate the immune response against antigens such as myelin main proteins, glycoproteins associated with myelin phosphodiesterase and S-100, resulting in the activation of autoimmune reaction via binding the target antigens by antigen-presenting cells, which includes cytokines, macrophages and complement[10-12](Fig 1). The attack of the immune system on the myelin causes the axons to become bare, and as a result, nerve conduction slows down and neurological symptoms appear[13]. Today, a large group of drugs with different molecular mechanisms are used in the treatment of multiple sclerosis; they play a significant role in reducing the recurrence of the disease, prescribing MRI and preventing the permanent disability of patients[14](Table 2). In terms of MS prevalence, among European countries, the highest prevalence is reported for Scotland and Northern Ireland (200 people per 100,000 people)[15,16]. In Iran, there are different statistics of MS prevalence (5 to 74 people per 100 thousand), yet in general, the prevalence rate is higher in Tehran and Isfahan[17]. The disease-related biomarkers provide information about the possible effects of treatment on the disease (predictive biomarkers), the presence of the disease (diagnostic biomarkers) and how a disease develops regardless of the type of disease (prognostic biomarkers)[18]. Predictive biomarkers provide information about possible responses to a specific type of treatment, while prognostic biomarkers provide information about disease progression, whether the patient is treated or not[19]. The aim of this study is to investigate the bioinformatics of gene biomarkers in multiple sclerosis.

Table 1. MS-related genes that play a role in the acquired immune system and influence susceptibility to other autoimmune diseases.[9]

Category	GO ID	Gene Ontology Term	Adjusted p-value	Genes
Biological Process	GO:1902652	secondary alcohol metabolic process	0.0006	LDLRAP1;CLN8
	GO:0090181	regulation of cholesterol metabolic process	0.0010	SP1;LDLRAP1
	GO:0016125	sterol metabolic process	0.0021	LDLRAP1;CLN8
	GO:0008203	cholesterol metabolic process	0.0028	LDLRAP1;CLN8
	GO:0090068	positive regulation of cell cycle process	0.0067	PKN2;DBF4B
Cellular Component	GO:0030665	clathrin-coated vesicle membrane	0.0039	LDLRAP1;VAMP3
	GO:0030136	clathrin-coated vesicle	0.0059	LDLRAP1;VAMP3
	GO:0030121	AP-1 adaptor complex	0.0080	LDLRAP1
	GO:0055037	recycling endosome	0.0083	LDLRAP1;VAMP3
	GO:0030130	clathrin coat of trans-Golgi network vesicle	0.0103	LDLRAP1
Molecular Function	GO:0035612	AP-2 adaptor complex binding	0.0091	LDLRAP1
	GO:0035005	1-phosphatidylinositol-4-phosphate 3-kinase activity	0.0091	PIK3C2B
	GO:0035615	clathrin adaptor activity	0.0125	LDLRAP1
	GO:0098748	endocytic adaptor activity	0.0125	LDLRAP1
	GO:0016307	phosphatidylinositol phosphate kinase activity	0.0182	PIK3C2B

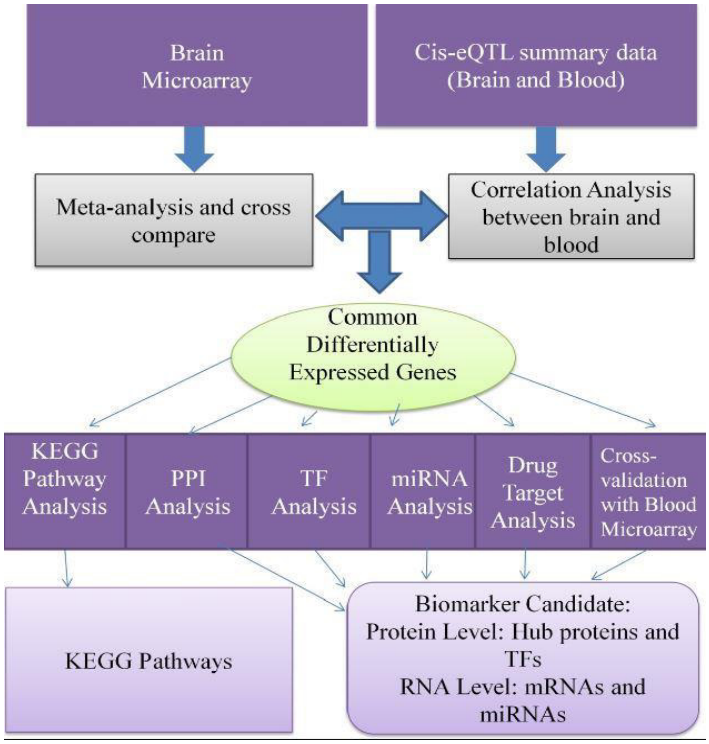

Fig 1: mechanisms of pathogenesis in Multiple Sclerosis

Table 2. The candidate drugs enriched by the differentially expressed genes were identified from the databases.

Drug/Small Molecule	P-value	Genes
Dorzolamide	0.0001	ZNF814;UPK3BL;NEAT1;VAMP3
Iohexol	0.0005	NFATC3;UPK3BL;NEAT1
Naringin	0.0009	NEAT1;VAMP3
Benzylpenicillin	0.0009	UPK3BL;NEAT1
Cicloheximide	0.0009	HN1L;NFATC3;KNOP1;PWP2;NEAT1
Mycophenolic Acid	0.0011	UPK3BL;NEAT1
Gsk461364	0.0012	PKN2;PIK3C2B
Disopyramide	0.0014	HN1L;VAMP3
H-89	0.0021	UPK3BL;VAMP3
0175029–0000	0.0022	ZNF814;NFATC3;KNOP1;PKN2;DBF4B;

2. MATERIALS AND METHODS

The present study is an analytical one. The data were extracted from NCBI, SWISSprot, Genrcards and Disaesome bioinformatics databases from samples of 10,000 MS patients and 20,000 healthy individuals. The genes involved in the disease have been extracted based on at least one in vivo or in vitro and in silico methods, and they were considered as Candidate Genes. In order to investigate the network connection of genes involved in MS disease and to calculate essential factors, genes involved in the disease were determined using text mining method. Then, the set of target genes in this disease was ranked using the Gene-Disease-Association-score (GDA score). After determining the candidate genes from related studies, expression data were collected, and in order to compare the results of the two experimental and control groups, the expression data obtained from each group was standardized compared to the control group (Table 3)(Fig 2). Then, the communication network of expression data of candidate genes was drawn in sick and healthy individuals separately using MATLAB (Version 9.1), and the structural parameters of communication networks of expression data were calculated and compared[20]. Significant parameters were introduced as potential biomarkers, and using rectome and diseasome databases, the validity of these determined networks and biomarkers were checked for a second time. All statistical calculations in this research were done using R and Matlab. In order to analyze the data, advanced descriptive and analytical statistical methods were used. Moreover, machine learning methods based on advanced bioinformatics algorithms were used to calculate features and network data analysis to extract biomarkers related to the structural characters of the network.

3. RESULTS

The results of the present study showed that according to the Maximum-Neighborhood-Component (MNC) index, the most effective biomarker of MS disease network is related to TNF and the least effective is related to CD2. According to the Degree index, the most effective biomarker of the MS disease network is related to TNF and the least effective is related to IL7R. In terms of Closeness criterion, the most effective biomarker of the MS disease network is related to TNF and the least effective is related to TYK2. In terms of Radiality criterion, the most effective biomarker of the MS disease network is related to TNF and the least effective is related to TYK2. In terms of Betweeness criterion, a gene with the highest score is likely to have the greatest effect on the

transmission of information in the biological network compared to other vertices of the network, and removing them from the network will disrupt the entire network communication. Based on this criterion, the most effective biomarker of the MS disease network is related to TNF and the least effective is related to CD40LG (Table 4).

Table 3. The expression levels of sorted candidate genes were extracted by Gene Enterz and Uniprot. In the next step, using the gephi platform, the communication network between the candidate genes was drawn, and while determining the communication structure network between the candidate genes, the structural concentration criteria of the network were calculated to determine the essential genes and proteins

Gene	Uniprot	GDA
HLA-DRB1	PO1911	0.5
IL2RA	PO1589	0.5
IL7R	PLE871	0.5
TNFRSF1A	P19456	0.5
CLEC16A	Q2K1013	0.5
CD40	P25942	0.5
CD58	P19256	0.5
TYK2	P29597	0.49
KIF1B	O60333	0.46
HLADRA	PO1903	0.44

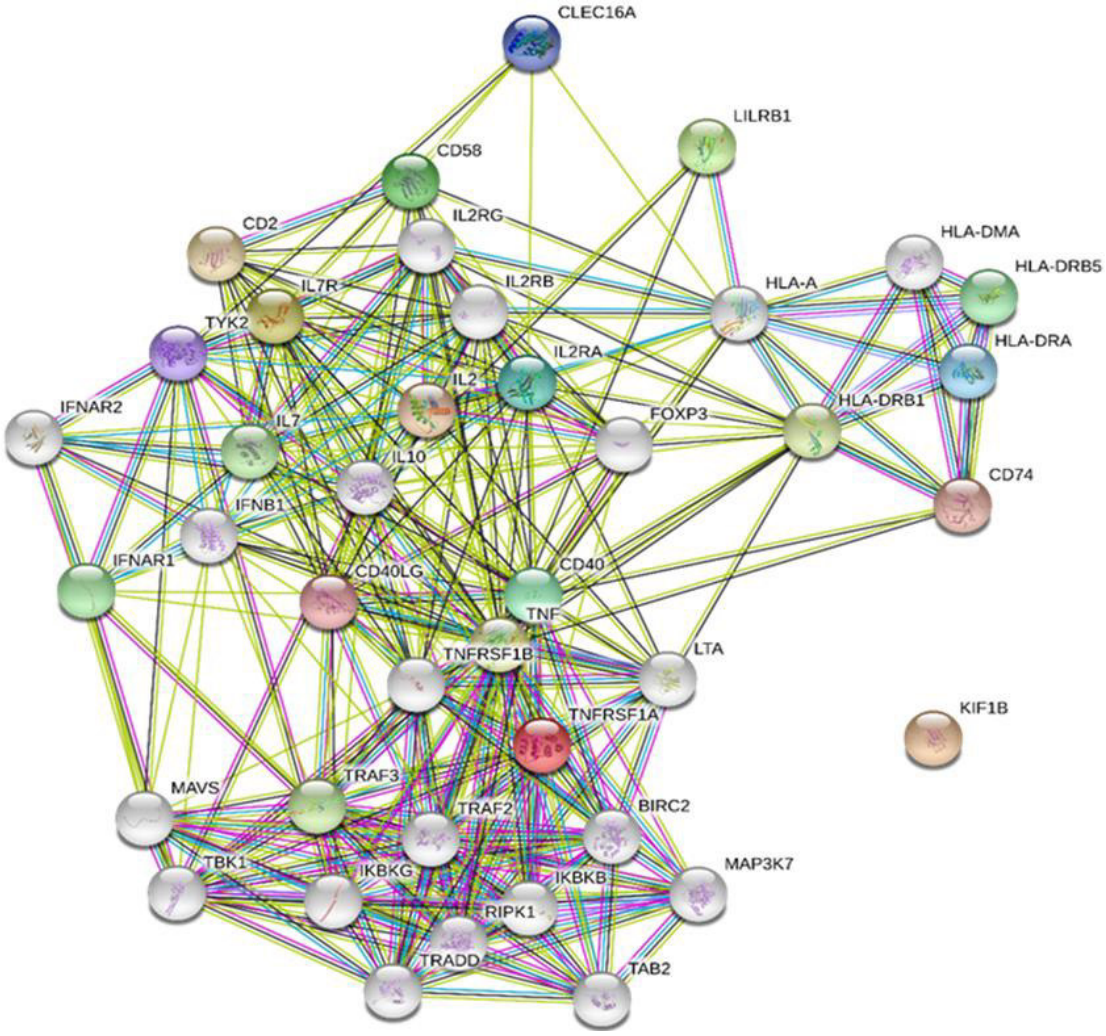

Fig 2: Network of communication between proteins in MS disease

Table 4. Maximum Neighborhood Component, Degree, Closeness, Radiality and Betweeness for MS disease protein association network

Criteria / Rating	Maximum Neighborhood Component	Degree	Closeness	Radiality	Betweeness
1	TNF	TNF	TNF	TNF	TNF
2	CD40	CD40	CD40	CD40	HLA-DRB1
3	IL2	IL2	IL2	IL2	CD40
4	IL2RA	IL2RA	IL2RA	IL2RA	IL2
5	IL7	CD40LG	IL7	IL7	CD74
6	IL7R	CD88	IL7R	IL7R	IL2RA
7	CD40IG	TYK1	CD40LG	CD40LG	TYK2
8	CD58	CD2	CD58	HLA-DRB1	IL7
9	TYK2	TYK2	HLA-DRB1	CD58	IL7R
10	CD2	IL7R	TYK2	TYK2	CD40LG

4. DISCUSSION

Today, the probability of developing chronic and incurable diseases has been on the rise. MS is the most common non-accidental disabling disease in young adults. The onset age of the disease is usually 20 to 40 years old and it is known as a chronic disease with unpredictable symptoms and trends in the reproductive age of the individual[21,22]. The prognosis of this disease has improved in recent decades due to new available treatments and more people are aging with this disease[23]. The recently discussed biomarkers have helped to solve the difficulties and heterogeneities of MS disease pathophysiology and can be a way to improve clinical tools for researchers and doctors. In this study, bioinformatics methods were used to investigate the set of essential genes in the diagnosis and treatment of MS. Based on this, by using 5 centrality criteria i.e. Degree, Closeness, Radiality, Betweeness and Maximum-Neighborhood-Component, the set of essential genes was identified. Among these, 5 genes including TNF-CD40-IL2-IL2RA-IL7 had the most repetition based on all the results of the above 5 central criteria methods. TNF encodes a multifunctional pro-inflammatory cytokine that belongs to the tumor necrosis factor superfamily[24]. The results of the present study showed that TNF is the most effective biomarker related to MS disease, which is consistent with the results of other studies[25]. It was also reported in the study conducted by Ribeiro et al. (2019), Soluble TNF receptor (sTNFR1) and age are the best predictive factors for the development of disability in MS patients[26]. CD40 biomarker is a member of the TNF receptor superfamily. The results of the present study indicate the key role of this biomarker after TNF in MS patients. In the study conducted by Titova et al. (2023), it was reported that the T-allele of the rs6074022 polymorphism of the CD40 gene has a significant relationship with the average rate of MS disease progression, and the GA genotype of the rs1800629 polymorphism of the TNF-α gene will cause MS exacerbation with a higher frequency[27]. In the study conducted by Pope et al. (2020), it was reported that IL2 biomarker in MS patients, in cooperation with other predisposing factors, will cause the activation of T cells and disease progression[28]. This is in line with the results of the present study. Interleukin-2 alpha (IL2RA) and beta (IL2RB) receptors together with the common gamma chain form the high-affinity IL2 receptor. The results of the present study showed that IL2RA is one of the effective genes in MS; this is in line with the results of other studies[29,30]. IL-7 is widely regarded as a key cytokine, which controls the differentiation and immune responses of several T cell subsets[31]. In the study by Simsek et al. (2019), which was conducted to investigate the relationship between L7R-Promoter-Polymorphisms and Multiple Sclerosis in Turkish population, it was reported that there is a significant relationship between IL7R promoter polymorphisms and the age of MS onset[32]; this is in line with the results of the present study.

5. CONCLUSION

The results of the present study showed that the most effective genes related to MS include: TNF-CD40-IL2-IL2RA-IL7. Thus, in the future, additional studies at the laboratory and clinical levels can be designed on the determined effective genes as diagnostic biomarkers of MS disease. Besides, by investigating the communication paths in the gene communication network of MS disease, one can recognize and study the communication in the disease and the interaction of this disease with other diseases. This can help to understand more about the mechanism of the disease.

6. AUTHOR'S CONTRIBUTION STATEMENT

Dr. Hossein Seidkhani. designed the model and the computational framework and analyzed the data. Dr. Reza Valizadeh. Critical revision of the manuscript for important intellectual content. Administrative, technical, and material support.

7. CONFLICT OF INTEREST

Conflict of interest declared none.

8. REFERENCES

1. Zamani F, Salami M, Soheili M. The effect of probiotic supplementation on motor skills, biochemical factors and histological evaluation in a multiple sclerosis model of mouse. Feyz. 2021;25(4):1031-9.
2. Goldenberg MM. Multiple sclerosis review. Pharm Ther. 2013;37(3):175-84. PMID 22605909.
3. Thompson AJ, Banwell BL, Barkhof F, Carroll WM, Coetzee T, Comi G et al. Diagnosis of multiple sclerosis: 2017 revisions of the McDonald criteria. Lancet Neurol. 2018;17(2):162-73. doi: 10.1016/S1474-4422(17)30470-2, PMID 29275977.

4. Kuhlmann T, Moccia M, Coetzee T, Cohen JA, Correale J, Graves J et al. Multiple sclerosis progression: time for a new mechanism-driven framework. Lancet Neurol. 2023;22(1):78-88. doi: 10.1016/S1474-4422(22)00289-7, PMID 36410373.
5. Broglio SP, Kontos AP, Levin H, Schneider K, Wilde EA, Cantu RC et al. National Institute of Neurological Disorders and Stroke and department of defense sport-related concussion common data elements version 1.0 recommendations. J Neurotrauma. 2018;35(23):2776-83. doi: 10.1089/neu.2018.5643, PMID 29717643.
6. Fazia T, Baldrighi GN, Nova A, Bernardinelli L. A systematic Review of Mendelian randomization studies on multiple sclerosis. Eur J Neurosci. 2023;58(4):3172-94. doi: 10.1111/ejn.16088, PMID 37463755.
7. Cox GM, Kithcart AP, Pitt D, Guan Z, Alexander J, Williams JL et al. Macrophage migration inhibitory factor potentiates autoimmune-mediated neuroinflammation. J Immunol. 2013;191(3):1043-54. doi: 10.4049/jimmunol.1200485, PMID 23797673.
8. Sargsyan SA, Shearer AJ, Ritchie AM, Burgoon MP, Anderson S, Hemmer B et al. Absence of Epstein-Barr virus in the brain and CSF of patients with multiple sclerosis. Neurology. 2010;74(14):1127-35. doi: 10.1212/WNL.0b013e3181d865a1, PMID 20220124.
9. Islam T, Rahman MR, Karim MR, Huq F, Quinn JMW, Moni MA. Detection of multiple sclerosis using blood and brain cells transcript profiles: insights from comprehensive bioinformatics approach. Inform Med Unlocked. 2019;16:100201. doi: 10.1016/j.imu.2019.100201.
10. Soldan SS, Lieberman PM. Epstein-Barr virus and multiple sclerosis. Nat Rev Microbiol. 2023;21(1):51-64. doi: 10.1038/s41579-022-00770-5, PMID 35931816.
11. Alizadeh Khatir A, Hojjati SMM, Ahmadi Ahangar A, Naghshineh H, Saadat P. Multiple sclerosis and its pathophysiology: A narrative review. Tabari Biomed Stu Res J. 2020;2(2):8-15. doi: 10.18502/TBSRJ.v2i2.3757.
12. James RE, Schalks R, Browne E, Eleftheriadou I, Munoz CP, Mazarakis ND et al. Persistent elevation of intrathecal pro-inflammatory cytokines leads to multiple sclerosis-like cortical demyelination and neurodegeneration. Acta Neuropathol Commun. 2020;8(1):66. doi: 10.1186/s40478-020-00938-1, PMID 32398070.
13. Sargsyan SA, Shearer AJ, Ritchie AM, Burgoon MP, Anderson S, Hemmer B et al. Absence of Epstein-Barr virus in the brain and CSF of patients with multiple sclerosis. Neurology. 2010;74(14):1127-35. doi: 10.1212/WNL.0b013e3181d865a1, PMID 20220124.
14. McGinley MP, Goldschmidt CH, Rae-Grant AD. Diagnosis and treatment of multiple sclerosis: a review. JAMA. 2021;325(8):765-79. doi: 10.1001/jama.2020.26858, PMID 33620411.
15. Rothwell PM, Charlton D. High incidence and prevalence of multiple sclerosis in south east Scotland: evidence of a genetic predisposition. J Neurol Neurosurg Psychiatry. 1998;64(6):730-5. doi: 10.1136/jnnp.64.6.730, PMID 9647300.
16. Gray OM, McDonnell GV, Hawkins SA. Factors in the rising prevalence of multiple sclerosis in the north-east of Ireland. Mult Scler. 2008;14(7):880-6. doi: 10.1177/1352458508090663, PMID 18573832.
17. Mirmosayyeb O, Shaygannejad V, Bagherieh S, Hosseinabadi AM, Ghajarzadeh M. Prevalence of multiple sclerosis (MS) in Iran: a systematic Review and meta-analysis. Neurol Sci. 2022;43(1):233-41. doi: 10.1007/s10072-021-05750-w, PMID 34787755.
18. Havasian MR, Panahi J, Khosravi A. Correlation between the lipid and cytokine profiles in patients with coronary heart disease (CHD)(Review article). Life Sci J. 2012;9(4):5772-77.
19. Cowen JB, Sjostrom BF, Doughty AS, Schiffer RB. Case-finding for MS prevalence studies in small communities requires a community-based approach. Neuroepidemiology. 2007;28(4):246-52. doi: 10.1159/000108599, PMID 17878739.
20. Elhorst JP. MATLAB software for spatial panels. Int Reg Sci Rev. 2014;37(3):389-405. doi: 10.1177/0160017612452429.
21. Romero-Pinel L, Bau L, Matas E, León I, Muñoz-Vendrell A, Arroyo P et al. The age at onset of relapsing-remitting multiple sclerosis has increased over the last five decades. Mult Scler Relat Disord. 2022;68(1):104103. doi: 10.1016/j.msard.2022.104103, PMID 36029708.
22. Oh J, Vidal-Jordana A, Montalban X. Multiple sclerosis: clinical aspects. Curr Opin Neurol. 2018;31(6):752-9. doi: 10.1097/WCO.0000000000000622, PMID 30300239.
23. Van Wijmeersch B, Hartung HP, Vermersch P, Pugliatti M, Pozzilli C, Grigoriadis N et al. Using personalized prognosis in the treatment of relapsing multiple sclerosis: A practical guide. Front Immunol. 2022;13(1):991291. doi: 10.3389/fimmu.2022.991291, PMID 36238285.
24. Kouchaki E, Akbari H, Mahmoudi F, Salehi M, Naimi E, Nikoueinejad H. Correlation of serum levels of interleukine-16, CCL27, tumor necrosis factor-related apoptosis-inducing ligand, and B-cell activating factor with multiple sclerosis severity. Iran J Allergy Asthma Immunol. 2022;21(1):27-34. doi: 10.18502/ijaai.v21i1.8610, PMID 35524375.
25. Marastoni D, Pisani AI, Schiavi G, Mazziotti V, Castellaro M, Tamanti A et al. CSF TNF and osteopontin levels correlate with the response to dimethyl fumarate in early multiple sclerosis. Ther Adv Neurol Disord. 2022;15(1):1756286422109212 4. doi: 10.1177/17562864221092124.
26. Ribeiro CM, Oliveira SR, Alfieri DF, Flauzino T, Kaimen-Maciel DR, Simão ANC et al. Tumor necrosis factor alpha (TNF-α) and its soluble receptors are associated with disability, disability progression and clinical forms of multiple sclerosis. Inflamm Res. 2019;68(12):1049-59. doi: 10.1007/s00011-019-01286-0, PMID 31559449.
27. Titova MA, Alifirova VM, Musina NF, Nikolaeva TN. The role of TNF-α, TNFRSF1A, and CD40 gene polymorphisms in multiple sclerosis in the Tomsk region. Neurochem J. 2023;17(3):412-7. doi: 10.1134/S1819712423020150.
28. Pope BJ, Sharma V, Meador W, Reynolds R, Bridges SL, Raman C. Interleukin 2 enhances and sustains IFNy induced STAT1 activation in subpopulations of CD4+ and CD8+ T lymphocytes in untreated relapsing remitting multiple sclerosis (RRMS) patients. J Immunol. 2020;204(1_Supplement):221.9-. doi: 10.4049/jimmunol.204.Supp.221.9.

29. Buhelt S, Søndergaard HB, Oturai A, Ullum H, von Essen MR, Sellebjerg F. Relationship between multiple sclerosis-associated IL2RA risk allele variants and circulating T cell phenotypes in healthy genotype-selected controls. Cells. 2019;8(6):634. doi: 10.3390/cells8060634, PMID 31242590.
30. Tempest A, Veettil SK, Maharajan MK, Earl JC, Ngorsuraches S, Chaiyakunapruk N. Genetic biomarkers in multiple sclerosis: an umbrella Review of meta-analyses of observational studies. Mult Scler Relat Disord. 2022;63(1):103834. doi: 10.1016/j.msard.2022.103834, PMID 35526474.
31. Irene Bishai PD, Mostafa L, Farid RJ, Shehata HSM, Riad NM, Raouf FHA. Association of IL-7Rα gene polymorphisms and serum interleukin 7 levels with multiple sclerosis and neuromyelitis optica in Egyptian patients. Med J. 2023; 91(1):33-42.
32. Simsek H, Geckin H, Sensoz NP, List EO, Arman A. Association between IL7R promoter polymorphisms and multiple sclerosis in Turkish population. J Mol Neurosci. 2019;67(1):38-47. doi: 10.1007/s12031-018-1205-0, PMID 30443838.