ScientificScholar®
Knowledge is power
Publisher of Scientific Journals

www.surgicalneurologyint.com

Surgical Neurology International

Editor-in-Chief: Nancy E. Epstein, MD, Professor of Clinical Neurosurgery, School of Medicine, State U. of NY at Stony Brook.

SNI: Infection

Editor
Ali Akhaddar, MD, IFAANS
Avicenne Military Hospital, Marrakech, Morocco

SNI
Open Access

Review Article

Epstein–Barr virus flare: A multiple sclerosis attack

Ali Naser Msheik[1], Zeinab Al Mokdad[2], Faten Hamed[3], Farah Assi[4], Ali Jibbawi[5], Jean-Pierre Saad[6], Rami Mohanna[6], Anthony Khoury[7], Mhamad Farhat[6], Rami Atat[8]

[1]Department of Neurosurgery, Faculty of Medical Sciences, [2]Masters In Medical Ethics, Registered Nurse, Lebanese University, [3]Department of Pharmacology, Lebanese International University, [4]Department of Infectious Diseases, Lebanese University, Beirut, [5]Department of Pediatrics, Saint Georges Hospital, Hadath, [6]Department of General Medicine, Saint Joseph University, [7]Department of General Medicine, [8]Professor of Neurology, Faculty of Medical Sciences, Lebanese University, Beirut, Lebanon.

E-mail: *Ali Naser Msheik - dr.alimsheik@gmail.com; Faten Hamed - dr.alimsheik@gmail.com; Farah Assi - dr.alimsheik@gmail.com; Ali Jibbawi - dr.alimsheik@gmail.com; Jean-Pierre Saad - dr.alimsheik@gmail.com; Rami Mohanna - dr.alimsheik@gmail.com; Anthony Khoury - dr.alimsheik@gmail.com; Mhamad Farhat - dr.alimsheik@gmail.com; Zeinab Al Mokdad - dr.alimsheik@gmail.com; Rami Atat - dr.alimsheik@gmail.com

***Corresponding author:**
Ali Naser Msheik,
Department of Neurological Surgery, Lebanese University, Lebanon.

dr.alimsheik@gmail.com

Received: 11 June 2024
Accepted: 24 July 2024
Published: 04 October 2024

DOI
10.25259/SNI_457_2024

Quick Response Code:

ABSTRACT

Background: Multiple sclerosis (MS)-Epstein–Barr virus (EBV) relation is similar to doing a complicated puzzle: it consists of many pieces that become more and more clear as the issue is viewed from different sides. Based on the research findings, there is powerful evidence that EBV and MS have a strong relation where high levels of EBV DNA are able to be shown in all the spinal cord and the blood of the MS patients, but these are shown during disease relapses, and this implies a role in these illnesses. It kind of narrows the choices that you have to look for, just like how gathering evidence can lead to finding the missing person. In the analysis, new ways of EBV participation in MS progression are expected to be installed, and even new therapeutics are expected to be made.

Methods: A comprehensive literature search of PubMed was conducted until November 2023 to identify studies investigating the association between Epstein-Barr virus (EBV) infection and multiple sclerosis (MS). Only articles that met stringent criteria, including validation of EBV infection through laboratory testing, were included in the analysis.

Results: A total of 16 articles were identified as applicable for the background review, and this conformed with the discovery that the initiation of EBV/IM was consistent across various studies, namely, retrospective, cross-sectional, or prospective. The statistics reveal a glimpse into the need for prolonged research in studying the pattern of this link between EBV and MS. Novel treatment approaches targeting EBV, including adoptive T-cell therapy and gene-based immunotherapy, show promise in mitigating MS progression by targeting EBV-infected cells.

Conclusion: Clinical trials investigating antiviral therapies and vaccination strategies are underway, aiming to translate these findings into effective treatments for MS. Despite promising advances, challenges remain in developing EBV-targeted therapies for MS, including safety concerns and the multifactorial nature of MS pathogenesis. Advance treatment options that focus on EBV, such as adoptive T-cell therapy and gene-based immunotherapy, are shown to be effective in the improvement of MS management that targets the viral-infected cell. The clinical trials for antiviral drugs and vaccination tactics are going on to benefit from these findings and eventually to invent effective therapeutics for MS. While these new therapeutic directions may offer great promise, challenges remain in these approaches as safety concerns and complex factors that underlie MS pathology need to be taken care of. The ethical aspects linked to picking the patients and giving informed consent make the progress of EBV-related treatments are even more difficult. Future research is recommended so that the primary mechanisms through which EBV contributes to MS development will be elucidated; in addition, the main MS subtype sources must be addressed. Longitudinal studies and other advanced research technologies will provide hope because they can solve the complicated problems of MS due to viruses and look for new therapeutic targets. The review brings up EBV/IM disease as a vital aspect of MS susceptibility, encouraging research in the field of

longitudinal studies. Although we have made advances, we are still far from clear on the labyrinthine pairing between EBV and MS and the development of therapeutic strategies to attack EBV infection in MS patients.

Keywords: Chronic infection, Epstein–Barr virus, Flare, Multiple sclerosis, Neurological disease, MS, EBV, FLARE, Neurological Disease, Chronic infection

INTRODUCTION

The association between multiple sclerosis (MS) and Epstein–Barr virus (EBV) infection is essential for understanding the etiology of MS. This relationship is evidenced by various studies, including meta-analyses that demonstrate a significant link between EBV and MS and laboratory reports that detect EBV-infected cells in MS cases, particularly those identified through asymptomatic EBV infections. Research indicates elevated levels of EBV DNA in the spinal fluid and blood of MS patients, suggesting a contributory role of EBV in MS relapses. These findings illustrate patterns of EBV infection and MS prevalence, highlighting differential risk exposures across populations. Early indicators, such as increased neurofilament levels, offer predictive insights into MS progression. Reviewing these components provides a comprehensive understanding of EBV's role in MS, guiding the development of novel therapeutic approaches. The study aims to examine EBV/infectious mononucleosis (IM) as an independent risk factor for MS onset instead of considering only the link between the risk of MS onset with the other factors. This can be done using advanced statistical analysis methods. Perfectly in line with Preferred Reporting Items for Systematic Reviews and Meta-Analysis protocol then, all the choices are standardized, which can be evaluated, in the end, as more clear and transparent.

Objectives

The objective of this research is to elucidate the role of EBV or infectious mononucleosis (IM) in the pathogenesis of MS. It is imperative to establish EBV/IM as an independent risk factor, potentially manifesting at any stage of a patient's life or over an extended period. The objectives include examining the temporal relationship between EBV/IM infection and the onset of MS and excluding studies that do not meet established criteria, such as availability of full text, relevance to the primary objective, or accurate presentation of the temporal association between EBV/IM infection and MS onset.

REVIEW

Methodology

Search strategy

Our search on the PubMed database was performed till November 2023, looking for papers that claim that it may be the risk factor of MS in the endpoint previous EBV/IM. The scan was completed without date or language restrictions and any subject restrictions; it involved all fields of knowledge. The used search terms included the following: "MS," "EBV," and "MS occurred, which is not a single form of the disease since there are different types of diseases manifesting in the form of MS. Thus, EBV is a member of the human herpesvirus (HHV)-4 family also known as HHV-4. IM, which is referred to as IM as well as EBV, has some kind of search phrases, and their titles and abstracts were scrutinized for relevance; the matching papers were screened according to the inclusion criteria, and the outputs of the papers were saved. We channeled the process of selection in accordance with the Preferred Reporting Items for Systematic Reviews and the Meta-Analysis diagram found in Figure 1.

Eligibility criteria

The following eligibility criteria were applied in the review of retrieved articles: studies that are aimed at evaluating EBV/IM as an independent risk factor for the occurrence of MS across a longitudinal time frame, studies that prospectively or retrospectively assess the risk of MS in relation to EBV/IM in a temporal relationship or a reverse temporal relationship, IM/EBV and MS have been documented according to a verified laboratory parameter or McDonald's/Poser criteria, and full-text availability. Articles not meeting the above criteria, studies with a main objective other than evaluating the risk of MS as a function of EBV infection, studies that are cross-sectional or do not retrospectively or prospectively follow the previously mentioned relationship in a temporal frame, articles that report EBV/IM infection through a validated or a self-reported questionnaire, reviews, systematic reviews, and case reports were excluded from the study.

Quality assessment

The included articles were of either two types: Case–controls or cohorts. After selecting the final included articles, the Newcastle-Ottawa Scale (NOS) tool was used to evaluate the quality of the articles. This tool assesses the quality of published articles using an asterisk scoring system with respect to three parameters: selection, comparability, and outcome (cohorts) or exposure (case–control studies). In the articles selected, the highest score was 8, and the lowest was 5. The average NOS score was 6.9.

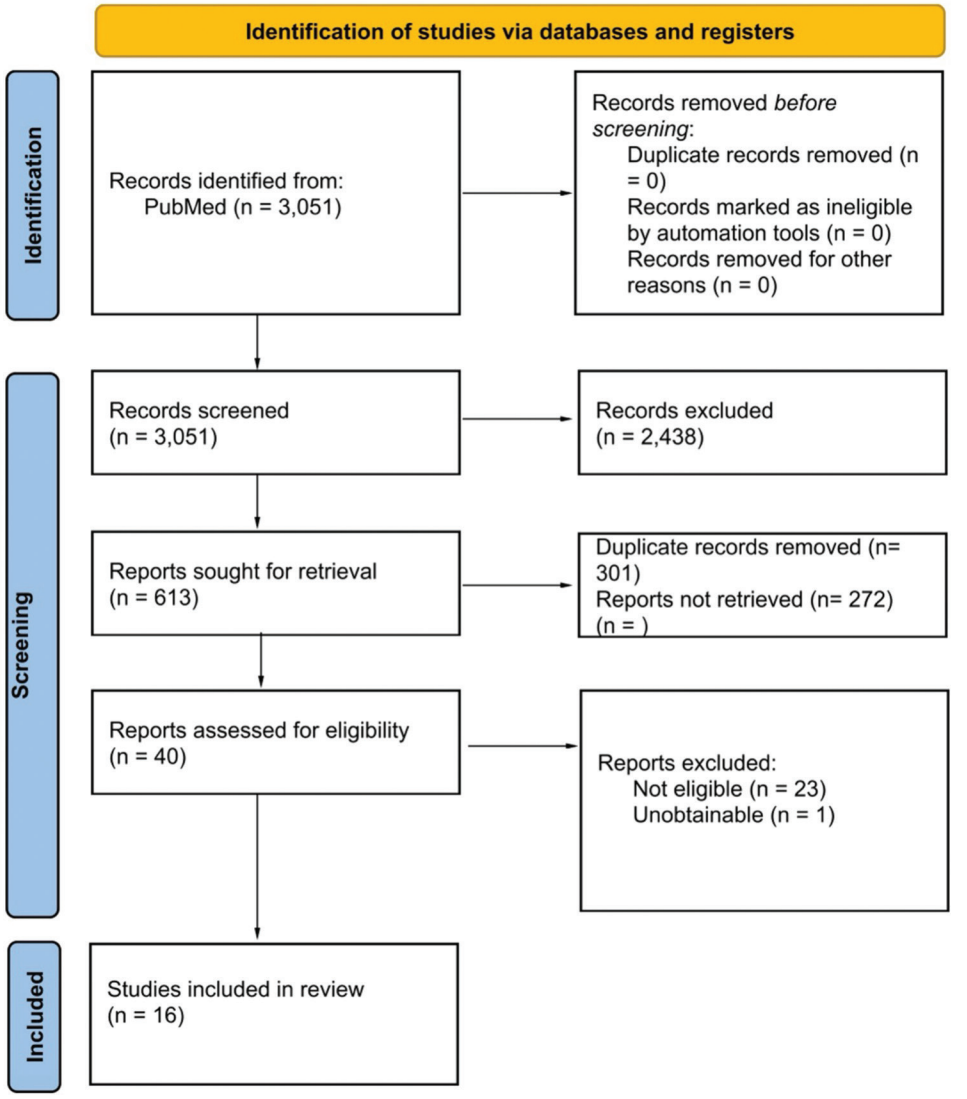

Figure 1: Diagram showing the Preferred Reporting Items for Systematic Reviews and Meta-Analysis protocol.

RESULTS

A total of 16 articles[2,4,10,12,13,19,23-30,34,43] were included in the final analysis that fit the eligibility criteria. The oldest-dated published article was from 1991, and the most recent was from 2023. All articles were either cohort studies (nine) or case–control studies (seven). The methodological study designs used were one of two designs. The first is the retrospective longitudinal cohort design, either starting from the point of diagnosis of IM/EBV infection until reaching the occurrence of MS as the outcome of interest or using subjects with an established diagnosis of MS and searching registry databases for a history of IM/EBV infection. The second design is selecting a cohort of MS-diagnosed individuals who had stored pre-symptomatically collected sera, on which testing for EBV titers and antigens was performed at the time of study conduction. Those two methods were used equally among the included articles. MS occurrence was the outcome of interest in five articles, whereas a history of IM/EBV infection was searched for retrospectively in patients already diagnosed with MS in four papers. The remaining seven articles evaluated the EBV serostatus and/or EBV titers/antigens and their significance regarding the risk of acquiring MS.

The choice of the main subjects was mainly based on the variety of registries that exist worldwide [Table 1]. The number of total subjects overall for studies was 72,868. However, this number could also be an underestimation, as the containment of the notorious articles sometimes could be solid and overlap among the continual version-change of some of the databases or registries. We merged the following databases: the Swedish MS Registry, the Total Population

Table 1: List of articles and corresponding findings.

Study		Source/Type	Sample size	Cases group			Control group		Covered Interval (y)	Time EBV/IM to MS (y)	Variable studied/ endpoint	Result	NOS
Author	Study			Start point	Age (y) (median or mean)	Female (%)	Sample size	Matching					
Jons *et al*., 2023[19]	Nested case-control	Swedish MS registry and local Swedish MS database	669	Established MS	25 (median, sampling) 33 (median, MS onset)	562 (84)	669	1:1	8 (median) (sampling to MS)	_	EBVNA1-Ab, VCAp18, gp350, ANO2, sNfL	Seropositivity: 94% of cases versus 93% of controls Significantly higher EBNA-1, VCA p18, gp350, and sNfL levels in cases versus matched controls Significantly higher EBNA1 10–15 years before MS ANO2 positivity was correlated with a higher risk of MS in EBNA-1 seropositive group	7
Loosen *et al*., 2022[25]	Retrospective cohort	The Disease Analyzer Database (IQVIA)	16, 058	IM history	31.6 (mean, MS onset)	9,407 (58.5%)	16, 058	1:1	2000–2018	_	Incidence of MS	22.6 cases/100,000 versus 11.9 controls/100,000 Significant association between MS and IM	8
Bjornevik *et al*., 2022[4]	Longitudinal cohort	US military	801	Established MS	78.1% <26 years	262 (32.7%)	1,566	1:2	1993–2013	7.5 y (median, estimated)	EBV serostatus	A 32-fold increase in the risk of MS	8
Xu *et al*., 2021[43]	cohort	Swedish Total Population Register	5,867	Established MS	31.5 md (MS dx)	4,017 (68.47%)	_	_	1978–2018	_	History of IM	Increased risk of MS diagnosis in childhood and adolescent age groups	8
Grut *et al*., 2021[13]	Nested case-control	Swedish MS Registry; local biobanks	670	Established MS	25 md (sampling age) 33 md (MS onset)	563 (84%)	670	1:1	_	_	EBNA-1 (trunc and prep), VCAp18,	Increased risk of MS in age groups 20–39 years	7
Ludvigsson JF *et al*., 2016[26]	Cohort	Swedish Total Population Register and National Patient Register	4,022	Established MS	30.11 mean (MS)	2,825 (70.24%)	_	_	_	_	History of IM	IM in adolescence increases the risk of MS diagnosis	8
Downham *et al*., 2017[9]	Case-control	The Clinical Practice Research Datalink (CPRD)	9,247	Established MS	41 (mean, MS) 21.7 (mean, IM)	6,565 (71%)	5,033	Up to six for each case	1990–2010	15 y median	History of IM	History of IM was more frequent in MS patients	6
Décard *et al*., 2012[7]	Retrospective cohort	Blood donation centers	25	First clinical symptom	33.4 (median, MS) 31.5 (median, sample)	21 (84%)	25	1:1	1997–2004	_	EBNA1 titers	EBNA1 was especially elevated in the 3 years before symptoms onset	7
De Jager *et al*., 2008[6]	Case-control	NHS and NHS II	18	Established MS	_	18 (100%)	288	1:2???	1976 NHS I and 1989 NHS II	_	History of IM; EBV titers	Association between MS susceptibility and history of IM and a 4-fold increase in EBV titers was found	6
Nielson *et al*., 2007[30]	Cohort	Statins Serum Institute; Danish National MS Registry; Danish National Discharge Registry	25,234	IM Diagnosis	_	_	_	_	1968–1996	_	MS occurrence	SIR for MS in IM cohort: 2.27 versus SIR 1.2 in non-IM group Risk of MS appears ≥10 years after IM	6
DeLorenze *et al*., 2006[8]	Retrospective case-control	KPNC; OFAS	42	Established MS	45 (median, MS onset) 32 (mean, sample)	36 (86%)	Matched: 79 All controls: 132	?	15 y (median from sample to MS)	_	EBV titers (EBNA complex; EBNA1)	Significantly higher Anti-EBNA complex and EBNA1 in MS versus matched and all controls RR of 4-fold increase in EBV complex and anti-EBNA-1 titers and MS acquisition was 2.1	8
Levin *et al*., 2005[23]	Nested case-control	US military; DoDSR	83	Established MS	24 (mean, sample) 27 (mean, MS onset)	29 (35%)	166	1:2	1988–2000	_	EBV titers	Higher baseline antibody titers (EBNA complex, VCA, EB-D) in cases versus controls A 4-fold increase in EBNA complex at baseline + subsequent serum analysis a/w a 3-fold increase in MS risk	6
Goldacre *et al*., 2004[12]	Cohort	NHS oxford database	2,767	IM diagnosis	18 (mean, IM)	_	640,163	_	1963–1999	14 y (mean)	MS occurrence	non-significant association between IM and the risk of MS significant risk only after 10 years of IM	7
Ascherio *et al*., 2001[2]	Nested case control	NHS and NHS II	18	Established MS	52 (median, MS onset)	18 (100%)	36 matched (Total 288)	1:2	1976 NHS I and 1989 NHS II- FU until 1999	_	EBV antibody titers	Statistically significant higher Antibody titers to EBNA1, EBNA2, and EA-D women with MS than their matched controls	7
Haahr *et al*., 1995[15]	Cohort	Statins Serum Institute; Danish National MS Registry	6,853	IM diagnosis (HA+)	16 y md (females) 19 md (males)	3,111 (45.4%)	12,886 (HA-)	_	1968–1978 (except 1975)	_	MS occurrence	Significantly higher risk of developing MS after IM than the general population (RR 2.81)	7
Lindberg *et al*., 1991[24]	Cohort	Hospital of Infectious Diseases in Göteborg; Sahlgren Hospital; Göteborg MS Register	494	IM diagnosis (HA+)	18 (median, IM) 30 (mean, MS onset)	_	_	_	1950–1967	12 y (mean)	MS occurrence	RR of developing MS is 3.7	5

MS: Multiple sclerosis, EBV: Epstein-Barr virus, IM: Infectious mononucleosis, NHS: Nurses' health study, KPNC: Kaiser Permanente Northern California, OFAS: Orentreich Foundation for the Advancement of Science, RR: Relative risk, EBNA: EBV nuclear antigen, DoDSR: Department of Defense Serum Repository, ICVIA: IQVIA: I (IMS Health), Q (Quintiles), and VIA (by way of) VCA: viral capsid antigen, SIR: Standardized incidence ratios

Register, the National health services (NHS)-I and T, the military database, and the Danish Meningitis Registry. In the articles that fully categorized gender, the National Institutes of Health reported a 27,434 total number of females, which made up 37.6% of the included number of subjects. The age group is not explicit anywhere because different intervals are reported as the average value of age in some articles while, in some others, it was reported as the medium value of age.

There were just four articles that provided the mechanism of action from the exposure of EBV or IM to the time of MS disease onset. Three of those articles[9,12,24] utilized a database search to determine the risk of MS after EBV/IM infection and reported a temporal mechanism derived from these data. The numbers of the major studies reported durations as 15 years,[9] 14 years,[13] and 12 years.[24] The fourth article presented a thorough longitudinal study of MS patients who were exposed to three different time intervals based on their pre-symptom corresponding samples picked and kept amenably afore. Using seroconversion rates along sequential samples to estimate the time any given pair had shared, their predilection was found to be around 7.5 years.

DISCUSSION

Data and results from the literature can be sorted into several subtopics. Below is a detailed discussion of each topic.

Epidemiological insights into the EBV-MS connection

MS is dominated by a number of risk factors, which include infection by EBV and such discovered association is considered a major element. An unfathomably vast meta-analysis has unearthed an association between the virus EBV and MS in as many as 96% of the included studies. It was 2011 when the most asked question for the profession was the reliability of correlation. This was answered when the B-cells of EBV-infected samples were treated in 21 out of 22 MS patients and were absent in other inflammatory neurological diseases.[35] Kids with this type of EBV are among the population that this virus would mostly impact, including those who live a life of utterment asymptomatic. The least severe among the lavish cases of this virus are mostly investigated, but those who experience symptoms tend to have severe outcomes. Despite serum EBV positive in the general population exceeding 90%, the patients with MS display an even higher proportion, which is in addition to the observation that the antibody levels are significantly correlated with unfavorable outcomes.[18]

In the relapsing-remitting MS (RRMS) study, RNA-precipitation analysis was used to analyze EBV nuclear antigen (EBNA)-1 DNA in cerebrospinal fluid (CSF) and blood samples, with higher viral loads in RRMS patients than in controls.[5] Besides that, a wide range of studies finds that EBV specific antibodies' seropositivity in the patients' blood is higher than in the general population, which suggests its potential role in this condition. EBV geographical and age-related distribution present things that are engaging and worth knowing more about. The inauguration of modern hygiene protocols can be associated with a difference in the number of people getting EBV infections for the first time, as the prevalence of this disease is higher in developed countries. MS deficiency amply demonstrated to be related to insufficient sunlight exposure, is among the risk factors, according to rat models.[1] Around 21 years is the age at which MS incidence rises following a northward and southward trend, which may be the result of both ecological factors and genetic predisposition.

The distinct EBV's role elicited in the MS early phase was significantly associated with a biomarker of neurofilaments that presented an increase 6 years before MS attack commencement while at the same time giving room for further exploration of other central nervous system biomarker changes, especially in the neurodegenerative disorder. An EBV infection of MS suffered patients was statistically demonstrated to rise following an EBV infection. This strongly revealed a possibility of a close and immunological mimicry mechanism. It is EBV and driven-interleukin-23 release and the activation mechanism of proinflammatory memory B-cells that become prominent in the disease progression within the central nervous system.[14] It is quite unusual that intrathecal immunoglobulin G production against EBV exhibits a lower ratio in MS patients in comparison with other viruses, possibly due to an enhanced permeability of the blood–brain barrier (BBB) during MS onset.[14]

Recent developments: Exploring EBV and MS relationship with novel methodologies

The most common techniques for studying the role of EBV in MS are observational studies that show higher levels of EBV in MS patients and assays such as enzyme-linked immunosorbent assay flow cytometry and genome-wide association studies. Studies that followed were focused on getting a grip on how the virus interacted with the disease to enable proper patient care. To pinpoint the certain transcription factors involved highly in the disease-related risk loci, Harley *et al.* developed the regulatory element locus intersection (RELI) algorithm.[16] RELI (an EBV-encoded B-cell line from the Muutu family) showed that EBNA2 protein, one of the EBV proteins, tagged 44 of the 109 MS risk loci.[16]

The researchers from Hassani *et al.* focused on producing a high level of quality analysis by relying on a few hundred brain specimens (122 MS and non-MS cases) to identify the existence of EBV at the tissue level.[17] Polymerase chain reaction (PCR), Epstein–Barr encoding region (EBER) *in situ* hybridization (EBER-ISH), and immunohistochemistry (IHC), which are very sensitive

and specific methods for studying the expression of EBNA1 and BZLF1, the key EBV proteins, were used to look at the expression of these EBV proteins. A combined scope of the cellular phenotype of infected cells was carried out by combining EBER-ISH with IHC. EBV had the highest prevalence (90%) among MS cases, although expressed in a low level that may have gone undetected but for meticulous and sensitive search. Unlike EBER-positive cells that were discovered to be scattered rather than clustered, a measure of cells other than B-cells that were proven to be infected with EBV, microglia, and astrocytes were among them.[17]

Sadam *et al*. implemented a hypothesis-free method based on multivalent antigen variants analysis (MVVD) to get antibody epitope profiles of patients with optic neuritis (ON), either those who progress to MS or who did not and discovered new epitope biomarkers for assessing ON progression to MS using blood samples.[33] Using the discriminant predictive model based on the plot of group comparative clusters of peptide epitope identified case-discriminating antigenic repertoires, two major clusters, A and B, were identified and analyzed as mimics of highly antigenic epitopes of gB cytomegalovirus and viral capsid antigen p18 EBV. The study confirmed that these two epitopes were among the most probable serosigns of MS. This was the first report showing a link between the highly antigenic epitopes of the two viruses and MS, indicating direct implication of the two viruses in the pathophysiology of the disease.

Smith *et al*. used the transcriptome analyses of resting B-cells and two types of B at 7 days after EBV infection or CD40L/interleukin-4 stimulation to explore the B-cell activation pathway.[36] Instead of B-cell Receptor (BR), which is already in an activated status, EBV was able to upregulate more genes, suggesting that EBV is a facilitator of B-cell activates. With the results of 3556 differentially expressed genes (DEG) obtained from primary B-cells after EBV infection, Hank discovered the intersection with 873 Multiple Sclerosis (MS)-related genes in B-cells. One hundred and fifty EBV-regulated MS-related genes comprising 18 most associated MS risk pathways were obtained through KEGG pathway enrichment analysis and hypergeometric test. Moreover, 5 hub genes (MALT1, BCL10, IFNGR2, STAT3, and CDK6) were recognized both based on the pathway-pathway and pathway-gene construction. In the final step, EBV miRNAs are predicted using bioinformatics and then experimentally validated for the direct regulation of genes related to MS phenotype.[37]

Using their expertise, Soldan *et al*. and Lieberman *et al*. studied the molecular techniques that could influence the process of B-cells binding with BBB and subsequently penetrating the BBB mediating B-cell neuroinvasion.[39] Green fluorescence protein and luciferase genes from fireflies were incorporated using expression vectors (melcp) into Burkitt lymphoma cells transfected with EBV+. Visualization of this process was typically done by IVIS bioluminescent imaging system. The application of RNA-seq to the question of B-cell neuroinvasion and brain penetration, as well as reverse transcription-PCR to find the molecular determinant of neuroinvasion, were our chosen techniques.[39] Cells with EBNA1 overexpressing and EBNA2 shortage, therefore, may correlate with the ability of B-cells to invade the host neurological system and restrict EBV latency (type I). The retention of EBNA1 may direct the development of epigenetic changes, furthering neuroinvasion. The model represents a novel way of exploring genetic factors inducing B-cell migration to the central nervous system (CNS) as well as therapeutics to halt B-cell neuroinvasibility.[39]

Keane *et al*., using an allele-specific chromatin immunoprecipitation PCR (allele-specific ChIP-PCR), analyzed 6 MS risk single nucleotide polymorphisms (SNPs) and observed that EBNA2 binding, based on the presence of the risk or protective allele, was discovered at five of these loci.[20] A dose-response trial was subsequently conducted on the lymphoblastoid cell lines (LCL), and, I administered the peptide inhibitor of EBNA2, which was my aim to ensure that the optimal concentration (EBNA2-TAT) that that the inhibition of the transcriptional activator of MS risk genes was achieved without being cytotoxic. It has been confirmed that EBNA2 binding suppression could be the cause of the allele's association disruption with the expression, which results in the overall expression.[20]

Although Ristori's crew member, by targeting the available recently mapped TT (TT) mainly came from the intergenic and intronic regions of half-life only minutes, have proven that the genomic region coding for TT is highly enriched with GWAS variants associated with MS.[42] In addition, EBV was mapped with over 487 DNA sites that serve as binding sites for molecular indicators transmitting non-genetic causes of MS. These data unequivocally showed up in the TT-coding regions as MS etiopathogenetic loci.[42]

The human microbiota plays a key role in health and disease. Microbiomes are a dynamic ecosystem that consists of organisms living in symbiotic relationships. These relationships can be either neutral, beneficial, or harmful. Microbiomes can be found in different environments, such as the skin, gut, and lungs. While studying the two-chain-recombined B-cell Receptor (BCR) repertoire from blood and Cerebrospinal fluid of Multiple Sclerosis (CSF of MS) patients, it was discovered that the mAB MS39p2w174 is an immunoglobulin heavy-chain variable region gene (IGHV)-3-7 derivative, cross-reactions both the Epstein–Barr virus (EBV) nuclear antigen 1 (EBNA1) and Glial cell adhesion molecule (CAM). The complex structure of EBNA1 peptide epitope in combination with the MS39p2w174 Fab indicated that the Complementarity-determining region (CDRs) of all domains, except for Light Chain-Complementarity-determining region(LC-CD)R2, were in close contact with

that region of EBNA1 which carries P394–P398 amino acids residues. It was also illustrated by peptide motif analysis of MS39p2w174, which revealed a Pro/Arg-rich motif that was quite similar to the central epitope in EBNA1 (AA395-AA399), confirming their interaction. Tissue-specific protein arrays, HuProt, were used that contain over 80% of the human proteome. Furthermore, GlialCAM (the chronic-active plaque of MS lesions) was revealed to be a critical binding partner with the MS39p2w174 (an MS candidate gene). BLI showed that the ancestor (germline) that unmutated (MS39p2w174) binds EBNA1 with about the same affinity as MS39p2w174.[21] Nevertheless, the affinity of the MS39p2w174 binding the target is ≥10× that of its to leave them the way they have been, which would avoid conflicts. Another important finding was the post-translational modifications of MS39p2w174 protein that increased its binding to GlialCAM protein, most specifically at the Ser376 site ~50-fold. Second, the mice were immunized with both EBNA1AA386-405 and then GlialCAM using the EAE model which is created with mice as being the MS simile.[21]

Exploring potential treatments: EBV-MS connection

The relationship between EBV and MS has turned out to be a search for therapy methods aimed at EBV. The current research considerations thoroughly explore the possibilities of targeting EBV in MS treatment, with adoptive T-cell therapy being one way of approaching the issue. This therapy focuses mainly on eliminating EBV-infected autoreactive B-cells, which contribute to the development of MS. It is based on the idea of reducing the EBV infection rate or virus load, thus reducing MS risk. We can apply another strategy focusing on improving the treatment ideology of MS that stops the main reason for the disease rather than its late factors. Considering the possibility of curing EBV disease through antiviral treatments that may slow disease progression and bring about the desired outcome is the objective of this approach. This is specifically so for the antiviral medicines comprising acyclovir and valacyclovir since the latter has received criticism for the ability to curtail EBV replication. The hoped result is the prevention or slowing down of MS progressing, or at least the reduction of risk factors for disease development.[19]

In addition, the third therapeutic strategy with proven effectiveness involves gene-based immunotherapy, in which cells targeted against EBV-infected malignant cells are administrated. The immunotherapeutic approach has also been under research predominated by B-cell depletion therapy (e.g., rituximab and ocrelizumab) that appear to be able to address B-cells, which perform the core function in EBV infection as well as MS. This multidimensional approach highlights the common purpose of developing those specific therapies against MS through understanding the regular dynamics between EBV and disease.

Clinical trials: EBV-targeted therapies for MS

Clinical studies are still independent and are mainly employing some antiviral agents, vaccination strategies, and cell-based interventions in patients with MS. The outcomes of these analyses are substantial in explaining EBV's modality in terms of being both a cause and a consequence of illnesses like systemic lupus erythematosus.[3,38] Earlier publication-based studies reported that patients with PPMS condition reported clinical amelioration subsequent to autologous EBV-specific T-cell therapy directed at EBNA1, latent membrane protein (LMP) 1, and LMP2A. The initial data indicate the acceptability and safety of ATA188, an incredible T-cell therapeutic. In addition, there is a decrease in EDSS scores, which indicates improvements in the quality of life. Another ammunition is the pursuance of immunotherapeutic strategies, including B-cell depletion treatments, which are indirectly targeted to eliminate chronic active EBV-infected B-cells. Trials sequentially investigating these therapies for patients suffering MS have proven both the reduction in the number, as well as the severity of relapses and the pace of disability progression. These efforts altogether help to create a glimpse of a more detailed picture of how different components are interrelated by EBV and the complex physiology of MS.

Challenges and limitations: Targeting EBV in MS patients

The medications directed against EBV in MS patients' development become a formidable task due to the side effects of the ongoing EBV infection, that is, cancer hazards, autoimmune disorders, and other serious illnesses in humans.[38]

Besides that, the fact that the multifactorial etiology of MS is also the basis of the difficulties and limitations in the conceptualization of EBV-directed therapies for the disease is also a feature. MS is manifested as a multifaceted disease, with both genetics and environmental factors playing major roles. Indeed, the efficacy of EBV as a single causative factor is questionable in the face of numerous players interacting in a highly complex system, as it is rather difficult to identify this sole contributor. Notwithstanding, safety issues are critical because the immune system can be disconnected from its natural state, or antiviral drugs can be administered, resulting in unwanted and even more severe reactions. Furthermore, MS shows profound diversity among its "classes" and "manifestations," which involve distinguishable patterns and progression. Although interventions that have been successful in one patient population cannot be generalized to all categories, they may still offer an optimistic prognosis for some. Moreover, the small number of mono-

specific MS patients in EBV-targeted therapies and the lack of evidence on the long-term safety and efficacy remain a challenge.[3] The origin and development of therapies being targeted for the treatment of virus EBV triggers ethical and regulatory issues such as immune system modulation and vaccine studies. It is essential to overcome these obstacles if we hope to see these advances translated from clinical trials to daily operations in the setting of MS treatment.

Ethical considerations: EBV-RELATED TREATMENTS for MS

Issues of a moral nature in the course of introducing the body environment virus as a treatment of MS include the meaning of selection and informed consent. Likewise, if interventions aimed at EBV-infected individuals, including antiviral treatments, were found to improve disease course or halt their progression, then we would have a clear identification of individuals who are highly vulnerable to MS post-EBV infection. The possibility for primary prevention among patients who are considered high-risk individuals is opened up as a result.[3] Patient education should be part of every phase of treatment, and the importance of informing about the risks and benefits associated with this kind of frame is a must. The primary goal is to maintain the safety of an individual and prevent him from any undesired effects by ensuring a clear understanding of the seriousness and potential side effects of MS treatments, including EBV therapy.[38]

Gaps and future directions

Present research about the role of EBV in MS would reveal that there are certain areas where our understanding is still lacking. The fact that the presence of EBV is 90%. Meanwhile, the specific mechanisms by which EBV contributes to MS pathogenesis are unknown and still emerge in clinical practice.[17,32] It is not completely clear how EBV enters the molecular pathways and affects those different cells, for example, microglia and astroglia, of the CNS. Moreover, the lawful approach to the sequence of EBV infection and MS onset should be investigated by means of longitudinal studies that develop temporal EBV dynamics in individuals who are later identified as having MS. The EBV proteins detected that were associated with MS risk loci raise doubts about their functional significance in the context of MS, requiring profound analysis. Besides, one of the complicating factors is the heterogenic nature of MS and the possible relationship variations among different MS subtypes that have not been specifically demonstrated in current studies do not explicitly address.[37]

Moreover, a number of the gaps in current research are related to the subject of the B-cell neuroinvasion on MS progression.[3,4,17,38] Although the literature masks B-cells' neuroinvasion and its link to EBV, it remains not fully understood how the issue affects the stage and course of MS and a thorough examination is needed.[15,16,37] Therefore, the clinical significance and specific biomarkers discovered in the literature with regard to ON progression need confirmation from bigger cohorts. The key to the implementation of these biomarkers in the diagnosis and the prognosis of MS lies in the fact that these biomarkers are predictive and in addition to that, they are a measure of disease activity. In future studies, instead of just broad clinical trials, researchers should also validate ML findings with well-designed clinical studies, determine how EBV infection in the CNS affects functionalities, clarify molecular mechanisms related to various MS pathological aspects, and address the challenges of B-cell invasion in the context of disease progression.[3] According to Rang *et al*., antibodies recognizing EBV latent proteins (such as EBNA1, LMP1, and LMP2a) have the potential to enhance immunity against EBV, consequently reducing susceptibility to MS.[32] In addition, strategies targeting B-cell pathways and inducing apoptosis in EBV-infected cells may prove effective in minimizing EBV reactivation, thereby holding implications for MS treatment and prevention. Lanz *et al*., in a comprehensive review, present key questions addressing how EBV triggers MS, covering both molecular mechanisms and potential therapeutic approaches.[22] The intriguing role of chloride channels in molecular mimics proximate to EBNA-1 is proposed as a subject for exploration by electrophysiologists and biochemists.

The therapeutic approach for MS treatment in regard to EBV virus might be to activate the lytic phase of EBV in B-cells, then to use an antiviral drug, with trials using tenofovir and other antivirals likely to be used. At present, trials are in progress concerning the approaches of the EBV-targeted T-cell immunotherapy, mRNA vaccines developed against the EBV, and the clinical investigations on the antiviral therapies' effects on EBV replication. Supplied by Massachusetts General Hospital, a Truvada therapeutic interventional clinical trial examines the effect of the drug on EBV levels in people with MS.[11] In addition, the institution also sponsors a study that is particularly oriented to issue the safety and effectiveness of tenofovir alafenamide as an MS treatment, especially among patients with RRMS.[40]

The next study is undertaken by Queen Mary University London in which the impact of famciclovir on EBV activity in RRMS patients is analyzed, and Atara Biotherapeutics conducts a trial to investigate the safety and efficacy of ATA188, which is an allogeneic T-cell immunotherapy in PPMS and SPMS patient groups.[11,31] A study of Nantes University Hospital financed by the latter is carried out to assess the safety and feasibility of adoptive cell therapy using

autologous EBV-specific cytotoxic T lymphocytes in patients with a clinical episode that appears to be suggestive of MS. The objective is to restore the control of EBV in MS patients with partially ineffectual control.[41]

CONCLUSION

This systematic review clarifies the complex connection between patients with previously diagnosed EBV infection or IM and their increased risk of developing MS. Through an extensive literature review and a thorough analysis of eligible articles with rigorous methods, we have emphasized a possible role of EBV/IM infection as one of the contributing factors of MS susceptibility. Our results strongly suggest that the EBV/IM relationship should be taken as the critical factor in the context of MS development, which provides an argument for the longitudinal studies to clarify the disease progression further. Although our systematic review highlights virtually all the connections between EBV and MS, additional studies are necessary to decipher the main trigger factors. Besides that, the identification of drugs targeting EBV may pave the way for the successful management or elimination of MS in such individuals.

Ethical approval

The Institutional Review Board approval is not required.

Declaration of patient consent

Patient's consent was not required as there are no patients in this study.

Financial support and sponsorship

Publication of this article was made possible by the James I. and Carolyn R. Ausman Educational Foundation.

Conflicts of interest

There are no conflicts of interest.

Use of artificial intelligence (AI)-assisted technology for manuscript preparation

The authors confirm that there was no use of artificial intelligence (AI)-assisted technology for assisting in the writing or editing of the manuscript and no images were manipulated using AI.

REFERENCES

1. Alfredsson L, Olsson T. Lifestyle and environmental factors in multiple sclerosis. Cold Spring Harb Perspect Med 2019;1:a028944.
2. Ascherio A, Munger KL, Lennette ET, Spiegelman D, Hernán MA, Olek MJ, *et al*. Epstein-Barr virus antibodies and risk of multiple sclerosis: A prospective study. JAMA 2001;286:3083-8.
3. Bar-Or A, Pender MP, Khanna R, Steinman L, Hartung HP, Maniar T, *et al*. Epstein-Barr virus in multiple sclerosis: Theory and emerging immunotherapies. Trends Mol Med 2020;26:296-310.
4. Bjornevik K, Cortese M, Healy BC, Kuhle J, Mina MJ, Leng Y, *et al*. Longitudinal analysis reveals high prevalence of Epstein-Barr virus associated with multiple sclerosis. Science 2022;375:296-301.
5. Cocuzza CE, Piazza F, Musumeci R, Oggioni D, Andreoni S, Gardinetti M, *et al*. Quantitative detection of Epstein-Barr virus DNA in cerebrospinal fluid and blood samples of patients with relapsing-remitting multiple sclerosis. PLoS One 2014;10:94497-10.
6. De Jager PL, Chibnik LB, Cui J, Reischl J, Lehr S, Simon KC, *et al*. Integration of genetic risk factors into a clinical algorithm for multiple sclerosis susceptibility: A weighted genetic risk score. Lancet Neurol 2009;8:1111-9.
7. Décard BF, von Ahsen N, Grunwald T, Streit F, Stroet A, Niggemeier P, *et al*. Low vitamin D and elevated immunoreactivity against Epstein-Barr virus before first clinical manifestation of multiple sclerosis. J Neurol Neurosurg Psychiatry 2012;83:1170-3.
8. DeLorenze GN, Munger KL, Lennette ET, Orentreich N, Vogelman JH, Ascherio A. Epstein-Barr virus and multiple sclerosis: Evidence of association from a prospective study with long-term follow-up. Arch Neurol 2006;63:839-44.
9. Downham C, Visser E, Vickers M, Counsell C. Season of infectious mononucleosis as a risk factor for multiple sclerosis: A UK primary care case-control study. Mult Scler Relat Disord 2017;17:103-6.
10. Effects of antiviral therapies on Epstein-Barr virus replication; (2023-2024). Available from: https://classic.clinicaltrials.gov/ct2/show/NCT05957913.
11. Famciclovir in multiple sclerosis; (2023-2024). Available from: https://www.hra.nhs.uk/planning-and-improving-research/application-summaries/research-summaries/famciclovir-in-multip.
12. Goldacre MJ, Wotton CJ, Seagroatt V, Yeates D. Multiple sclerosis after infectious mononucleosis: Record linkage study. J Epidemiol Community Health 2004;58:1032-5.
13. Grut V, Biström M, Salzer J, Stridh P, Lindam A, Alonso-Magdalena L, *et al*. Free vitamin D3 index and vitamin D-binding protein in multiple sclerosis: A presymptomatic case-control study. Eur J Neurol 2022;29:2335-42.
14. Guan Y, Jakimovski D, Ramanathan M, Weinstock-Guttman B, Zivadinov R. The role of Epstein-Barr virus in multiple sclerosis: From molecular pathophysiology to *in vivo* imaging. Neural Regen Res 2019;14:373-86.
15. Haahr S, Koch-Henriksen N, Møller-Larsen A, Eriksen LS, Andersen HM. Increased risk of multiple sclerosis after late Epstein-Barr virus infection: A historical prospective study. Mult Scler 1995;1:73-7.
16. Harley JB, Chen X, Pujato M, Miller D, Maddox A, Forney C, *et al*. Transcription factors operate across disease loci, with EBNA2 implicated in autoimmunity. Nat Genet 2018;50:699-707.
17. Hassani A, Corboy JR, Al-Salam S, Khan G. Epstein-

Barr virus is present in the brain of most cases of multiple sclerosis and may engage more than just B cells. PLoS One 2018;13:e0192109.
18. Hedström AK. Risk factors for multiple sclerosis in the context of Epstein-Barr virus infection. Front Immunol 2023;24:1212676.
19. Jons D, Grut V, Bergström T, Zetterberg H, Biström M, Gunnarsson M, *et al*. Seroreactivity against lytic, latent and possible cross-reactive EBV antigens appears on average 10 years before MS induced preclinical neuroaxonal damage. J Neurol Neurosurg Psychiatry 2023;95:325-32.
20. Keane JT, Afrasiabi A, Schibeci SD, Swaminathan S, Parnell GP, Booth DR. The interaction of Epstein-Barr virus encoded transcription factor EBNA2 with multiple sclerosis risk loci is dependent on the risk genotype. EBioMedicine 2021;71:103572.
21. Lanz TV, Brewer RC, Ho PP, Moon JS, Jude KM, Fernandez D, *et al*. Clonally expanded B cells in multiple sclerosis bind EBV EBNA1 and GlialCAM. Nature 2022;603:321-7.
22. Lanz TV, Robinson WH, Ho PP, Steinman L. Roadmap for understanding mechanisms on how Epstein-Barr virus triggers multiple sclerosis and for translating these discoveries in clinical trials. Clin Transl Immunology 2023;12:e1438.
23. Levin LI, Munger KL, Rubertone MV, Peck CA, Lennette ET, Spiegelman D, *et al*. Temporal relationship between elevation of Epstein-Barr virus antibody titers and initial onset of neurological symptoms in multiple sclerosis. JAMA 2005;293:2496-500.
24. Lindberg C, Andersen O, Vahlne A, Dalton M, Runmarker B. Epidemiological investigation of the association between infectious mononucleosis and multiple sclerosis. Neuroepidemiology 1991;10:62-5.
25. Loosen SH, Kostev K, Jördens MS, Luedde T, Roderburg C. Overlap between irritable bowel syndrome and common gastrointestinal diagnoses: A retrospective cohort study of 29,553 outpatients in Germany. BMC Gastroenterol 2022;5:48.
26. Ludvigsson JF, Almqvist C, Bonamy AK, Ljung R, Michaëlsson K, Neovius M, *et al*. Registers of the Swedish total population and their use in medical research. Eur J Epidemiol 2016;31:125-36.
27. Lünemann JD, Tintoré M, Messmer B, Strowig T, Rovira A, Perkal H, *et al*. Elevated Epstein-Barr virus-encoded nuclear antigen-1 immune responses predict conversion to multiple sclerosis. Ann Neurol 2010;67:159-69.
28. Munger KL, Chitnis T, Ascherio A. Body size and risk of MS in two cohorts of US women. Neurology 2009;10:1543-50.
29. Munger KL, Levin LI, O'Reilly EJ, Falk KI, Ascherio A. Anti-Epstein-Barr virus antibodies as serological markers of multiple sclerosis: A prospective study among United States military personnel. Mult Scler 2011;17:1185-93.
30. Nielsen TR, Rostgaard K, Nielsen NM, Koch-Henriksen N, Haahr S, Sørensen PS, *et al*. Multiple sclerosis after infectious mononucleosis. Arch Neurol 2007;64:72-5.
31. Phase 1/2 study to evaluate the safety and efficacy of ATA188 in subjects with progressive multiple sclerosis; (2023-2024). Available from: https://classic.clinicaltrials.gov/ct2/show/NCT03283826.
32. Rang X, Liu Y, Wang J, Wang Y, Xu C, Fu J. Identification of multiple sclerosis-related genes regulated by EBV-encoded micrornas in B cells. Mult Scler Relat Disord 2022;59:103563.
33. Sadam H, Pihlak A, Jaago M, Pupina N, Rähni A, Toots M, *et al*. Identification of two highly antigenic epitope markers predicting multiple sclerosis in optic neuritis patients. EBioMedicine 2021;64:103211-10.
34. Schmidt M, Schmidt SA, Sandegaard JL, Ehrenstein V, Pedersen L, Sørensen HT. The Danish national patient registry: A review of content, data quality, and research potential. Clin Epidemiol 2015;7:449-90.
35. Serafini B, Rosicarelli B, Franciotta D, Magliozzi R, Reynolds R, Cinque P, *et al*. Dysregulated Epstein-Barr virus infection in the multiple sclerosis brain. J Exp Med 2007;26:2899-12.
36. Smith C, Khanna R. Adoptive T-cell therapy targeting Epstein-Barr virus as a treatment for multiple sclerosis. Clin Transl Immunol 2023;12:e1444.
37. Smith N, Tierney R, Wei W, Vockerodt M, Murray PG, Woodman CB, *et al*. Induction of interferon-stimulated genes on the IL-4 response axis by Epstein-Barr virus infected human b cells; relevance to cellular transformation. PLoS One 2013;27:64868.
38. Soldan SS, Lieberman PM. Epstein-Barr virus and multiple sclerosis. Nat Rev Microbiol 2023;21:51-64.
39. Soldan SS, Su C, Lamontagne RJ, Grams N, Lu F, Zhang Y, *et al*. Epigenetic plasticity enables CNS-trafficking of EBV-infected B lymphocytes. PLoS Pathog 2021;9:1009618.
40. Tenofovir alafenamide for treatment of symptoms and neuroprotection in relapsing remitting multiple sclerosis; (2023-2024). Available from: https://classic.clinicaltrials.gov/ct2/show/NCT04880577.
41. Trial to assess the safety and feasibility of adoptive cell therapy with autologous EBV-specific cytotoxic T lymphocytes (CTL) in patients with a first clinical episode highly suggestive of multiple sclerosis; (2023-2024). Available from: https://classic.clinicaltrials.gov/ct2/show/NCT02912897.
42. Umeton R, Bellucci G, Bigi R, Romano S, Buscarinu MC, Reniè R, *et al*. Multiple sclerosis genetic and non-genetic factors interact through the transient transcriptome. Sci Rep 2022;9:7536-10.
43. Xu Y, Hiyoshi A, Smith KA, Piehl F, Olsson T, Fall K, *et al*. Association of infectious mononucleosis in childhood and adolescence with risk for a subsequent multiple sclerosis diagnosis among siblings. JAMA Netw Open 2021;1:e2124932.

How to cite this article: Msheik AN, Al Mokdad Z, Hamed F, Assi F, Jibbawi A, Saad J, *et al*. Epstein–Barr virus flare: A multiple sclerosis attack. Surg Neurol Int. 2024;15:355. doi: 10.25259/SNI_457_2024