Supplemental Information

HIV-associated alterations of the biophysical features of maternal antibodies correlate with their reduced transfer across the placenta

Sean A. Taylor[a#], Shilpee Sharma[b#], Christopher A. L. Remmel[a], Beth Holder[c], Christine E. Jones[d*], Arnaud Marchant[b*], and Margaret E. Ackerman[a*]

[a]Thayer School of Engineering, Dartmouth College, Hanover, NH, USA

[b]Institute for Medical Immunology, Université Libre de Bruxelles, Brussels, Belgium

[c]Institute of Reproductive and Developmental Biology, Department of Metabolism, Digestion and Reproduction, Imperial College, London, UK

[d]Faculty of Medicine and Institute for Life Sciences, University of Southampton and NIHR Southampton Clinical Research Facility and NIHR Southampton Biomedical Research Centre, University Hospital Southampton NHS Foundation Trust, Southampton, UK

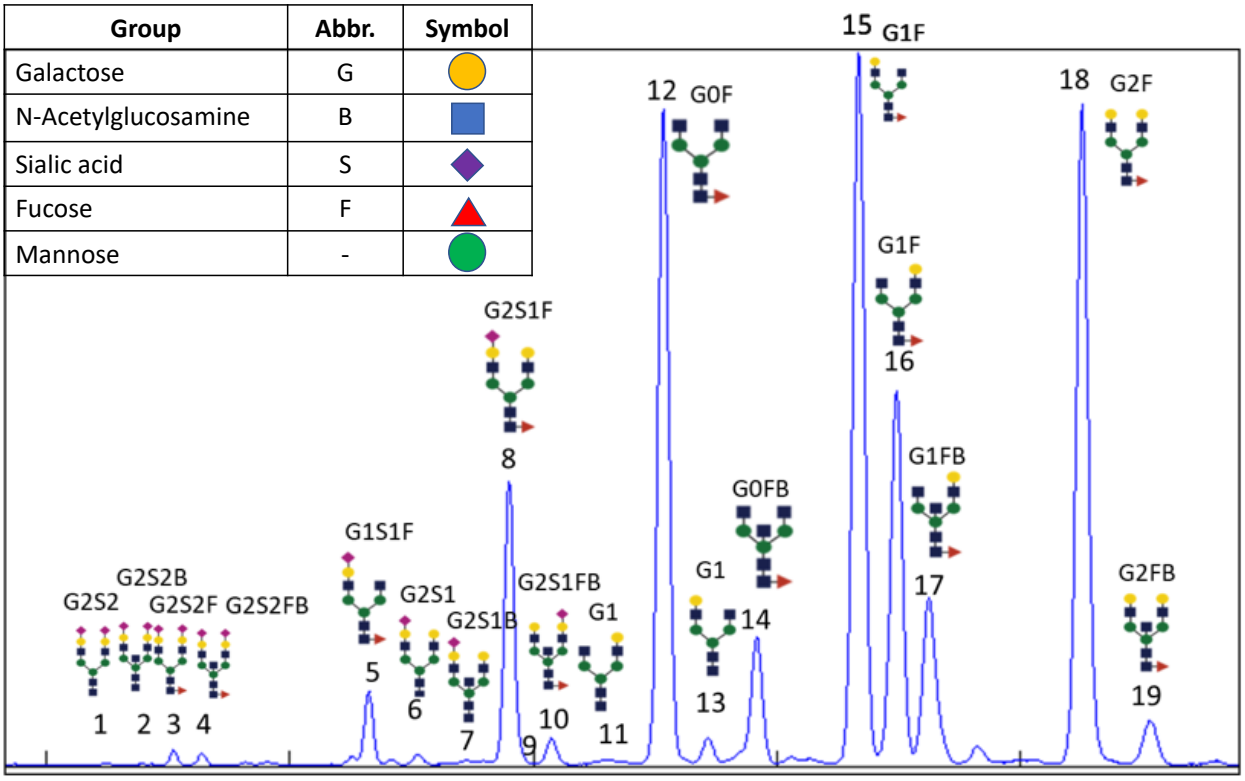
[#]Share first authorship

*Corresponding authors:
Christine E. Jones
NIHR Southampton Clinical Research Facility and Biomedical Research Centre
University Hospital Southampton, Tremona Road
SO16 6YD Southampton, United Kingdom
Email: C.E.Jones@soton.ac.uk

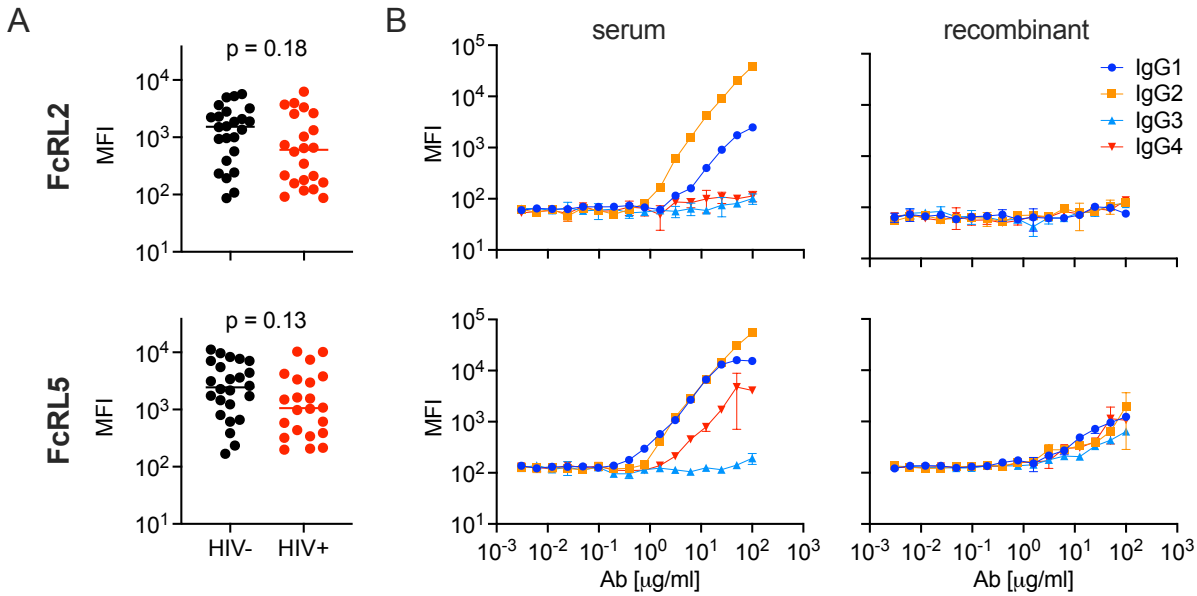
Arnaud Marchant
Institute for Medical Immunology
Campus Erasme
808 Route de Lennik
1070 Bruxelles, Belgium
Email: arnaud.marchant@ulb.be

Margaret E. Ackerman
Thayer School of Engineering, Dartmouth College
14 Engineering Dr
Hanover, NH 03755 USA
Email: Margaret.E.Ackerman@Dartmouth.edu

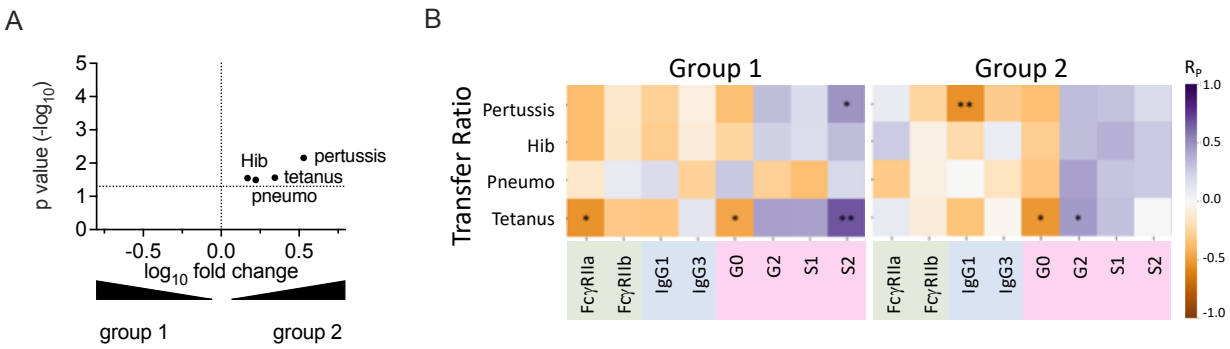
Supplemental Figures	
Supplemental Figure 1	Representative CE spectrum for glycan analysis by capillary electrophoresis
Supplemental Figure 2	FcRL binding according to infection status and among serum and recombinant IgG subclasses
Supplemental Figure 3	Relationships between transfer efficiency and serum IgG characteristics by UMAP group
Supplemental Tables	
Supplemental Table 1	Study reagents



Supplemental Figure 1: Representative CE spectrum for glycan analysis by capillary electrophoresis. Fluorescently-labeled Fc glycans were analyzed by capillary electrophoresis. Individual peaks and their representative glycan structures are shown. The core glycan, consistent across IgG Fc glycoforms in not included in the text label for each glycan.



Supplemental Figure 2: FcRL binding according to infection status and among serum and recombinant IgG subclasses. A. Maternal serum antibody binding to FcRL2 (top) and FcRL5 (bottom) according to HIV infection status and UMAP group. Statistical significance determined by unpaired t-testing. **B**. Binding of serum-derived (left)and recombinantly expressed (right) IgG subclasses (Ab) to FcRL2 (top) and FcRL5 (bottom). Mean fluorescent intensity (MFI) is reported for multiplex assay results. Lines indicate group mean. Error bars indicate standard deviation.



Supplemental Figure 3: **Relationships between transfer efficiency and serum IgG characteristics by UMAP group. A.** Volcano plot showing the magnitude and statistical confidence (unpaired t test) of differences in antibody transfer ratios between subjects according to UMAP group status. The dotted horizontal line represents a p value of 0.05 and dotted vertical lines indicates no fold-change. **C**. Heatmaps depicting magnitude and statistical significance of Pearson correlation coefficients (R_P) between transfer ratios and IgG subclasses,Fc receptor binding, and the prevalence of major Fc glycoforms in UMAP groups 1 (left) and 2 (right). Color indicates correlation coefficient magnitude and direction, significance is indicated as *$p < 0.05$; **$p < 0.01$.

Supplemental Table 1. Study Reagents

Regeant	Source	Catalog number/reference
anti-Ig	Southern Biotech	JDC-10
anti-IgG1	Invitrogen	A10630
anti-IgG2	Southern Biotech	31-7-4
anti-IgG3	Invitrogen	053600
anti-IgG4	Southern Biotech	HP6025
Goat anti-human IgG Fc-PE	Southern Biotech	1030-09
Mouse anti-human IgG1 Fc-PE	Southern Biotech	9054-09
Mouse anti-human IgG2 Fc-PE	Southern Biotech	9070-09
Mouse anti-human IgG3 Fc-PE	Southern Biotech	9210-09
Mouse anti-human IgG4 Fc-PE	Southern Biotech	9200-09
FcγRI	Boesch, 2014	doi: 10.4161/mabs.28808
FcγRIIA	Boesch, 2014	doi: 10.4161/mabs.28808
FcγRIIB	Boesch, 2014	doi: 10.4161/mabs.28808
FcγRIIIA	Boesch, 2014	doi: 10.4161/mabs.28808
FcRn	Feng, 2020	doi: 10.1016/j.pep.2011.03.012
FcRL2	R&D Systems	2048-FC-050
FcRL5	R&D Systems	2078-FC-050
FcgBP	Novus Biologicals	NBP190462PEP
FcRLB	R&D Systems	4868FC050