

## Presence of Herpesviridae Genome in CSF of GBS Patients

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### Abstract:

**Background:** Guillain Barre Syndrome (GBS) is considered as an immune mediated inflammatory disease of peripheral nerves and nerve roots. Herpes viruses like CMV, EBV, HSV and VZV infections are associated with GBS. The aim of the study was to identify the presence and frequency of different Herpes virus genome by PCR assay in CSF of GBS patients **Methods:** An observational, cross sectional study was carried out in the Department of Neurology, BSMMU, and Dhaka. A total 50 (fifty) admitted GBS patients were included after fulfilling the inclusion and exclusion criteria. About 2 ml of CSF was taken for detection of viral nucleic acid of CMV, EBV, HSV1, HSV2, VZV and HHV6 by Multiplex PCR method. **Results:** Herpesviridae genome in CSF of study population was present in 9 (18%) patients. Maximum 3 (33.3%) cases were HSV1, EBV and HSV2 found in 2 (22.2%) patients, CMV and VZV in 1 (11.1%) patient respectively. AMAN (66.6%) was the most frequent electrophysiological pattern; followed by AIDP (33.3%), mean CSF protein was (159±81) mg/dl and mean cell count was (2±3)/cmm in these patients. Herpes virus genome positive group patients were more disabled and received definitive treatment more than the others. **Conclusion:** Herpesviridae genome was present in CSF of GBS especially in early collected CSF sample. Antiviral drugs might have a role in treating GBS patients having Herpes virus genome in CSF.

**Key words:** Guillain Barre Syndrome (GBS), Herpes Virus genome, PCR, CSF.

### Materials and methods:

This cross sectional observational study. It was conducted in department of Neurology (In patient department), BSMMU, Dhaka. Male and female of 18 years or above who were diagnosed clinically by accepted clinical criteria as Guillain-Barre syndrome in inpatient department, department of Neurology, BSMMU, Dhaka was taken as study population. About 2-3 ml of CSF was taken for detection of PCR for viral nucleic acid of six virus of Herpesviridae group (CMV, EBV, HSV1, HSV2, VZV, and HHV6) by Multiplex PCR method (Seeplex meningitis ACE kit) in Microbiology department of BSMMU. Pattern of clinical

presentation was documented and disability was graded by Hughes functional grading scale. Routine CSF study was done in the Biochemistry and Clinical Pathology department for CSF protein and cytology respectively. Nerve conduction study was performed in the "Neurophysiology laboratory" of BSMMU and pattern of findings of these investigations were recorded. Presence and frequency of different Herpes virus genome was expressed in percentage and their association with variation in clinical presentation, electro-physiological findings and routine CSF findings was statistically analyzed by Chi-square test and t-test. The P value <0.05 was considered statistically significant.

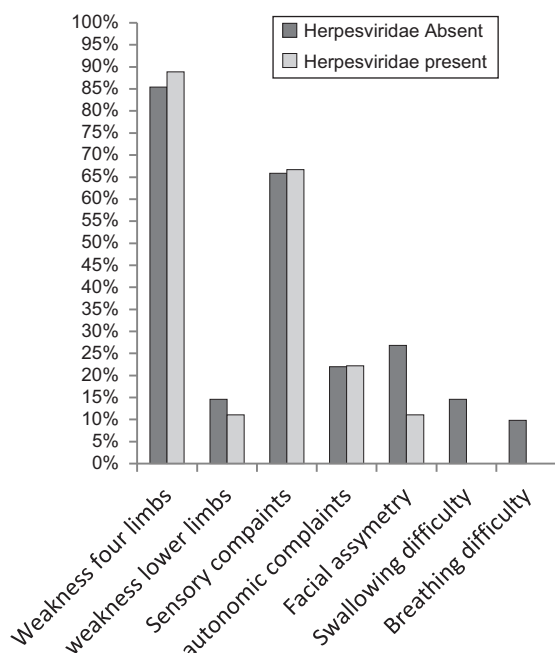
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**Results and observations:**

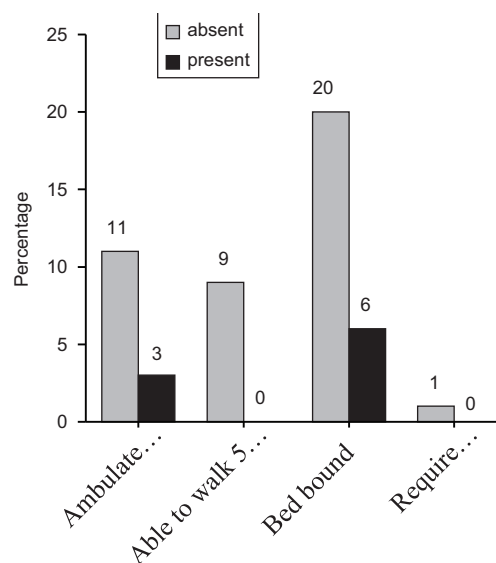
**Table-I**

*Distribution of study population by age in relation to Herpesviridae genome in CSF (n=50)*

Age in years	CSF viral genome by PCR			P-Value
	Absent n (%)	Present n (%)	Total n (%)	
< 20	3 (7.3)	3(33.3)	6 (12)	0.029 <sup>s</sup>
21- 30	13(31.7)	4(44.4)	17 (34)	0.465 <sup>ns</sup>
31- 40	9 (22)	0 (0)	9 (18)	0.120 <sup>ns</sup>
41- 50	6 (14.6)	0 (0)	6 (12)	0.221 <sup>ns</sup>
>50	10(24.4)	2(22.2)	12 (24)	0.890 <sup>ns</sup>
Total	41 (100)	9 (100)	50 (100)	0.098 <sup>s</sup>



**Fig-1 (Bar diagram):** Presenting complaints of study population.



**Fig-2 (Bar diagram):** Disability status of study population.

**Table-II**

*Presence of Herpesviridae genome in CSF of study population (n=50)*

Name of the Herpes virus	Number	Percentage
CMV	01	11.1
EBV	02	22.2
HSV 1	03	33.3
HSV 2	02	22.2
VZV	01	11.1
HHV 6	00	0.00
Total	09	100

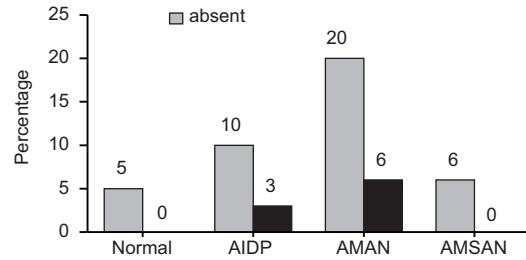
**Table-III**

*CSF protein level of study population (n=50)*

	Absent	Present	Total	P-Value
	Group	Group		
Mean	CSF protein (mg/dl)	CSF protein (mg/dl)	175	0.774
Standard Deviation	178	159	179	

**Table-IV:**  
CSF cytology of study population (n=50)

	Absent	Present	Total	P-Value
	Group (CSF cell/cmm)	Group (CSF cell/cmm)		
Mean	12	2	10	0.523
Standard Deviation		47	3	42



**Fig-3: (Bar diagram):** Electrophysiological pattern of study population

**Table-V**

CSF collection time to detect Herpesviridae genome among cases at time interval in weeks (n=50)

CSF collection time	CSF viral genome by PCR		Total n (%)	P-Value
	Absent n (%)	Present n (%)		
< 7 days	0 (0)	1(11.1)	1 (2)	0.031 <sup>s</sup>
8-14 days	22(53.7)	7(77.8)	29 (58)	0.184 <sup>ns</sup>
15-28 days	19(46.3)	1(11.1)	20 (40)	0.050 <sup>ns</sup>
Total	41 (100)	9 (100)	50(100)	0.023 <sup>s</sup>

**Table-VI**

Distribution of study population by received treatment (n=50)

Treatment modalities	CSF viral PCR positivity		Total n(%)	P-Value
	Absent n(%)	Present n(%)		
Conservative	29 (70.7)	4(44.4)	33 (66)	0.131 <sup>ns</sup>
Plasma exchange	9 (22)	5(55.6)	14 (28)	0.042 <sup>s</sup>
IVIg	3 (7.3)	0 (0)	3 (6)	0.402 <sup>ns</sup>
Total	41 (100)	9 (100)	50 (100)	0.110 <sup>ns</sup>

**Discussion:**

Guillain-Barre Syndrome (GBS) is considered as an immune mediated inflammatory disease of peripheral nerves and nerve roots. Two thirds of patient has a preceding history of various viral and bacterial infections before the symptoms onset<sup>1</sup>. GBS is thought to be result of immune reaction against these infectious agents having epitope similar to peripheral nerves myelin sheath or axonal membrane component. This type of molecular mimicry was found mainly those of bacterial infectious agent like Campylobacter jejuni, Hemophilus influenzae and Mycoplasma. Herpes

viruses like CMV, EBV, HSV and VZV infections are associated with GBS<sup>3</sup>. The above mentioned mechanism of nerve injury cannot be proved in virus associated GBS patients who may have different pathogenesis described in some study.

Different study shows that antibody against herpes virus and PCR for herpes virus genome was found in serum and CSF of GBS patients. As their study found PCR for Herpes virus genome was present in CSF of GBS patients after disease onset so they claimed that there may be ongoing infection or presence of Herpes virus genome in CSF has a different role in GBS pathogenesis. This cross-

sectional observational study was carried out primarily with an aim to identify the presence of different Herpes virus genome by multiplex PCR method in CSF of GBS patients, to identify their frequency, any association with variation in clinical presentation, electrophysiological pattern and routine CSF findings of them. A total 50 (fifty) GBS patients admitted in the Neurology department of BSMMU, Dhaka, were included after fulfilling the inclusion and exclusion criteria. GBS patients of any age and sex were included in our study.

We have made two groups on the basis of presence or absence Herpes virus genome in CSF of GBS patients within 4 weeks of disease onset. We have also compared between two groups to find out any association of CSF Herpes virus genome positivity with any variation in pattern of clinical presentation, electrophysiological findings, cytological and biochemical parameter of CSF. The timing of presence of Herpes virus genome by multiplex PCR method in CSF of GBS patients and its significance was also observed.

Analysis of age distribution (Table-I) of two groups showed that the mean age ( $\pm$ SD) was 39 ( $\pm$ 15) years in CSF Herpes virus genome absent group and 30 ( $\pm$ 16) years in CSF Herpes virus genome present group. Male patients were 42 (84%) and female patients were 8 (16%). Higher frequency was found in male than female. Most of the study patients were young adults of 21-30 yrs, 4 (44.4%) patients in CSF Herpes virus genome positive group and 13 (31.7%) patients in CSF Herpes virus genome negative group. So our study showed that Herpes virus genome in CSF is found in younger GBS patients. In this study presenting complaints (Figure-1, Bar diagram) was weakness of all four limbs in 43 (86%) patients, lower limb weakness in 7 (14%) patients. Sensory complaints were present in 33 (66%) of patients. Complaints of facial weakness or deviation were present in 12 (24%) patients, 4 (8%) patient had breathing difficulty and 6 (12%) patients had dysphagia. There was no significant difference found in terms of clinical presentation in between two groups. In our study maximum number of patients 20 (48.8%) were in the grade 4 disability scale in Herpes virus genome absent group and 6 (66.7%) patients were in

Herpes virus genome present group. Patients who had Herpes virus genome in CSF were more disabled during admission than who had absent Herpes virus genome in CSF (Figure-II). Our study found Herpes virus genome in CSF was present in 9 (18%) patients and was absent in 41 (82%) patients. Maximum 3 (33.3%) cases were HSV1 followed by 2 (22.2%) cases were EBV and HSV2, 1 (11.1%) was CMV and VZV respectively (Table-II). This result has similarity with that of Armin S. et al., (2014)<sup>13</sup> who found Herpes virus genome in CSF of GBS patients in 20% cases (3 out of 15 patients).

In our study CSF was drawn between 8-14 days of disease onset in maximum 22 (53.7%) patients followed by 19 (46.3%) patients within 15-28 days in CSF Herpes virus genome negative group. In contrast CSF was drawn between 8-14 days of disease onset 7 (77.8%) patients and 1 (11.1%) patient within 7 days in CSF Herpes virus genome positive group. So our study found that Herpesviridae genome was found more in GBS patients whose CSF was taken early after disease onset (Table-V). Our one specific objective was to find out any correlation between two groups in regards of CSF findings. Mean protein was (178 $\pm$ 195) mg/dl in Herpes virus genome absent group and (159 $\pm$ 81) mg/dl in Herpes virus genome present group respectively. Mean cell count was (12 $\pm$ 47)/cmm in Herpes virus genome absent group and (2 $\pm$ 3)/cmm in Herpes virus genome present group respectively (Table-III). No significant difference was found between two groups but CSF cell count and protein was higher in Herpesviridae genome negative group (Table-IV).

We also had a specific objective to see association of presence of Herpes virus genome in CSF with electrophysiological pattern of our patients. The present study found predominant electrophysiological subtype in both group was AMAN found in 6 (66.7%) patients in Herpes virus genome positive group and 20 (48.8%) patients in CSF Herpes virus genome negative group. Next common electrophysiological pattern was AIDP observed in 3 (33.3%) patients in CSF Herpes virus genome positive group and 10 (24.4%) patients in CSF Herpes virus genome negative group

respectively. NCS was normal in 5 (12%) patients (Figure-III). No significant association was found between presence of Herpes virus genome in CSF and electrophysiological pattern. But axonal variants (AMAN and AMSAN) constitute the predominant subtype 64% (52% AMAN and 12% AMSAN) in our population. A study done by Islam et al. (2010)<sup>7</sup> in Bangladesh, also found higher frequency of axonal variants in 67% (AMAN-56% and AMSAN-11%) followed by AIDP in 22% of cases and 11% were unclassified. Thus the present study has similarities with the previous study although AIDP was the predominant subtype in many developed countries including Europe and North America. Another observation of our study is presence of CSF Herpes virus genome render the patient to receive more specific treatment than who has absent Herpes virus genome in CSF. Definitive treatment was given more in CSF Herpes virus genome positive group (55.6%) patients than CSF Herpes virus genome negative group (49.3%) patients. Fourteen (14, 28%) patients were treated with plasma exchange and 3 (6%) patient received intravenous immunoglobulin during their course of illness. Majority 33 (66%) of patients had received conservative treatment (Table-VI).

#### **Conclusion:**

The present study showed that Herpesviridae genome may be found in CSF of GBS patients after the disease onset especially in younger patients. The presence of Herpes virus genome by PCR assay in CSF of GBS patients early in the disease course carries significance. It may be related with ongoing or current infection or a different pathogenesis may coexist in these GBS patients but actual mechanism of Herpesviridae genome presence in CSF yet to be elucidated. Present study also showed that Herpes virus genome positivity was associated with greater disability of the GBS patients and necessitated specific costly treatment more than the other patients. But present study failed to correlate the presence of Herpesviridae genome in CSF of GBS patients with routine CSF findings and with the electrophysiological pattern. As our study showed that early in the disease course of GBS patients Herpesviridae genome is present in CSF, so whether antiviral drug can be

effective or not in these patients is a matter of debate and demand for further study

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