Role of host factors in the pathogenesis of Guillain-Barré syndrome

ABSTRACT

of

THESIS

SUBMITTED TO

BABASAHEB BHIMRAO AMBEDKAR UNIVERSITY

LUCKNOW

BABASAHEB
BHIMRAO
AMBEDKAR
UNIVERSITY

LUCKNOW

प्रज्ञा शील करुणा

ESTABLISHED 1996

FOR THE DEGREE OF

Doctor of Philosophy

IN

BIOTECHNOLOGY

Submitted by

Nagendra Kumar Kharwar

Supervisor

Dr. D. R. Modi

Associate Professor,
Department of Biotechnology
BBAU, Lucknow (U.P.)

Co-Supervisor

Dr. K. N. Prasad

Professor,
Department of Microbiology
SGPGIMS, Lucknow (U.P.)

DEPARTMENT OF BIOTECHNOLOGY
BABASAHEB BHIMRAO AMBEDKAR UNIVERSITY
(A CENTRAL University; NAAC-'A' GRADE)
VIDYA VIHAR, RAEBARELI ROAD, LUCKNOW-226 025

Enrolment Number: 477/11

2016

Abstract

Guillain Barré syndrome (GBS) is an immune-mediated inflammatory disease mainly affecting the myelin and axons of peripheral nerves with heterogeneous pathological features. GBS is often triggered by an aberrant immune response towards an infectious pathogen and is preceded by an infectious illness usually 1-3 weeks before the onset of neurological symptoms. Epidemiological studies linked it with *Campylobacter jejuni*, Cytomegalovirus, Epstein Barr virus and *Mycoplasma pneumoniae*. The mechanisms involved in immunopathogenesis of GBS are still unclear. The hypothesis put forward for the immunopathogenesis of GBS points to molecular mimicry between lipopolysaccharide (LPS) and ganglioside like epitopes in host nerve cells, which leads to cross reactivity of immune response after the infection. Besides microbial factors, host susceptibility may also play an important role in the etiology of GBS, because not all infected individuals develop this disorder. It is estimated that only 1:1000 people develop GBS after *C. jejuni* enteritis, thus highlighting the role of host genetic factors. However, studies demonstrating for identifying the potential host factors that may impart susceptibility to GBS are least understood. In present study, we investigated the polymorphism (TLR-2, IL-8, NLRs, IL-17 and ICAM-1) and expression (TLR-2, IL-8, IL-17 and ICAM-1) of host factor molecules to define and understand the role in GBS.

Keeping the above observations in mind, the present study was planned. The major features of the present study are summarized as follows:

1. One hundred five patients with GBS (male/female, 84/21; mean age ± SD, 30.20±10.98 years) were enrolled for the study.

2. Age and sex matched 100 individuals (male/female, 78/22; mean age ± SD, 28.12 ± 16.97 years) without any history of apparent infectious illnesses within the last four weeks, were included as healthy control group.

3. History of preceding infection was found in 51 (48.6%) patients. Either alone or in combination, 21(20%) patients had respiratory infection or 10 (9.5%) patients had diarrhoea prior to the onset of GBS.

4. 25 (23.8%) of the 105 GBS cases had infection with *C.jejuni* and 10 (9.52%) of GBS patients had *M.pneumoniae* infection and 2(1.9%) in HC.

5. EBV-VCA infection was detected in 13 (12.4%) of GBS patients however, 15 (14.28%) of GBS patients had CMV infection.

6. Genotypic distribution of TLR2 Arg753Gln and Arg677Trp polymorphism demonstrated increased risk of GBS patient for heterozygous genotypes Arg/Gln (34.3% vs 6%) and Arg/Trp (46.67% vs 1%), when compared with healthy control.

7. Frequency of wild homozygous genotypes was observed at 65.7% vs 94% for TLR2Arg753Gln and 53.33% vs 99% for Arg677Trp polymorphisms in a patient and control wise manner.

8. Patients with GBS showed significantly higher frequency in comparison to controls for allele 753Gln (17.14% vs 6%) and allele 677Trp (23.34% vs 0.5%).

9. The genotypic frequencies of both homozygous (Lys/Lys) and heterozygous (Glu/Lys) variants of NOD1 were higher in GBS patients than healthy controls.

10. Homozygous variant showed increased risk to GBS (40% vs 29%) and heterozygous variant also showed a trend towards risk to GBS than healthy controls (48.57% vs 47%).
11. The frequency of variant allele (NOD1 266Lys) was also higher among GBS patients (64.29%) than healthy controls (52.5%) and showed 1.63 fold risk association with GBS.
12. NOD1 variant allele (266Lys) showed increased susceptibility to AMAN and AIDP subtypes.
13. Increased risks for the development of GBS were revealed with heterozygote genotypes of NOD2, Arg702Trp and Gly908Arg.
14. NOD2 alleles i.e., 702Trp & 908Arg also showed significant risk association with GBS.
15. Significant protective association for AMAN, AMSAN and AIDP subtypes were found with NOD2 Arg702Trp heterozygote genotype. On the contrary, predisposing association was observed with NOD2 702Trp allele for AMAN, AMSAN and AIDP subtypes.
16. Heterozygote (Gly/Arg) genotype of NOD2 Gly908Arg revealed protective association and variant allele (Arg) of Gly908Arg polymorphism showed risk associations with AIDP subtype.
17. The occurrence of Arg702-Gly908 haplotype comprising of wild genotype (Arg/Gly) among control and patients was 79% and 68.5% respectively.

18. Significant risk with Trp702-Arg908 haplotype and protective association with Arg702-Arg908 haplotype for GBS were observed.

19. The LD analysis showed five significant associations in this study and it is evident from the analysis that significant risk was associated with GBS in the presence of mutant alleles.

20. Pair-wise LD showed risk for GBS in the presence of Glu (Glu266Lys)-Lys(Glu266Lys), Glu (Glu266Lys)-Trp (Arg702Trp), Glu (Glu266Lys)-Arg (Gly908Arg), Trp (Arg702Trp)-Arg (Gly908Arg) and Trp (Arg702Trp)-Arg (Gly908Arg) alleles among the studied individuals.

21. Only homozygous variant (Gly/Gly) of IL-17F (Glu126Gly) showed increased susceptibility to GBS (53.34% vs. 23%, $p \leq 0.0001$). Further, G allele also showed increased risk to GBS (71.43% vs. 53.5%, $p=0.0002$).

22. IL-17F His161Arg gene polymorphism did not show any association with GBS susceptibility.

23. The logistic regression analysis revealed no significant association of IL-8-251A/T polymorphism (polymorphic and heterozygous) with GBS ($p=0.8748$ and $p=0.0698$).

24. The logistic regression analysis revealed that heterozygous genotype (G/A) of ICAM-1 Gly241Argpolymorphism had association only with GBS ($p<0.0001$) while not any homozygous polymorphic (AA) genotype was detected either in patients or in controls and frequency of homozygous wild type was (30.47% vs 64%) between patients and controls.

25. In expression studies, mRNA expression of TLR 2 was increased significantly in GBS patients than controls (1.727±0.412 vs 0.319±0.113).
26. Enhanced expression of TLR-2 was observed in GBS patients (fold change-1.112) compared to healthy controls (fold change-1) by immunoblotting.
27. mRNA expression of IL-8 was increased significantly in GBS patients than controls (3.47±0.14 vs 1.27±0.07).
28. Level of IL-8 was elevated in sera of GBS Patients than healthy controls (112.55±13.96 vs 34.79±4.373; $p \leq 0.001$).
29. The mRNA level of IL-17 was found increased trend ($p=0.066$, 3.66±0.792 vs 2.05±0.299) in patient than control but statically not significant.
30. The level of IL-17 in sera was elevated in GBS patients when compared with healthy control (42.00±5.33Vs 6.0±0.77; $p \leq 0.001$).
31. In ICAM-1, mRNA expression ($p=0.012$, 2.04±0.261 vs 1.18±0.202) increased in patient compared to control.
32. The elevated level was observed for ICAM-1(5.33±0.661 vs 2.25±0.288) in serum, when compared with HC.