P 406

Baseline Glycolipid IgG Autoantibodies in GBS Are Prevalent and Show Cross-Regional Pattern Concordance

Susan Halstead[1], Preeti Paliwal[2], Henk André Kroon[2], David Lorenz[3], Zhahirul Islam[4], Kazim Sheikh[5], Kenneth C. Gorson[6], Hugh Willison[1]

[1]University of Glasgow, Glasgow, UK; [2]Annexon Biosciences, Brisbane, CA, USA; [3]Excelra Inc, Boston, MA, USA; [4]Gut-Brain Axis Laboratory, icddr,b, Dhaka, Bangladesh; [5]McGovern Medical School, UTHealth, Houston, TX, USA; [6]Tufts University School of Medicine, Boston, MA, USA

Introduction: Guillain-Barré syndrome (GBS) is an autoantibody-mediated disease[1-3]

- GBS is an acute, immune-mediated polyradiculoneuropathy resulting in mild weakness to severe paralysis[1]
- GBS pathogenesis is associated with autoantibodies directed against peripheral nerve glycolipids, which contribute to complement activation, neuroinflammation, and axonal injury[1,2]
- Recent studies highlight the mechanistic and clinical significance of these complex profiles, emphasizing the need to define antibody-driven disease biology to guide therapies addressing GBS presentations and outcomes[2,3]

Study aim: To evaluate the spectrum and cross-regional concordance of anti-ganglioside IgG signatures in GBS-02 and US cohorts

Methods

- GBS-02 (NCT04701164) was a Phase 3, multi-center, double-blind, placebo-controlled study in Bangladesh and the Philippines of tanruprubart examining treatment with patients recently diagnosed with moderate to severe GBS[4]
- Tanruprubart is a monoclonal antibody that rapidly and completely blocks C1q, inhibiting the classical complement pathway[5,6]
- Baseline serum samples from GBS-02 patients (n=239), an independent US cohort of patients with GBS (n=142), and heathy controls (normal/healthy serum controls [age: 35–37 years]) from Bangladesh (n=50) and the US (n=71) were included
- Baseline anti-glycolipid immunoglobulin G (IgG) autoantibody reactivity patterns were screened by glycolipid antigen array (glycoarray) and analyzed by unsupervised hierarchical clustering using the Ward.D2 method with Manhattan distance.[7] Consensus *k*-means clustering was used to partition participants into subsets (**Figure 1**)

GBS-02 study cohort (n=239)

Key inclusion/exclusion criteria:

- Aged ≥16 years
- Had a GBS Disability Score 3, 4, or 5
- ≤10 days from onset of weakness
- Included classical motor or motor/sensory (mixed) GBS; other variants excluded
- Did not have access to intravenous immunoglobulin (IVIg) or plasma exchange

Stratified by baseline prognostic factors: muscle strength and time from onset of weakness until hospital admission

Conducted at sites in Bangladesh and the Philippines

US GBS cohort (n=142)

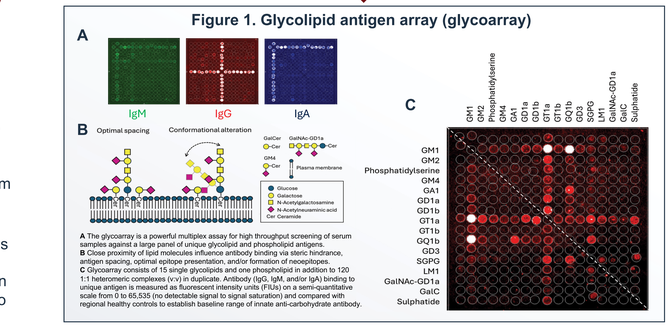
- GBS (96.5%)
- Non-GBS (3.5%)
- Includes other GBS clinical variants (MFS 8.1%), all GBS Disability Score <3 (20.3%) or ≥3 (79.7%)

Healthy controls

- Baseline anti-glycolipid antibody assessed per region
- Healthy US controls (n=71)[a]
- Healthy Bangladesh controls (n=50)[a]

[a]To assess baseline glycolipid expression.

Figure 1. Glycolipid antigen array (glycoarray)



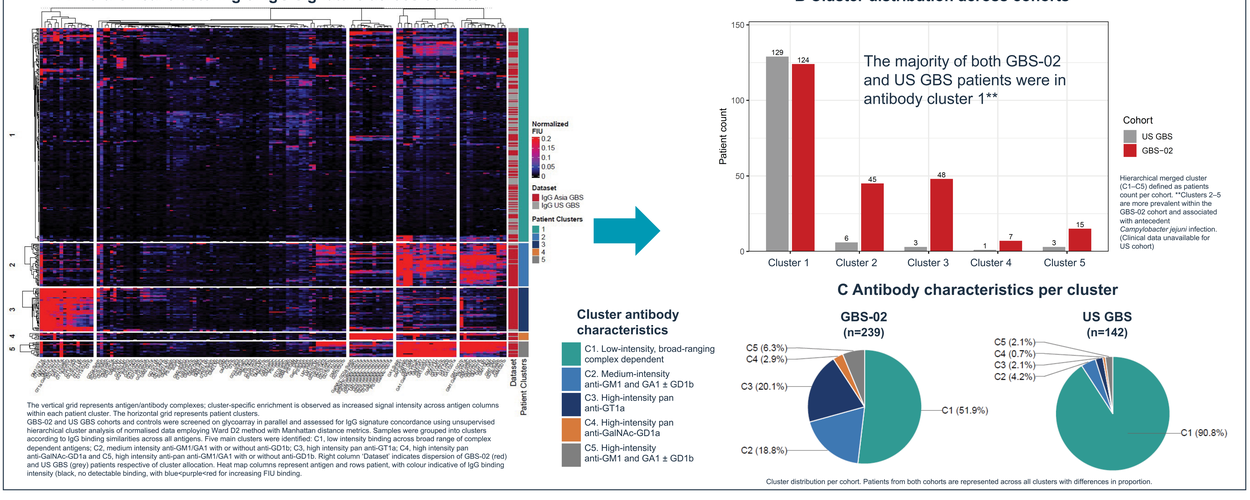
A The glycoarray is a powerful multiplex assay for high throughput screening of serum samples against a large panel of unique glycolipid and phospholipid antigens. **B** Close proximity of lipid molecules influence antibody binding via steric hindrance, antigen spacing, optimal epitope presentation, and/or formation of neoepitopes. **C** Glycoarray consists of 15 single glycolipids and one phospholipid in addition to 120 1:1 heteromeric complexes (v:v) in duplicate. Antibody (IgG, IgM, and/or IgA) binding to unique antigen is measured as fluorescent intensity units (FIUs) on a semi-quantitative scale from 0 to 65,535 (no detectable signal to signal saturation) and compared with regional healthy controls to establish baseline range of innate anti-carbohydrate antibody.

Results: The spectrum of IgG signatures are concordant between GBS-02 and an independent US GBS cohort

- IgG reactivity to one or more glycolipids/complexes was detected in 95.0% of GBS-02 and 82.4% of the US GBS cohort at baseline
- High-intensity (abundance and/or affinity) IgG signatures were observed in both cohorts, with reactivity to GM1, GA1, GD1b, GT1a, GD1a, and/or GalNAc-GD1a, independent of complex with other glycolipids ('pan' reactivity; **Figure 2**)
- Low-intensity (abundance and/or affinity) IgG signatures of broad-ranging, glycolipid complex-dependent reactivity were observed across both cohorts (GBS-02: 51.9%; US GBS: 90.8%), representing the predominant cluster 1 (**Figure 2**)

- **Baseline IgG autoantibodies are highly prevalent in GBS**
- **Five antibody clusters (1–5) conserved across GBS-02 and US cohorts, consistent with common disease biology**
- **Cluster 1 (low affinity/broad glycolipid antigen reactivity) predominant in both populations**

Figure 2. Distribution of anti-glycolipid IgG signatures across US and GBS-02 cohorts

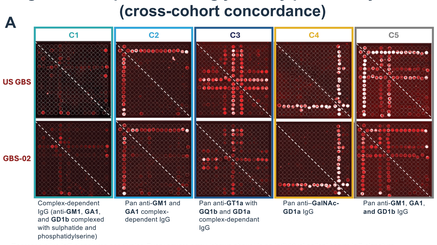


The vertical grid represents antigen/antibody complexes; cluster-specific enrichment is observed as increased signal intensity across antigen columns within each patient cluster. The horizontal grid represents patient clusters.
GBS-02 and US GBS cohorts and controls were screened on glycoarray in parallel and assessed for IgG signature concordance using unsupervised hierarchical cluster analysis of normalised data employing Ward D2 method with Manhattan distance metrics. Samples were grouped into clusters according to IgG binding similarities across all antigens. Five main clusters were identified: C1, low intensity binding across broad range of complex dependent antigens; C2, medium intensity anti-GM1/GA1 with or without anti-GD1b; C3, high intensity pan anti-GT1a; C4, high intensity pan anti-GalNAc-GD1a and C5, high intensity anti-pan anti-GM1/GA1 with or without anti-GD1b. Right column 'Dataset' indicates dispersion of GBS-02 (red) and US GBS (grey) patients respective of cluster allocation. Heat map columns represent antigen and rows patient, with colour indicative of IgG binding intensity (black, no detectable binding, with blue>purple>red for increasing FIU binding.

Cluster distribution per cohort. Patients from both cohorts are represented across all clusters with differences in proportion.

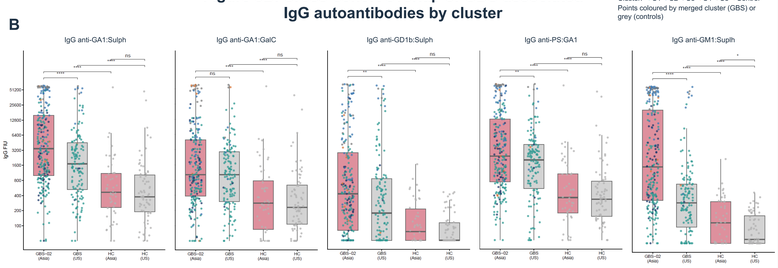
- Key glycolipid features driving cluster separation are consistently enriched in both datasets, supporting concordant biological signatures and confirming association of antibody reactivity with disease subtypes (**Figure 3A**)
- Glycolipid features are highly concordant between US and GBS-02 cohorts, with aligned direction and magnitude (**Figure 3B**)

Figure 3A. Representative glycoarray patterns by cluster (cross-cohort concordance)



Representative IgG signatures per cluster. Every square array (shown above) is an individual patient serum IgG signature. C1 features low-intensity, complex-dependent binding including IgG reactivity to GM1, GA1, GD1b, GT1a, GQ1b, LM1 and/or GalNAc-GD1a heteromeric complexes, and with no anti-glycolipid IgG binding detected in some patients. C1 examples shown demonstrate enhanced binding to GM1, GA1, or GD1b when complexed with sulphatide or phosphatidylserine. C2–C5 signatures are generally defined by medium-high intensity pan-antigen binding, irrespective of heteromeric complex. Red spots indicate IgG binding per antigen, with white spots indicating maximum binding (65,535 FIUs). Clusters 2–5 are more prevalent within the GBS-02 cohort and are associated with antecedent *Campylobacter jejuni* infection (clinical data unavailable for US cohort).

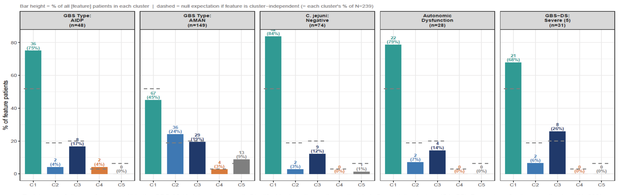
Figure 3B. Cross-cohort overlap of GBS-associated IgG autoantibodies by cluster



IgG heteromeric complex-specific intensity distribution per cohort and regional controls. Glycolipid complexes are significantly elevated in both cohorts relative to controls. IgG-enriched binding to GM1-, GA1-, and GD1b-containing complexes is measured within all clusters and across both GBS cohorts, with significant increase in GBS-02 driven in part by increased prevalence within C2 and C5. No significant difference determined for GA1:GalC complex between GBS cohorts. Typically, lower intensity reactivity was observed with C1 with overlapping distribution measured across US and GBS-02 populations. Wilcoxon test p-value: **** <0.0001, ** <0.01.

- Disease characteristics (acute inflammatory demyelinating polyradiculoneuropathy [AIDP], *Campylobacter jejuni*-negative serology, autonomic dysfunction) are dominant in cluster 1 vs other GBS-02 clusters (**Figure 4**). Clinical and electrophysiologic data were limited/unavailable for the US cohort, restricting cross-cohort phenotype comparisons

Figure 4. Across multiple disease characteristics, cluster 1 predominates in GBS-02



Enrichment of disease features within the GBS-02 (Asia) cohort overall. Stratified by baseline anti-ganglioside IgG reactivity clusters (C1–C5). Percentage for the overall cohort (N=239) shown by dashed lines for each feature. Clinical and electrophysiologic data were limited/unavailable for the US cohort, restricting cross-cohort phenotype comparisons.
AMAN, acute motor axonal neuropathy.

Conclusions: Anti-glycolipid IgG signatures across GBS cohorts show cross-regional concordance

- Autoantibodies to glycolipids were detected in both a GBS-02 (Bangladesh/Philippines) and US cohort, consistent with their central role in disease
- Baseline IgG reactivity patterns were concordant across the GBS-02 and US GBS cohorts
- These findings support biological comparability across cohorts; further studies linking antibody signatures to clinical phenotype and outcomes are warranted

References

1.Willison HJ, et al. *Lancet*. 2016;388:717–27. 2. Gorson KC. *Front Neurol*. 2025;16:1572949. 3. Pascual-Goñi E. *Neurology*. 2024;103:e209725. 4. Kroon H-A, et al. Presented at the Neuromuscular Study Group Annual Scientific Meeting, September 20–22, 2022; Tarrytown, NY, USA. 5. Lansita JA, et al. *Int J Toxicol*. 2017;36:449–62. 6. Suri P, et al. *Neurology*. 2022;98(18 Suppl):3867. 7. Thomma RCM, et al. *Brain*. 2025;148:4000–15.

Acknowledgments

The study was sponsored by Annexon Biosciences (Brisbane, CA, USA). We thank Ethan Loo (Excelra) for additional bioinformatic analysis. Medical writing and editing assistance were provided by Envision Ignite, an Envision Medical Communications agency, a part of Envision Pharma Group, and were funded by Annexon Biosciences.

Disclosures

SH: Research funding from Annexon Biosciences and argenx. H-AK, PP: Employee and shareholder of Annexon Biosciences. DL: External contractor of Annexon Biosciences to perform data analysis. ZI: Research funding from Fogarty International Center, National Institute of Neurological Disorders and Stroke of the National Institutes of Health, USA, and Annexon Biosciences. KS: Research funding from NINDS/NIH and DoD. Consultant for Annexon Biosciences. KCG: Consultancy/advisory role with Annexon Biosciences, argenx, Janssen, and Sanofi. HW: Research funding, consultancy payments, and share options from Annexon Biosciences.

For additional information, please reach out to: Susan Halstead (susan.halstead@glasgow.ac.uk) and/or Preeti Paliwal (ppaliwal@annexonbio.com).