

DOI: 10.5455/medarh.2012.66.s37-s40

Med Arh. 2012 Jun; 66(3, suppl 1): 37-40

Received: May 15th 2012

Accepted: June 20th 2012

CONFLICT OF INTEREST: NONE DECLARED

ORIGINAL PAPER

Role of Early Viral Infections in Development of Multiple Sclerosis

Jasminka Djelilovic - Vranic, Azra Alajbegovic

Neurology Clinic, Clinical Center of University of Sarajevo, Sarajevo, Bosnia and Herzegovina

Introduction: Multiple sclerosis is a chronic inflammatory, autoimmune, demyelinating, disease but also degeneration of axons, with mainly progressive course, causing greater or lesser degree of disability. In addition to genetic predisposition the environmental factors, with particular importance of early viral infection, have an essential role in the development of MS. These are called long-acting viruses that remain hidden in the body for years by encouraging latent immunological changes in the body, eventually resulting in autoimmune demyelination and the appearance of disease symptoms, which confirms the high titer of antibodies to certain viruses in patients with the MS. To first of all herpes simplex virus, Epstein Barr virus, cytomegalovirus and rubella virus. **Goal:** Goal of this study is to analyze the incidence of early infection with rubella virus, herpes simplex, cytomegalovirus and Epstein-Barr, in MS patients using titers of IgG and IgM antibodies. **Material and methods:** The study included patients treated at the Neurology Clinic in Sarajevo, with a diagnosis of multiple sclerosis (newly discovered) in the period January 2009 - December 2011. To all patients beside history and neurological examination and tests to confirm the MS (brain MRI, evoked potentials and CSF examination) made serological tests for viruses, HSV, Rubella virus, cytomegalovirus and Epstein-Barr's virus, with reference to the previous parameters (old) and new viral infection. **Results:** In this period there were 118 newly diagnosed multiple sclerosis from which 69.5% (82) female and 30.5% (36) male patients aged 23-56 years. IgG antibodies to herpes simplex virus was positive in 93.2% (110 patients) (72 F and 38 M and IgM only in 0.84% (1 patient). Ig G in Cytomegalovirus was positive in 86.44% (102 subjects, 71 females and 31 males), while IgM was negative in whole sample. IgG Rubella virus was positive in 61.01% (72 patients, 52 F and 20 M) and IgM was negative in all, while IgG in Epstein-Barr's virus was positive in 83% (98 patients). **Conclusion.** Early infection by herpes simplex virus, cytomegalovirus, Epstein-Barr and Rubella is present in patients with multiple sclerosis in a significant number so the conclusion is the fact that in the development of multiple sclerosis an important role early exposure to these viruses. **Key words:** early viral infection, multiple sclerosis.

Corresponding author: Prof Jasminka Djelilovic-Vranic, MD, PhD. Neurology clinic, Clinical center of Sarajevo University, Bosnia and Herzegovina. Sarajevo, Bolnicka 25.

1. INTRODUCTION

Multiple sclerosis is an inflammatory, autoimmune disease, mainly of white mass of the brain, which some-

times may involve the gray matter, primarily subcortical and those in the anterior horns of the spinal cord (1, 2). Today from this disease in the world pop-

ulation suffers about 2.5 million people, with a tendency of increase in the number of patients. Prevalence increases with the distance from the equator.

In the etiology of MS occurrence are noted early viral infections, genetic predisposition and environmental factors. (3)

Viruses that are brought in connection with the development of multiple sclerosis are: family of DNA viruses (herpes simplex virus, human type 6, Epstein Barr's, Cytomegalovirus) and RNA viruses the Rubella virus.

Herpes simplex virus is a DNA virus from the family subfamily Alpha-herpesvirinae Herpesviridae with size of 110-120 nm. It multiplies in the nucleus and cytoplasm of infected cells. Herpes simplex virus infections occur during the first years of life, passing mostly as latent, asymptomatic and result in the presence of virus in sensory ganglia n. Trigeminal where they remain present throughout life (4).

IgM antibody titer which is taken as a positive is titer greater than 0.200 IU/M and IgG antibodies 1:231 IU/M. 70-80% of patients with MS have antibodies for HSV type I. A study of American Pathologists, which was performed on brain tissue of deceased persons with a diagnosis of MS, confirmed the existence of this virus in the demyelinating plaques in 37%. Human herpes simplex virus type 6A is DNA from subfamily Betaherpesvirinae-

rus size 160-210 nm. It is now considered one of the cofactors in the development of MS especially in time of relapse. A study of American pathologists, which was done on brain tissue of deceased persons with a diagnosis of MS, confirmed the existence of this virus in the demyelinating plaques in 32% of respondents (5, 6, 8).

Cytomegalovirus is a DNA virus of the subfamily Betaherpesvirinae size of 150-200 nm. The primary infection is usually unnoticed with unapparent latent retention in the parenchymatous organs and cells of the salivary glands. 50-74% of the world's population has positive IgG antibodies to this virus. Titers of IgM antibodies that are taken as positive for this virus is greater than 0.200, and IgG 1.231. In the active infections may develop infectious mononucleosis syndrome. CMV infection usually occurs between 10 and 35 year of life. The virus can pass through the blood-brain barrier in two ways, directly and indirectly as a viral load over the cells. A study of American pathologists, which was done on brain tissue of deceased persons with a diagnosis of MS, confirmed the existence of this virus in the demyelinating plaques in 16% of respondents and elevated titers of CMV antibodies are present in 47% of patients (4, 9).

Epstein-Barr virus is a DNA virus of the subfamily Gamaherpesvirinae size of 150-170 nm. Acute viral infections can pass unnoticed and go into a latent phase of infection, when viral genome remains in the infected B lymphocytes. Acute infection of EB virus is expressed as infectious mononucleosis. About 90% of the population (between 50 and 100%) has signs of the old EB virus infection. A study of American pathologists, which was done on brain tissue of deceased persons with a diagnosis of MS, confirmed the existence of this virus in the demyelinating plaques in 27% of respondents, but in 87% of patients who suffer from MS elevated titers of antibodies to this virus and that the average time of illness after infection with the virus was 10 years. In the peripheral blood of patients suffering from MS, by specific staining were detected in T lymphocytes specific antibodies to EB virus. (4,7)

Rubella is virus from the RNA group. Research shows that this virus can be associated with MS patients in 20 to 60% of patients. A positive IgM antibody titer is greater than 0.200 and IgG > 4 IU/M (4,9).

2. GOAL

To analyze the incidence of early infection by herpes simplex virus, cytomegalovirus, Epstein-Barr and rubella, in MS patients using IgG titer and the titer of any of fresh IgM antibodies.

3. MATERIAL AND METHODS

In this paper we analyzed the patients, newly diagnosed with Multiple Sclerosis in a two-year period January 2009-December 2011 to which during hospitalization besides medical history and neurological examination was performed brain MRI, evoked potentials, cerebrospinal fluid examination and serological tests for viruses, HSV, cytomegalovirus, Epstein-Barr's and Rubella virus, with reference to the previous parameter (old) or new viral infections, as well as the possible impact of the MS occurrence.

4. RESULTS

During the two-year period was discovered a total of 118 new patients with a diagnosis of multiple sclerosis during the first or second hospitalization

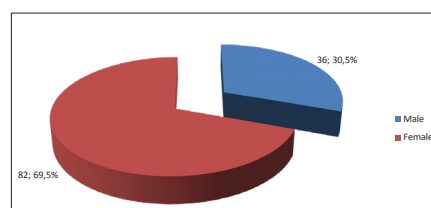


Figure 1. Gender structure of patients suffering from Multiple Sclerosis

from which 82 (69.5%) female and 36 (30.5%) male.

The average age of patients with multiple sclerosis was 36.529 years, for the group of men 36.5 years and for women 35 years. The largest number of patients, 56.90% of them had been in the age group from 20-39 years.

Analysis of viral infection is followed by a parameter IgG (old) and IgM (fresh) infections. We can see that there is a significant difference between the number of patients with positive IgG

	Positive	Negative	Total
N	110	8	118
Percent (%)	93.22%	6.77%	100,0%
$\chi^2=60.229$ $p<0.001$			

Table 1. Relation between the number of patients with and without HSV IgG antibodies present

	IgM -	IgM +	IgG -	IgG+
Male	33	0	5	28
Female	85	0	11	74
Total	118	0	16	102

Table 2: The results of serological tests for CMV

	IgG	IgM
Positive	102	0
Negative	16	118
$p<0.001$ $\chi^2=102.508$		

Table 3: The frequency of IgG and IgM CMV antibodies in patients with MS

	IgM -	IgM +	IgG -	IgG NT
Male	36	0	4	24 5
Female	82	0	12	48 9
Total	118	0	32	72 14
$p<0.001$ $\chi^2=107.127$				

Table 4: The results of serological tests for Rubella virus

and IgM antibodies, or that positive IgG antibody occurring in a significant number of patients ($\chi^2=105,245$; $p<0,001$). The table shows that there is a significant difference in the percentage of patients with multiple sclerosis who have a positive titer of IgG antibodies to HSV compared to those with negative, or a positive titer of IgG antibodies is found in a significant number of patients suffering from multiple sclerosis, compared to patients with negative IgG antibody titer.

Infection with this type of virus was equally present in men and women.

The largest number of patients

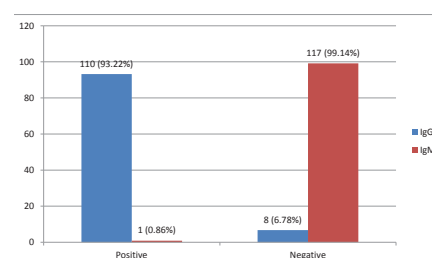


Figure 2. The frequency of IgG and IgM antibodies in patients with HSV MS

	IgM -	IgM +	IgG -	IgG +	NT
Male	46	0	2	30	1
Female	58	0	4	58	13
Total	104	0	6	98	14

Table 5: The results of serological tests for Epstein Barr virus

	IgG	IgM
Positive	98	0
Negative	6	104
p=0.000411		

Table 6: The frequency of IgG and IgM antibodies to Epstein Barr virus in patients with MS

	HSV	Cytomegalovirus
Positive	93.2%	86.44%
Negative	6.8%	13.56%
p=1,000 $\chi^2=0$		
	Epstein-Barr virus (n=83)	Rubella (n=83)
Positive	83.00	61.01%
Negative	5.00	26.09%
p=1.000 $\chi^2=0$		

Table 7: patient ratio and the presence of IgG antibodies to different viruses

tested for HSV or 56.63% of them are in the age group of 20-39 years. In this group there is also the largest number of IgG + patients, 52.24%

The Table shows that after testing on cytomegalovirus positive IgM antibodies were not found, while 102 (86.44%) patients had positive IgG antibodies.

The Table shows that there is a significant difference between the number of patients with positive IgG and IgM antibodies to cytomegalovirus, or that positive IgG antibody are found in a significant number of patients (86,44%), mostly aged 20-39 years (45%) The Table shows that IgM antibodies to Rubella virus did not have a single patient, while 72 (66.01%) patients had positive IgG antibodies with fact that 14 (11.86%) was not tested.

There is a significant difference between the number of patients who have a positive IgG and IgM antibodies to Rubella virus, or that positive IgG antibody occurring in a significant number of patients, mostly aged 20-39 years (42.4%).

Epstein-Barr virus

The Table shows that of 104 patients tested for Epstein-Barr virus IgM antibody none had positive result, while

98 (83.00%) patients had positive IgG antibodies.

The table shows that there is a significant difference between the number of patients with positive IgG and IgM antibodies to Epstein Barr virus, as well as statistically significant difference in the number of IgG positive and negative patients.

Infection by this virus type was equally represented in both men and women.

Usually aged 20-39 years (47.2%).

The Table shows that there was no statistically significant differences in the number of patients with positive IgG antibodies to the virus observed (noting that 12% are not tested for rubella and Epstein-Barr virus).

5. DISCUSSION

Multiple sclerosis is a disease which in 70% of cases occurring between 20 and 40 years of age and can occur at the age from 10 to 60 years. Women two times more likely than men suffer from MS.

In our study from 118 patients there were 69.5% of women with newly diagnosed MS compared to the 30.5% men, which is in accordance with other author's findings. The average age of patients with multiple sclerosis was 36.529 years. There was no significant difference between the average age for women and men suffering from multiple sclerosis ($p=0.687$).

The largest number of patients was in the group between 20 and 39 years (56.90%) while in a group of 40-59 years was 41.18% patients. The average duration of illness of all patients was 1.451 years, one year for men and for women 1.605 years. There was no statistically significant difference in the average duration of illness between the sexes ($p=0.369$). Our results correlate with studies of other authors (10).

Positive IgG antibodies to herpes simplex virus in our study had 93.2% of patients, and only one patient (0.84%) had positive IgM antibodies. There was a statistically significant difference in the incidence of the above ($p<0.001$, $\chi^2=105.245$). Also, there is a significant difference in the percentage of patients who were positive and those who had negative findings, positive findings had

a significantly higher percentage of patients ($p<0.001$, $\chi^2=60.229$). There was no difference in gender distribution of IgG antibodies ($p=0.873$, $\chi^2=0.027$) and the highest percentage of patients with positive HSV IgG antibody found in the age group 20-39 years 52.24% (11,12).

According to the literature, early viral infection with herpes simplex virus in patients with multiple sclerosis has been found in 70-80% of cases and in the general population in about 50% and some say up to 70%. Somewhat higher percentage of early viral infection detected in herpes simplex virus in our population could explain the low standard of living and continuously exposure to stress of our population, which contributes to global modification of the immune system and weaken the body's defense system (13).

Positive IgG antibodies to cytomegalovirus had in our sample 86.44% of patients and there were those with positive IgM antibody titers while there was a statistically significant difference in the incidence of the above ($p<0.001$, $\chi^2=102.508$). Also, there is a significant difference in the percentage of patients who were positive and those who had negative findings, or positive results are observed in a significant percentage of patients. ($p<0.001$, $\chi^2=55.525$). There was no difference in gender distribution of IgG antibodies ($p=0.097$, $\chi^2=0$), while the largest number of patients with positive IgG antibodies to CMV was in the age group of 20-39 years—45%.

According to research by other authors positive IgG antibodies to CMV occur in 40-74% of the world's population, which refers to the general population, while the study of association of early viral infection with cytomegalovirus and the occurrence of multiple sclerosis suggest that early viral infections with cytomegalovirus in patients with multiple sclerosis is present in about 50% of cases.

In our sample there is greater representation of early cytomegalovirus infection (86,44%) which could also be explained by the global social situation and constant stress, which contributes to the modification of the immune defense system and the reduced ability of organisms to fight infection (14).

In our study positive for IgG anti-

bodies to Rubella virus had 61.01% of patients, and none positive for IgM antibodies, so there was a statistically significant difference in the incidence of the above ($p < 0.001$, $\chi^2 = 107.127$). There is a significant difference in the percentage of patients who were positive and those who had negative findings, or positive finding is observed in a significant percentage of patients ($p < 0.001$, $\chi^2 = 55.570$). There was no difference in gender distribution of IgG antibodies ($p = 0.086$, $\chi^2 = 0.017$), while the largest number of patients with positive IgG antibodies to rubella was in the age group of 20-39 years—42.4%.

According to research by other authors signs of infection with the Rubella virus are present in 30–80% of respondents in the general population and in 20-60% of patients with MS, which correlates with our results. It should be noted that implementation of immunization also contributes to the prevalence of rubella antibodies (15).

For the Epstein-Barr virus in our study, elevated titers of IgG antibodies was observed in 83.00% of patients, the percentage is much higher than the number of negative patients and no patient had a positive IgM antibody, which in this case indicates a statistically significant difference in frequency of IgG and IgM antibodies in seropositive patients ($p < 0.0047$, $\chi^2 = 8.001$). There were no significant differences in gender distribution ($p = 0.001$) and most patients that were serologically positive in this case had between 20 and 39 years - 47.2%.

According to the literature data, infection with Epstein Barr virus is found in 50-100% of persons in the general population and in 87% of patients with MS so our results confirm the presence of this virus in 83% of patients with multiple sclerosis is fully in accordance of results by other researchers (16,17).

Mutual comparison of positive IgG findings for each particular virus did not show statistically significant difference in the appearance of any in particular, which points to the fact that none of them by itself is significant, but ob-

served together, these viruses have a significant impact and certainly contribute to the development of multiple sclerosis. We did not observed any gender differences in the incidence of any form of virus and age groups are correlated with the age groups of patients with MS.

The data from the literature shows that in the general population positive antibodies to individual viruses occur in a significant number, over 95%, which would mean that the viral infection may be associated with the development of multiple sclerosis. Taking into account the fact that statistically significant in the first place are IgG antibodies over IgM type, it can be said that it was an earlier, rather than acute viral infection as an aetiological factor of the disease (6,17).

A positive family history, which is predisposing factor for multiple sclerosis, according to our study was confirmed in 14% of patients, which is in accordance with findings of other authors, according to which this percentage ranges from 3-35% (18,19).

6. CONCLUSION

Women more often suffer from MS than men, and mostly at age of 30-40 years. A positive IgG antibody as a sign of early infection for HSV had 93.2% patients, the CMV 86.44%, to rubella 61.01%, and 83% on EBV. A family predisposition was found in 13.73% patients. Early viral infections with viruses HS, cytomegalovirus, rubella, and Epstein-Barr, is certainly an important factor in the development of multiple sclerosis.

REFERENCES

1. Kantardžić Dž. i sar. Klinička neurologija. Svjetlost, Sarajevo, 2001.
2. Reiber H, Ungefer S, Jacobi C. The intrathecal, polyspecific and oligoclonal immune response in multiple sclerosis, 12 april 2011.
3. World Health Organizations: Atlas Multiple sclerosis resources in the world, 2008.
4. Zvizdić Š. Opća medicinska virusologija, univerzitetski udžbenik, Sarajevo, 2002.
5. Alvarez-Lafuente R, García-Montojo M, De Las Heras V, Domínguez-Mozo MI, Bartolome M, Benito-Martin MS, Arroyo R. Herpes viruses and human endogenous

- retroviral sequences in the cerebrospinal fluid of multiple sclerosis patients, 2008 Jun;14; 595-601.
6. A Nationwide, Population-Based Study Jinn-Hong Kang I, Jau-Juan Sheu, Senyeong Kao and Heng-Ching Lin. Increased Risk of Multiple Sclerosis Following Herpes Zoster, The Journal of infectious diseases. 07/2011; 204(2): 188-192.
7. Lowry F. Herpes Zoster Boosts Risk for Multiple Sclerosis, [Published online June 7, 2011](#).
8. Garcia-Montojo M, De Las Heras V, Dominguez-Mozo M, Bartolome M, Garcia-Martinez MA, Arroyo R, Alvarez-Lafuente R; On behalf of the HHV-6 and Multiple Sclerosis Study Group: Human herpesvirus 6 and effectiveness of interferon beta 1b in multiple sclerosis patients, 25 april 2011.
9. Perron H, Bernard C, Bertrand JB, Lang AB, Popa I, Sanhadji K, Portoukalian J. JNeurol Sci. [Endogenous retroviral genes, Herpes-viruses and gender in Multiple Sclerosis](#). 2009 Nov 15; 286(1-2): 65-72.
10. Cassels C. Multiple Sclerosis Prevalence Increasing Faster Among Women Than Men April 24, 2007.
11. Howard M, Sellors JW, Jang D, Robinson NJ, Fearon M, Kaczorowski J, Chernesky M. Regional Distribution of Antibodies to Herpes Simplex Virus Type 1 (HSV-1) and HSV-2 in Men and Women in Ontario, Canada 2003.
12. Malkin JE, Morand P, Malvy D, Ly TD, Chanzy B, de Labareyre C, El Hasnaoui A, Hercberg S. Seroprevalence of HSV-1 and HSV-2 infection in the general French population, 2002; 78: 201-203.
13. Vyse AJ, Gay NJ, Slomka MJ, Gopal R, Gibbs T, Morgan-Capner P, Brown DW. The burden of infection with HSV-1 and HSV-2 in England and Wales: implications for the changing epidemiology of genital herpes, 2000; 76: 183-187.
14. Sotgiu S, Arru G, Mameli G, Serra C, Pugliatti M, Rosati G, Dolei A. Multiple sclerosis-associated retrovirus in early multiple sclerosis: a six-year follow-up of a Sardinian cohort, 2006 Dec; 12(6): 698-703.
15. Sever JL, Schiff GM, Bell JA, Kapikian AZ, Huebner RJ, Traub RG. Rubella: frequency of antibody among children and adults, 2009.
16. Lassmann H, Niedobitek G, Aloisi F, Middelorp JM, The NeuropoMiSe EBV Working Group: Epstein - Barr virus in the multiple sclerosis brain: a controversial issue - report on a focused workshop held in the Centre for Brain Research of the Medical University of Vienna, Austria, June 25, 2011.
17. Jilek S, Schluep M, Meylan P, Vingerhoets F, Guignard L, Monney A, Kleeberg J, Le Goff G, Pantaleoand G, Du Pasquier RA. Strong EBV - specific CD8+ T-cell response in patients with early multiple sclerosis, May 9, 2008.
18. Cassels C. Men and Women Equally Transmit Genetic Risk for MS to Offspring 2007.
19. [Authors and Disclosures](#): Genetics and Natural History of Multiple Sclerosis: Genetics of Multiple Sclerosis, 2008,