Serum Iron and Zinc Levels in Lebanese Multiple Sclerosis Patients

Alaa Matar[1], Shada Jennani[1], Hassan Abdallah[2], Nabil Mohsen[3], Jamilah Borjac[1]

Abstract

Purpose: Multiple sclerosis (MS) is an autoimmune disease of the central nervous system that attacks mainly the myelin sheath covering the axons of neurons. Various studies have reported a potential role of zinc and iron in MS disease. The aim of this study is to estimate the serum level of iron and zinc in Lebanese MS patients.

Methods: Sixty-nine participants were enrolled in this study, 27 were diagnosed with MS according to McDonald's criteria and 42 were normal control. Subjects were matched in age. Serum iron and zinc levels were measured using colorimetric methods. Descriptive methods and Mann-Whitney U test were used in the statistical analysis.

Results: The mean age of MS patients and healthy subjects was 42.8 and 38.3 years respectively. The mean serum iron level in patient and control groups was 84.7 and 83.3 μg/dl respectively. The mean serum zinc level in patient and control groups was 80.6 and 82.0 μg/dl respectively. No significant association was observed between serum iron and zinc levels in both groups. No association was also observed between serum iron and zinc levels in terms of gender.

Conclusion: Our results showed no significant difference in serum iron and zinc levels between MS patients and healthy controls.

Keywords: Multiple Sclerosis, Zinc, Iron, Serum

Acta Neurol Taiwan 2020;29:5-11

INTRODUCTION

Multiple sclerosis (MS) is a chronic neurodegenerative disease of the central nervous system (CNS) affecting mainly young adults [(1)]. Demyelination, degeneration and progressive axonal loss are the main characteristics of this autoimmune disorder that may progress from relapsing-remitting course to much more complicated forms [(2, 3)]. Although its etiology is not well understood, the onset of the disease may happen due to genetic [(4)] and environmental factors [(5)], infectious diseases [(6)], nutrition [(7)], as well as exposure to several minerals [(8)]. Toxic

From the [1]Department of Biological Sciences, Faculty of Science, Beirut Arab University, Debbieh, Lebanon; [2]Department of Neurology, Faculty of Medicine, Lebanese University, Beirut, Lebanon; [3]Department of Nursing, Faculty of Public Health, Lebanese University, Tripoli, Lebanon.
Received November 20, 2019. Revised December 27, 2019.
Accepted February 10, 2020.

Correspondence to: Jamilah Borjac, Department of Biological Sciences, Faculty of Science, Beirut Arab University, Debbieh, Lebanon.
E-mail: j.borjac@bau.edu.lb

mineral levels can provoke metabolic, immunological, and neurological damage affecting the structure of diverse proteins in the myelin sheath altering the normal function of the central nervous system (CNS) [9]. Autoimmune demyelination may arise through autoantibodies secretion leading to phagocytic actions of myelin, thus resulting in progressive damage of the oligodendrocytes and formation of plaques that can be visualized by magnetic resonance imaging (MRI) [10].

Minerals are essential for normal body growing and they play a fundamental role as cofactors in cellular energy production and other biochemical processes [11,12]. Zinc (Zn) is one of the most important mineral in the body especially for the nervous system as it acts as cofactor for distinct enzymes such as matrix metalloproteinase (MMP) and plays a central role in various proteins like myelin basic protein (MBP) [13]. A network of protein including zinc transporters, intracellular binding proteins and channels contributes in the translocation and homeostasis of zinc in the CNS [14]. However, an abnormal vesicular zinc release may lead to matrix metallopeptidase-9 (MMP-9) stimulation causing blood-brain barrier (BBB) disruption accompanied with an infiltration of autoimmune cells and antibodies that attack myelin sheath [15]. A high level of zinc in neurons may also contribute in protein kinase C (PKC) and NADPH oxidases activation and superoxides production resulting in poly (ADP-ribose) polymerase-1 (PARP-1) activation in the nucleus, leading to neurotoxicity [16].

Iron (Fe) is a crucial element for human life as it enters in the structure of different proteins like hemoglobin and acts as cofactor for several enzymes especially those engaged in neurotransmitter synthesis and myelination process [17]. However, an imbalance in iron homeostasis is thought to contribute to the neurodegeneration and inflammation in MS, due to microglia activation, mitochondrial dysfunction and free radicals release in the entire body especially in the CNS [18]. Iron accumulation in the brain was observed in case of BBB impairment due to reduced iron clearance as a result of axonal damage or dysregulation of iron transport proteins caused by inflammation [19].

The most common clinical features of the disease are cerebellar symptoms, motor and sensory dysfunction, bowel and bladder perturbations, eye movement abnormalities as well as numbness and spasticity [20]. Studies revealed that diet containing magnesium and zinc were able to improve MS symptoms implicating their role in MS. In contrast, high levels of metals such as iron, copper, lead and cadmium are believed to play a role in MS etiology [21]. The assessment of these metals in MS patients may lead to an effective progress towards MS treatment through nutritional supplementation.

The aim of the current study is to investigate the levels of zinc and iron in the serum of Lebanese MS patients.

METHODS

A. Participants

Twenty-seven patients with relapsing-remitting Multiple sclerosis (RRMS) disease course according to the diagnostic criteria of McDonald's and 42 healthy controls with no neurological disease from the same geographical areas (Tripoli and Bekaa cities of Lebanon) were enrolled in this study. Informed consent was obtained from all participants. The study was approved by Beirut Arab University institutional review board with a code number of 2019A-0042-S-P-0337. Relevant clinical data including age, family neurological history, medical history, treatment and clinical complication were collected through a well-designed questionnaire.

All participants were not using any mineral supplements. None of the participants had mal-absorption syndrome, cardiovascular diseases, cancer, nor chronic inflammatory disorders such as diabetes mellitus, rheumatoid arthritis and asthma. The MS patients and controls were matched in age.

B. Quantification of Zinc and Iron Levels in Serum

Five ml of venous blood was drawn from controls and MS patients during remission stage. Serum was separated by centrifugation at 9,000 x g for ten minutes and was immediately tested or stored at -20 ˚C until analysis. The serum Zn and Fe and were measured by colorimetric method using specific kits from DIALAB, Austria. Analyses were performed in triplicate and the mean value was calculated.

C. Statistical Analyses

All statistical analyses (percent, mean, Mann-Whitney

U-test) were performed using SPSS version 20. P-values smaller than 0.05 were considered statistically significant.

RESULTS

A. Demographic Data of Participants

The characteristics of both MS patients and controls are shown in Table 1. Thirteen MS patients were male and 14 were female. As for the controls, 13 were male and 29 were female. The average age of MS patients was 42.8 years versus 38.3 years for controls. All MS patients were in the relapsing-remitting multiple sclerosis (RRMS) disease course with an Expanded Disability Status Scale (EDSS) mean of 3.8 and a Multiple Sclerosis Severity Score (MSSS) of 5.5. The mean age of all male and female participants was 43.4 and 37.7 years respectively. Male participants' age ranged between 18 and 72 years while the female participants' age ranged between 22 and 73 years.

B. Clinical Profile of MS Patients

The peak age of MS onset in our study was in the third decade with 60.7% of cases developing their first symptoms between 20 and 40 years where they represent 75% of cases. Disease duration was ranging from 7 to 25 years. The female to male ratio was 1.2:1. A positive family history for MS was present in 28.6% of patients.

The most common symptoms were sensory (53.4%), visual (53.4%) and motor (25%) perturbations. Bladder dysfunction was noticed in approximately 50% of the MS patients. Constipation was a frequent trait which may be due to spinal cord impairment, reduced motility and diet composition that has been registered in 50% of the study population. Low physical disability was reported in 46.4% of patients and 28.6% of the patients were ambulant, self-dependent and capable of performing daily activities. This is correlated with the partially preserved cognitive functions of the patients in the current study.

C. Iron and Zinc Serum Levels in MS Patients and Controls

The mean iron serum level in MS patients and controls was 84.7 and 83.3 μg/dl respectively while the mean zinc serum level in MS patients and controls was 80.6 and 82.0 μg/dl respectively.

No significant difference in terms of iron (P=0.796) and zinc (P=0.640) serum levels was registered between MS patients and controls (Table 2).

D. Iron and Zinc Serum Levels in Different Groups according to Gender

The mean iron serum level in male of MS patients and controls was 94.0 and 92.7 μg/dl respectively while

Table 1. Characteristics of participants.

	Cases (n=27)	Controls (n=42)
Mean age ± SD	42.8 ± 12.9	38.3 ± 16.0
Male (mean age ± SD)	13 (46.8 ± 12.0)	13 (41.0 ± 19.8)
Female (mean age ± SD)	14 (38.1 ± 13.2)	29 (36.8 ± 13.7)
Disease course	RRMS	----------
EDSS mean	3.8	----------
MSSS mean	5.5	----------

EDSS: Expanded Disability Status Score; MSSS: Multiple Sclerosis Severity Score; SD: Standard Deviation; RRMS: relpasing-remitting multiple sclerosis

Table 2. Iron (Fe) and zinc (Zn) serum levels in MS patients and healthy individuals.

Metal serum level (μg/dl)	Cases (n=27)	Controls (n=42)	P-value	CI 95%
Fe (mean ± SD)	84.7 ± 35.1	83.3 ± 39.7	0.796	-17.35 ; 20.07
Zn (mean ± SD)	80.6 ± 15.2	82.0 ± 17.6	0.640	-9.71 ; 6.78

Table 3. Iron (Fe) serum level in different groups according to gender.

Iron serum level (μg/dl)	Cases (n=27)	Controls (n=42)	P-value	CI 95%
Male (mean ± SD)	94.0 ± 40.3	92.7 ± 31.8	0.758	-27.46 ; 29.88
Female (mean ± SD)	80.3 ± 30.2	79.2 ± 42.7	0.824	-27.52 ; 29.58

Table 4. Zinc serum level in different groups according to gender.

Zinc serum level (μg/dl)	Cases (n=27)	Controls (n=42)	P-value	CI 95%
Male (mean ± SD)	79.5 ± 17.2	83.8 ± 19.0	0.410	-18.76 ; 10.11
Female (mean ± SD)	81.7 ± 11.7	81.0 ± 16.9	0.766	-10.48 ; 12.00

the mean iron serum level in female of MS patients and controls was 80.3 and 79.2 μg/dl respectively.

No significant difference was shown between males (P=0.758) and females (P=0.824) of both groups (Table 3).

The mean zinc serum level in male MS patients and controls was 79.5 and 83.8 μg/dl respectively while the mean zinc serum level in female MS patients and controls was 81.7 and 81.0 μg/dl respectively.

No significant difference was noticed between males (P=0.410) and females (P=0.766) of both groups (Table 4).

DISCUSSION

Multiple sclerosis is an autoimmune neurodegenerative disease attacking brain and nerves [1]. Recently, many studies shed the light about the implication of minerals in the pathogenesis of the disease [22-24]. In our study, no significant difference was obtained between MS patients and healthy subjects in terms of iron and zinc serum levels and according to gender. Similar results were shown in studies performed on Greek, Italian and Egyptian populations [22,24-26]. However, other studies showed higher iron serum levels in MS patients compared to control groups [23,27-29]. Higher iron level in urine was also detected in MS group in comparison to control group [30]. In active MS lesions, iron is released from dying oligodendrocytes, leading to extracellular accumulation of iron in brain, causing symptoms as severe as iron deposit [31-33]. Hence, iron imbalance may be the cause of brain tissue inflammation leading to MS disease [26, 34]. Furthermore, brain iron excess levels contribute to oxidative stress with an abnormal iron metabolism leading also to iron accumulation causing neurodegeneration [35,36]. Generation of free radicals through the reduction of free iron causes lipid peroxidation and mitochondrial dysfunction [37]. Free radicals attack also the myelin sheath and axons [38]. Iron chelators, that bind free iron in the blood circulation, inhibit its entry in redox reactions [39].

On the other hand, many studies investigated zinc association with MS. Zinc levels were lower in serum of MS patients compared to controls [28,40,41]. Other studies done in Sweden and Iran reported a higher level of zinc in MS patients in comparison to controls [40]. Dore-Duffy et al. showed that zinc serum level was significantly higher only in male MS patients [42]. In our study, no significant difference was observed in zinc serum levels between MS patients and controls. Similar results were obtained in different studies [43-47]. Findings showed that zinc deficiencies induce apoptosis in neural cells while high zinc levels exert neurotoxic effect [48,49].

Zinc low levels reduce distinct functions in the immune system including T-helper cell function, peripheral T-cell number, NK-cell and cytotoxic T-cell activities, and neutrophil and macrophage functions [50]. Zinc deficiency is directly involved in the exacerbation of MS. In fact, a decreased level of intracellular zinc can activate dendritic cell maturation that results in increased of costimulatory molecules and cell-surface expression of MHC, leading probably to the activation of T-cells and T-cell mediated autoimmunity [51]. High levels of zinc alone allow the over-expression of the high-affinity receptor for IL-2, essential for the proliferation and differentiation of CD4+

and CD8+ lymphocytes to effector cells, in addition to microglia activation which are the residential immune cells of the CNS playing an important role in the pathogenesis of MS [52]. Matrix metalloproteinases zinc dependent are involved in the remodeling of the extracellular matrix like the degradation of BBB [53] and myelin basic protein (MBP) [54], conferring a potential important function in the pathogenesis of MS from one side and in the development of new treatments for MS from another side [55].

CONCLUSION

Our findings do not show significant correlation between serum iron and zinc levels and MS disease in Lebanese population living in Bekaa and Tripoli cities confirming other studies done on other populations. They do not contradict the contribution of iron and zinc in the disease but show them as non-reliable MS biomarkers. Measurements of these two metals in other body fluids and biological tissues as well as the recruitment of a larger number of participants in other Lebanese regions is needed for a better understanding of the involvement of these metals with the pathogenesis of MS.

ACKNOWLEDGEMENTS

The authors are grateful to the MS patients and controls without whom this study would not have been possible. We also thank the laboratory medical stuff in Al-Abdallah hospital-Riyak and Al-Makarem Primary Health Care especially Mrs. Diala Shalhoub, Siham Mazbouh and Rahmeh Nasser, for facilitating sample collection and Dr. Noura Yassine for her assistance in statistical analyses.

REFERENCE

1. Cooper GS, Stroehla BC. The epidemiology of autoimmune diseases. Autoimmunity reviews. 2003; 2(3):119-25.
2. Milo R, Kahana E. Multiple sclerosis: geoepidemiology, genetics and the environment. Autoimmunity reviews. 2010;9(5):A387-94.
3. Mehta BK. New hypotheses on sunlight and the geographic variability of multiple sclerosis prevalence. Journal of the neurological sciences. 2010;292(1-2):5-10.
4. Zhang Y, Zhang J, Liu H, He F, Chen A, Yang H, et al. Meta-analysis of FOXP3 gene rs3761548 and rs2232365 polymorphism and multiple sclerosis susceptibility. Medicine. 2019;98(38):e17224.
5. Sadovnick D. The place of environmental factors in multiple sclerosis: Genes, environment and the interactions thereof in the etiology of multiple sclerosis. Revue neurologique. 2019.
6. Serafini B, Rosicarelli B, Veroni C, Mazzola GA, Aloisi F. Epstein-Barr virus-specific CD8 T cells selectively infiltrate the multiple sclerosis brain and interact locally with virus infected cells: clue for a virus-driven immunopathological mechanism. Journal of virology. 2019.
7. Riccio P, Rossano R. Nutrition facts in multiple sclerosis. ASN neuro. 2015;7(1).
8. Mezzaroba L, Alfieri DF, Colado Simao AN, Vissoci Reiche EM. The role of zinc, copper, manganese and iron in neurodegenerative diseases. Neurotoxicology. 2019;74:230-41.
9. Maiuolo J, Macri R, Bava I, Gliozzi M, Musolino V, Nucera S, et al. Myelin Disturbances Produced by Sub-Toxic Concentration of Heavy Metals: The Role of Oligodendrocyte Dysfunction. International journal of molecular sciences. 2019;20(18).
10. Melo TM, Larsen C, White LR, Aasly J, Sjobakk TE, Flaten TP, et al. Manganese, copper, and zinc in cerebrospinal fluid from patients with multiple sclerosis. Biological trace element research. 2003;93(1-3):1-8.
11. Kennedy DO, Stevenson EJ, Jackson PA, Dunn S, Wishart K, Bieri G, et al. Multivitamins and minerals modulate whole-body energy metabolism and cerebral blood-flow during cognitive task performance: a double-blind, randomised, placebo-controlled trial. Nutrition & metabolism. 2016;13:11.
12. Majewski M, Kozlowska A, Thoene M, Lepiarczyk E, Grzegorzewski WJ. Overview of the role of vitamins and minerals on the kynurenine pathway in health and disease. Journal of physiology and pharmacology : an official journal of the Polish Physiological Society. 2016;67(1):3-19.
13. Frederickson CJ. Neurobiology of zinc and zinc-containing neurons. International review of neuro-

biology. 1989;31:145-238.
14. Kimura T, Kambe T. The Functions of Metallothionein and ZIP and ZnT Transporters: An Overview and Perspective. International journal of molecular sciences. 2016;17(3):336.
15. Choi BY, Jung JW, Suh SW. The Emerging Role of Zinc in the Pathogenesis of Multiple Sclerosis. International journal of molecular sciences. 2017; 18(10).
16. Noh KM, Koh JY. Induction and activation by zinc of NADPH oxidase in cultured cortical neurons and astrocytes. The Journal of neuroscience : the official journal of the Society for Neuroscience. 2000;20(23):RC111.
17. Radlowski EC, Johnson RW. Perinatal iron deficiency and neurocognitive development. Frontiers in human neuroscience. 2013;7:585.
18. Sheykhansari S, Kozielski K, Bill J, Sitti M, Gemmati D, Zamboni P, et al. Redox metals homeostasis in multiple sclerosis and amyotrophic lateral sclerosis: a review. Cell death & disease. 2018;9(3):348.
19. Ge Y, Jensen JH, Lu H, Helpern JA, Miles L, Inglese M, et al. Quantitative assessment of iron accumulation in the deep gray matter of multiple sclerosis by magnetic field correlation imaging. AJNR American journal of neuroradiology. 2007;28(9):1639-44.
20. Yamout B, Barada W, Tohme RA, Mehio-Sibai A, Khalifeh R, El-Hajj T. Clinical characteristics of multiple sclerosis in Lebanon. Journal of the neurological sciences. 2008;270(1-2):88-93.
21. Stankiewicz J, Panter SS, Neema M, Arora A, Batt CE, Bakshi R. Iron in chronic brain disorders: imaging and neurotherapeutic implications. Neurotherapeutics: the journal of the American Society for Experimental NeuroTherapeutics. 2007;4(3):371-86.
22. Rieder HP, Schoettli G, Seiler H. Trace elements in whole blood of multiple sclerosis. European neurology. 1983;22(2):85-92.
23. Forte G, Visconti A, Santucci S, Ghazaryan A, Figa-Talamanca L, Cannoni S, et al. Quantification of chemical elements in blood of patients affected by multiple sclerosis. Annali dell'Istituto superiore di sanita. 2005;41(2):213-6.
24. Visconti A, Cotichini R, Cannoni S, Bocca B, Forte G, Ghazaryan A, et al. Concentration of elements in serum of patients affected by multiple sclerosis with first demyelinating episode: a six-month longitudinal follow-up study. Annali dell'Istituto superiore di sanita. 2005;41(2):217-22.
25. Sfagos C, Makis AC, Chaidos A, Hatzimichael EC, Dalamaga A, Kosma K, et al. Serum ferritin, transferrin and soluble transferrin receptor levels in multiple sclerosis patients. Multiple sclerosis. 2005;11(3):272-5.
26. Abo-Krysha N, Rashed L. The role of iron dysregulation in the pathogenesis of multiple sclerosis: an Egyptian study. Multiple sclerosis. 2008;14(5):602-8.
27. Johnson S. The possible role of gradual accumulation of copper, cadmium, lead and iron and gradual depletion of zinc, magnesium, selenium, vitamins B2, B6, D, and E and essential fatty acids in multiple sclerosis. Medical hypotheses. 2000;55(3):239-41.
28. Alimonti A, Ristori G, Giubilei F, Stazi MA, Pino A, Visconti A, et al. Serum chemical elements and oxidative status in Alzheimer's disease, Parkinson disease and multiple sclerosis. Neurotoxicology. 2007;28(3):450-6.
29. Craelius W, Migdal MW, Luessenhop CP, Sugar A, Mihalakis I. Iron deposits surrounding multiple sclerosis plaques. Archives of pathology & laboratory medicine. 1982;106(8):397-9.
30. Exley C, Mamutse G, Korchazhkina O, Pye E, Strekopytov S, Polwart A, et al. Elevated urinary excretion of aluminium and iron in multiple sclerosis. Multiple sclerosis. 2006;12(5):533-40.
31. Hametner S, Wimmer I, Haider L, Pfeifenbring S, Bruck W, Lassmann H. Iron and neurodegeneration in the multiple sclerosis brain. Annals of neurology. 2013;74(6):848-61.
32. Haacke EM, Garbern J, Miao Y, Habib C, Liu M. Iron stores and cerebral veins in MS studied by susceptibility weighted imaging. International angiology : a journal of the International Union of Angiology. 2010;29(2):149-57.
33. Burgetova A, Seidl Z, Krasensky J, Horakova D, Vaneckova M. Multiple sclerosis and the accumulation of iron in the Basal Ganglia: quantitative assessment of brain iron using MRI t(2) relaxometry. European neurology. 2010;63(3):136-43.

34. Mills E, Dong XP, Wang F, Xu H. Mechanisms of brain iron transport: insight into neurodegeneration and CNS disorders. Future medicinal chemistry. 2010;2(1):51-64.
35. Zecca L, Youdim MB, Riederer P, Connor JR, Crichton RR. Iron, brain ageing and neurodegenerative disorders. Nature reviews Neuroscience. 2004; 5(11): 863-73.
36. Schneider SA, Hardy J, Bhatia KP. Iron accumulation in syndromes of neurodegeneration with brain iron accumulation 1 and 2: causative or consequential? Journal of neurology, neurosurgery, and psychiatry. 2009;80(6):589-90.
37. Halliwell B. Oxidative stress and neurodegeneration: where are we now? Journal of neurochemistry. 2006; 97(6):1634-58.
38. Sjogren B, Iregren A, Frech W, Hagman M, Johansson L, Tesarz M, et al. Effects on the nervous system among welders exposed to aluminium and manganese. Occupational and environmental medicine. 1996; 53(1):32-40.
39. Fulgenzi A, Zanella SG, Mariani MM, Vietti D, Ferrero ME. A case of multiple sclerosis improvement following removal of heavy metal intoxication: lessons learnt from Matteo's case. Biometals : an international journal on the role of metal ions in biology, biochemistry, and medicine. 2012;25(3):569-76.
40. Palm R, Hallmans G. Zinc and copper in multiple sclerosis. Journal of neurology, neurosurgery, and psychiatry. 1982;45(8):691-8.
41. Ghazavi A, Kianbakht S, Ghasami K, Mosayebi G. High copper and low zinc serum levels in Iranian patients with multiple sclerosis: a case control study. Clinical laboratory. 2012;58(1-2):161-4.
42. Dore-Duffy P, Catalanotto F, Donaldson JO, Ostrom KM, Testa MA. Zinc in multiple sclerosis. Annals of neurology. 1983;14(4):450-4.
43. Ho SY, Catalanotto FA, Lisak RP, Dore-Duffy P. Zinc in multiple sclerosis. II: Correlation with disease activity and elevated plasma membrane-bound zinc in erythrocytes from patients with multiple sclerosis. Annals of neurology. 1986;20(6):712-5.
44. Kapaki E, Segditsa J, Papageorgiou C. Zinc, copper and magnesium concentration in serum and CSF of patients with neurological disorders. Acta neurologica Scandinavica. 1989;79(5):373-8.
45. Smith DK, Feldman EB, Feldman DS. Trace element status in multiple sclerosis. The American journal of clinical nutrition. 1989;50(1):136-40.
46. Giacoppo S, Galuppo M, Calabro RS, D'Aleo G, Marra A, Sessa E, et al. Heavy metals and neurodegenerative diseases: an observational study. Biological trace element research. 2014;161(2):151-60.
47. Kazmierczak K, Malukiewicz G, Lesiewska-Junk H, Laudencka A, Szady-Grad M, Klawe J, et al. Blood plasma levels of microelements in patients with history of optic neuritis. Current eye research. 2014;39(1):93-8.
48. Yokoyama M, Koh J, Choi DW. Brief exposure to zinc is toxic to cortical neurons. Neuroscience letters. 1986;71(3):351-5.
49. Adamo AM, Zago MP, Mackenzie GG, Aimo L, Keen CL, Keenan A, et al. The role of zinc in the modulation of neuronal proliferation and apoptosis. Neurotoxicity research. 2010;17(1):1-14.
50. Rink L, Gabriel P. Zinc and the immune system. The Proceedings of the Nutrition Society. 2000;59(4):541-52.
51. Rink L, Haase H. Zinc homeostasis and immunity. Trends in immunology. 2007;28(1):1-4.
52. Higashi Y, Segawa S, Matsuo T, Nakamura S, Kikkawa Y, Nishida K, et al. Microglial zinc uptake via zinc transporters induces ATP release and the activation of microglia. Glia. 2011;59(12):1933-45.
53. Kurzepa J, El-Demerdash FM, Castellazzi M. Matrix Metalloproteinases as a Pleiotropic Biomarker in Medicine and Biology. Disease markers. 2016; 2016: 9275204.
54. Shiryaev SA, Savinov AY, Cieplak P, Ratnikov BI, Motamedchaboki K, Smith JW, et al. Matrix metalloproteinase proteolysis of the myelin basic protein isoforms is a source of immunogenic peptides in autoimmune multiple sclerosis. PloS one. 2009;4(3):e4952.
55. Ram M, Sherer Y, Shoenfeld Y. Matrix metalloproteinase-9 and autoimmune diseases. Journal of clinical immunology. 2006;26(4):299-307.