Multi-regional comparative N-glycomics reveals both spatial and phylogenetic gradients in mammalian brain N-glycome complexity

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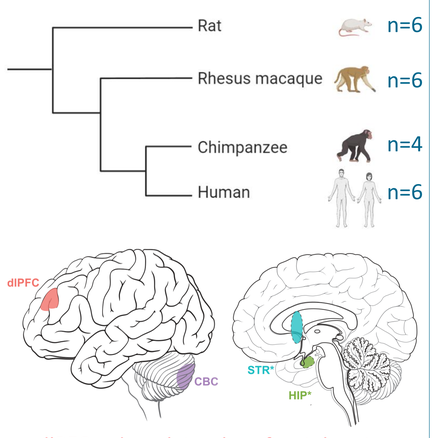
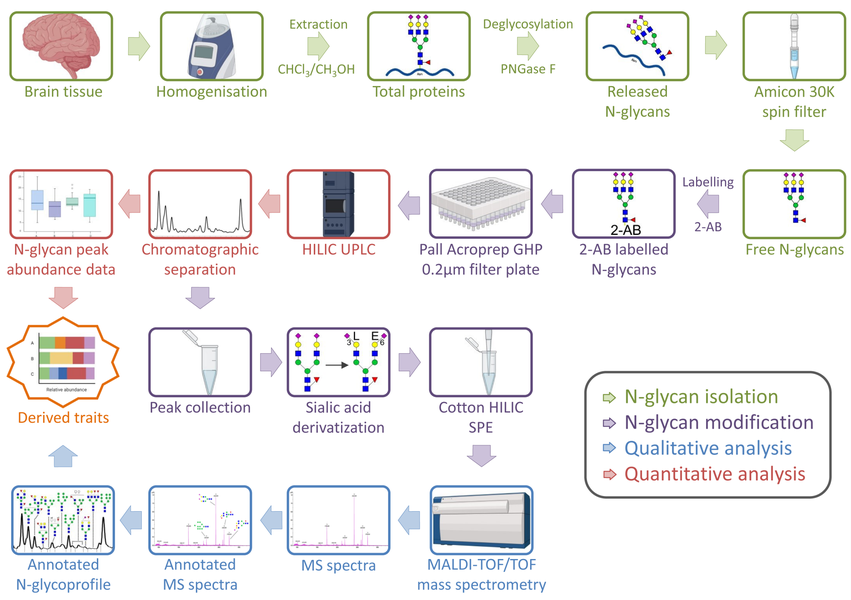
Study design

Four brains regions from four mammalian species



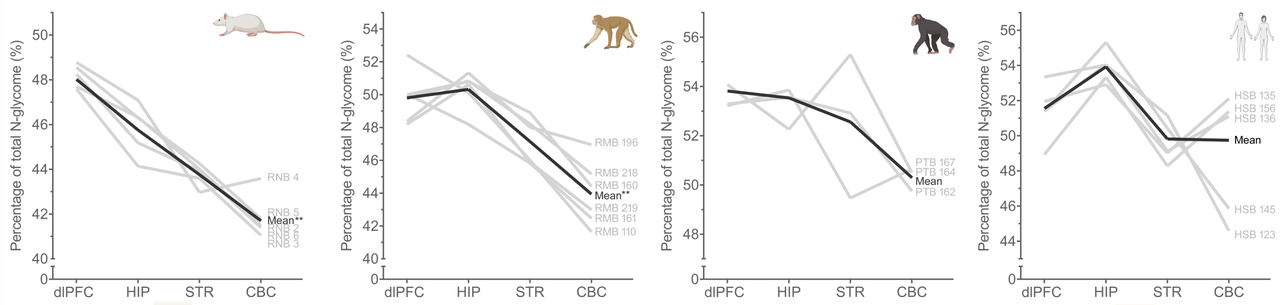
dlPFC – dorsolateral prefrontal cortex
CBC – cerebellar cortex
STR – striatum
HIP – hippocampus
*regions not visible from this view

Sample preparation and analytical workflow

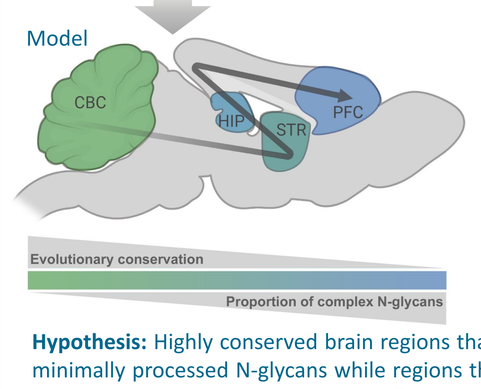


Conserved anatomical gradient in brain N-glycome complexity

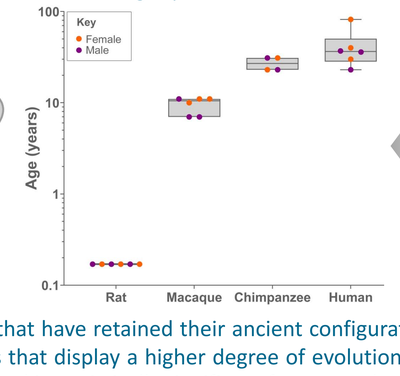
Total abundance of complex N-glycans by brain region in each species



Model



Age spread of donors



Weakest trend seen in human cohort which also had greatest variation in:

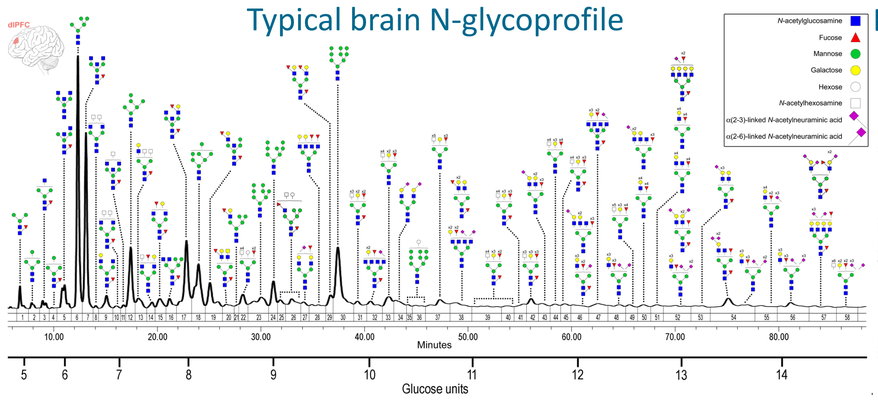
- age
- post-mortem interval
- genome
- environment
- diet
- lifestyle

} epigenome

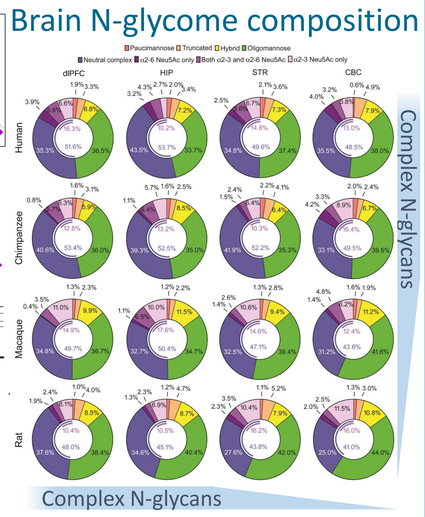
Hypothesis: Highly conserved brain regions that have retained their ancient configuration have a higher proportion of ancient, minimally processed N-glycans while regions that display a higher degree of evolutionary change across species have acquired N-glycoprofiles of greater complexity. This trend is statistically significant for rats and macaques; to be validated for hominids.

Overview of the mammalian brain N-glycome

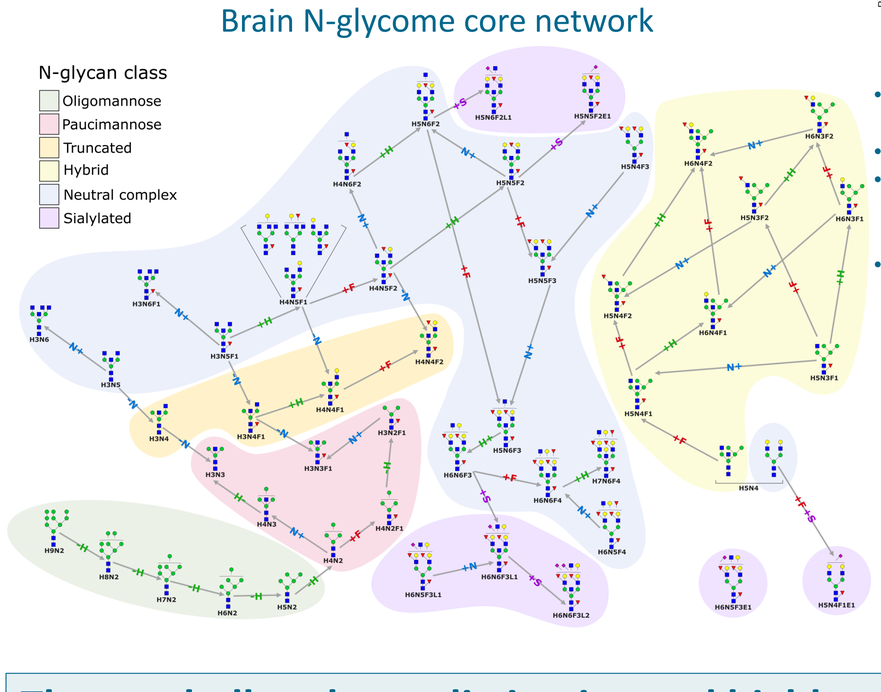
Typical brain N-glycoprofile



Brain N-glycome composition

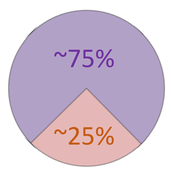


Brain N-glycome core network



- 287 distinct N-glycan structures detected across all species and brain regions
- Only 60 are ubiquitous → 'core network'
- Core N-glycans are biosynthetically related → constitute the dominant N-glycosylation pathway in the brain
- They are overrepresented → combined abundance is ~75% of brain N-glycome

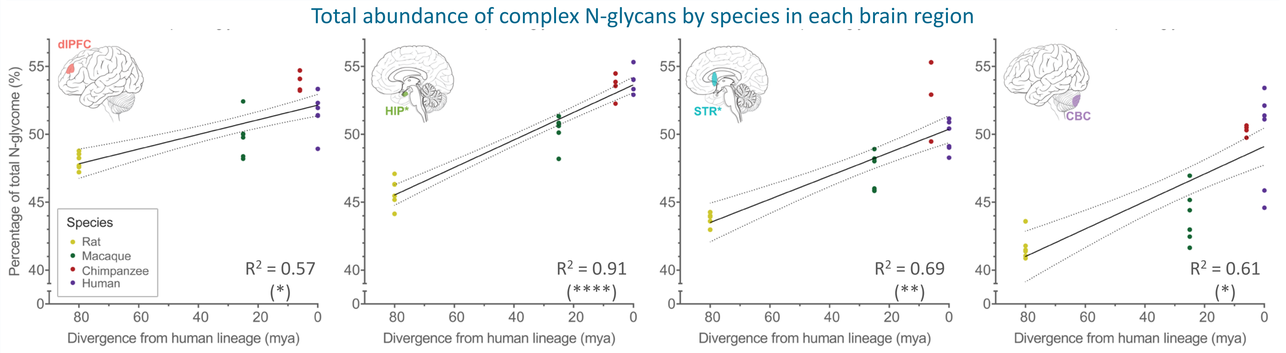
- Conserved 'core network' (60 N-glycans)
  - Ubiquitous brain proteins?
  - Brain homeostasis?



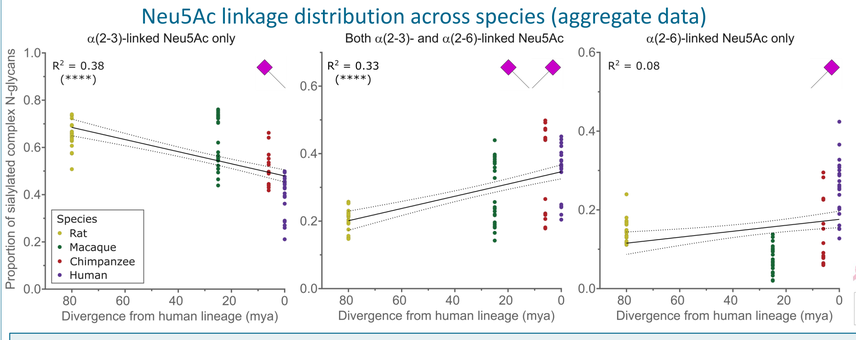
- Region/species-specific (>220 N-glycans)
  - Diverse, highly variable
  - Specialised brain functions?

Global increase in N-glycome complexity in the hominid lineage

Total abundance of complex N-glycans by species in each brain region

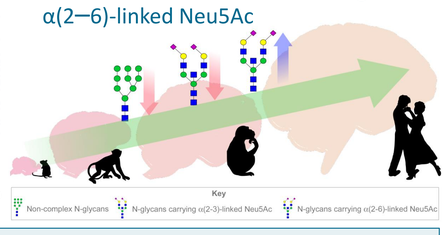


Neu5Ac linkage distribution across species (aggregate data)



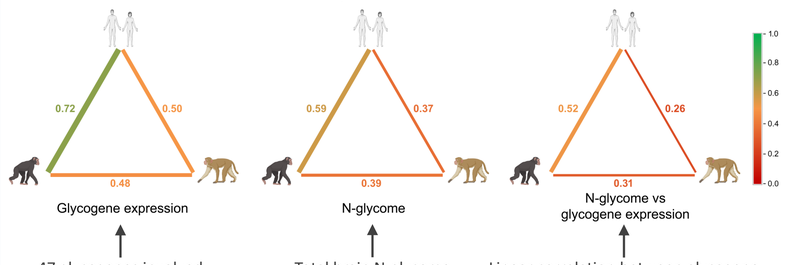
Hominid brain evolution has been characterised by two global trends:

- an increase in N-glycome complexity
- a shift towards increased usage of α(2—6)-linked Neu5Ac



Rapid evolutionary divergence of the primate brain N-glycome

Inter-species correlation



47 glycogenes involved in N-glycan synthesis

Total brain N-glycome

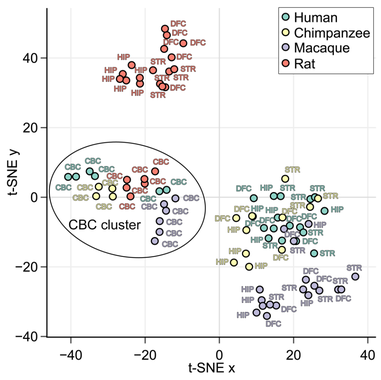
Linear correlation between glycogene expression and the N-glycome phenotype

Integration of N-glycomic and complementary bulk RNA-seq transcriptomic data revealed that the primate brain N-glycome evolves more rapidly than the underlying transcriptional framework.

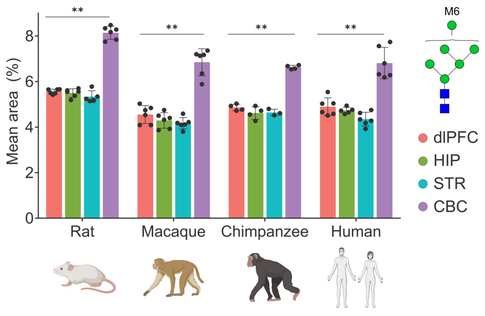
This supports the notion that post-translational modification of conserved proteins via N—glycosylation is a means of relatively rapidly generating additional inter-species diversity among genetically similar organisms.

The cerebellum has a distinctive and highly conserved N-glycome

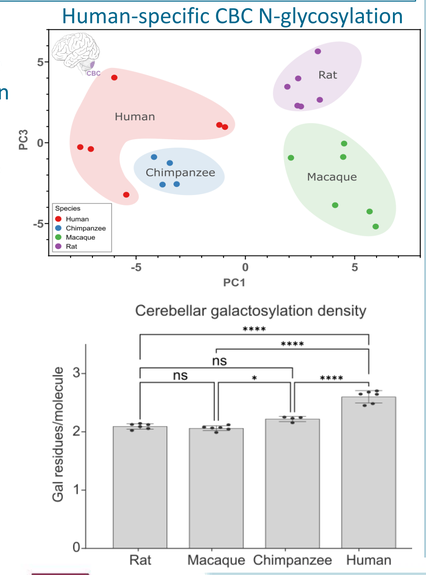
Global N-glycome clustering by PCA



The defining feature of the cerebellar N-glycome is an abundance of the M6 oligomannose N-glycan



Human-specific CBC N-glycosylation



Conclusions

The composition of the brain N-glycome is generally well conserved across different brain regions and mammalian species - a core network of N-glycans defines the archetypal brain N-glycoprofile. Nevertheless, in comparison to glycogene expression networks, the brain N-glycome evolves rapidly resulting in a wealth of both spatial and inter-species variation. As a result of overlapping anatomical and phylogenetic gradients in the abundance of complex N-glycans, peak N-glycome complexity was found in the hominid cortical regions. We hypothesise that increased diversity and complexity of sugar modifications on neural N-glycoproteins contributed to the emergence of novel cognitive functions, including those unique to the human neocortex.

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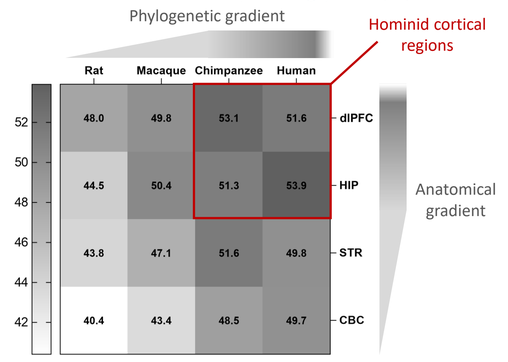
Mean abundance of complex N-glycans (%)



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