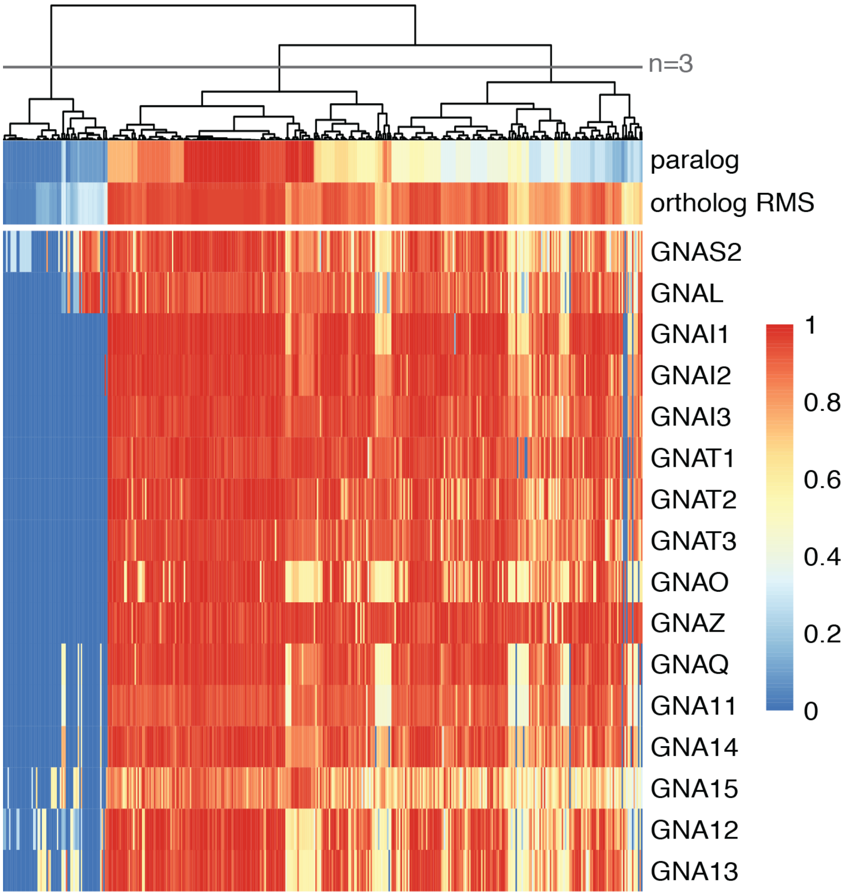

Supplementary Figure 1 | Selectivity barcode of G proteins. Hierarchical clustering of G protein conservation scores in ortholog alignments of G protein subfamilies and the human paralog alignment. The orthoRMS (root mean square of the conservation of a CGN position in all 16 subfamily alignments) gives a score for conservation of each CGN position (G protein numbering) across all G protein subfamilies (1 conserved in all subfamilies, 0 conserved in none, 0-1 conserved in few to many subfamilies). Hierarchical clustering of the subfamily and paralog positional conservation reveals three main clusters: (1) positions conserved in both paralog alignment and the G protein subfamilies (highly conserved positions); (2) positions conserved in the ortholog alignments but not in the paralog alignment (subtype-specifically conserved positions); (3) positions not conserved in the paralog nor the ortholog alignments (neutrally evolving positions). As this analysis assigns these roles to CGN positions (as we base it on a protypical G protein) rather than for individual G proteins, we used discrete cut-offs in the main manuscript to provide a simplified G protein barcode for each subfamily individually. We provide the readers/users the opportunity to choose their own cut-offs in the GPCRdb web resource for identifying such positions each of the 16 Gα proteins (e.g. for GNAS2 http://www.gpcrdb.org/signprot/gnas2_human/). In this manner, researchers can be liberal or conservative in inferring such positions in any human Gα protein of interest.