

Age-dependent changes in lipid composition of cerebral organoids relevant to Alzheimer's disease

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OVERVIEW

Aim: To utilize a 3D cell biology model of the human brain tissue and analyze aging-induced changes in lipid composition in the cerebral organoids and differential lipid composition between organoids with AD mutation and WT.

Methods: Lipids extracted in an organic solvent were quantified using selected reaction monitoring (SRM) assay combined with UHPLC.

Achievements: Aging-induced changes in membrane glycolipids were observed during the growth of the cerebral organoids.

INTRODUCTION

- Lipid rafts are lipid enriched microdomains containing glycolipids and cholesterol, aiding in anchoring transmembrane proteins and signal transduction.
- Gangliosides (GS's) are glycosphingolipids with one or more sialic acid residue in glycan moiety present in the lipid draft and known to sequester A β [1].
- During aging and neurodegeneration, the membranes' physicochemical properties are altered, so profiling these lipids could augment our understanding of the pathogenesis of Alzheimer's disease (AD).

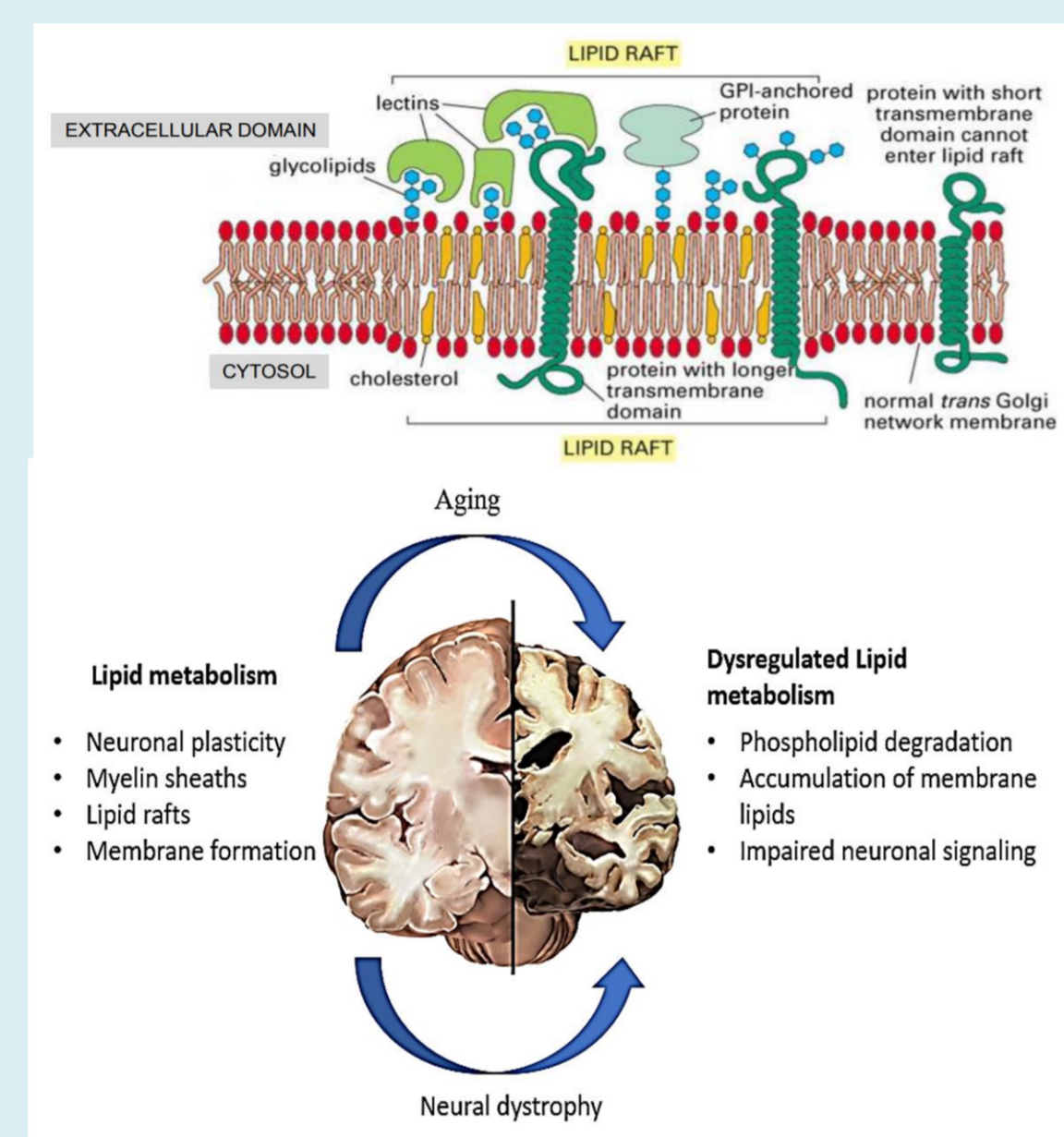


Figure 1: GS in lipid raft [2] and the aging-induced dysregulation of lipid metabolism relevant to AD pathogenesis

METHODS

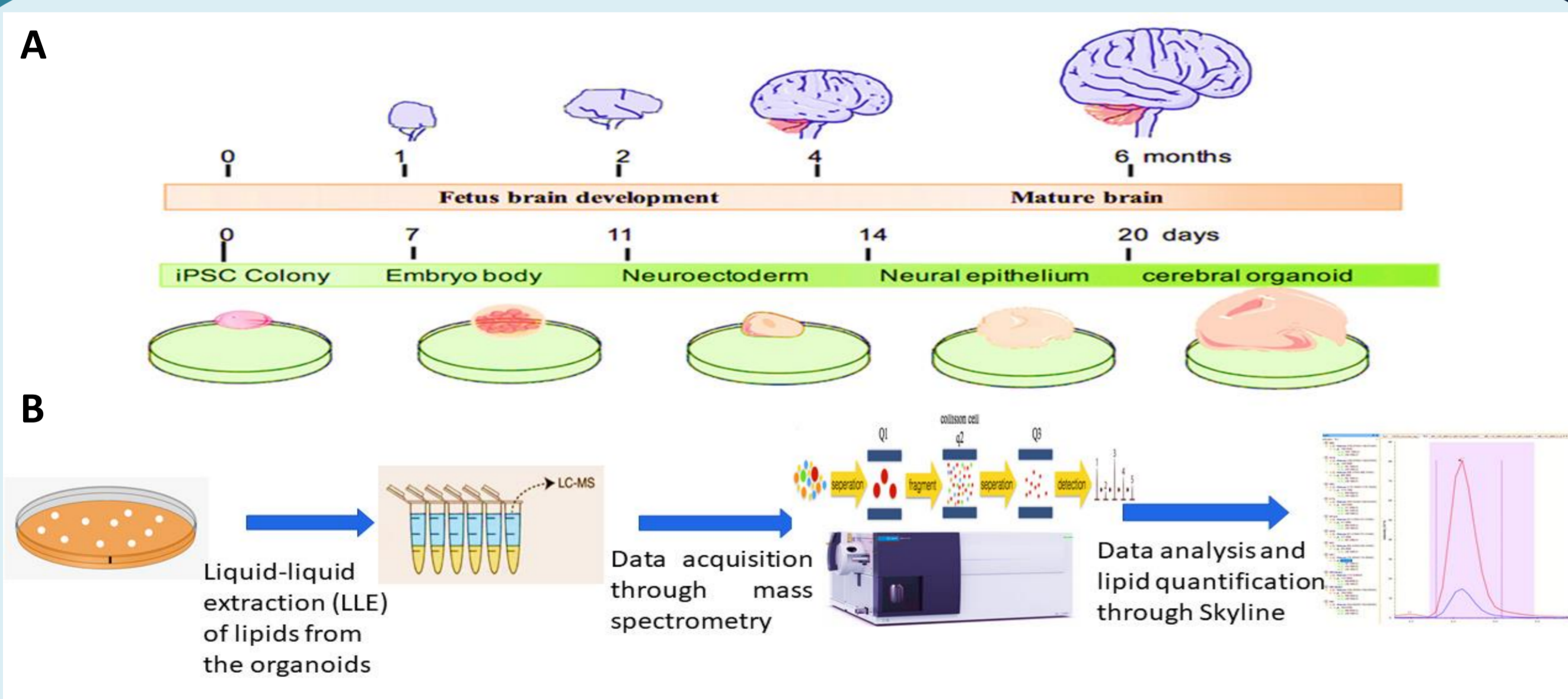


Figure 2: A. The organoids are a 3D cellular system that could be a model system for brain development and A β pathologies relevant to AD [3].

B. General workflow of the analysis of membrane lipids in cerebral organoids.

- Cerebral organoids (CO's) were cultured and provided to us by the Department of Histology and Embryology, Faculty of Medicine, MU.
- CO's were harvested at six different time points - 48, 75, 95, 100, 135, 160 days.
- CO's were washed with cell recovery solution immediately after harvesting to effectively removed the matrigel utilized for their cultivation. Next, CO's were washed with PBS afterward and lyophilized.
- Lipids were extracted using 80% isopropanol followed with vortex and ultrasonication, which were analyzed in both positive and negative mode in an SRM assay.
- The raw files were processed in the skyline, and quantification was done using labeled internal standards.

RESULTS

- Brain development is known to be accompanied by changes in the expression of glycosphingolipids.
- GS's, which are expressed at the early stages of development (neural stem cells), are simpler gangliosides such as GD3 and GM3, which contain only glucose, galactose, and sialic acid (1-2).
- GS containing complex carbohydrate moieties are elevated during neuro- and astrocytogenesis [4].
- With aging, the level of gangliosides decreases significantly.

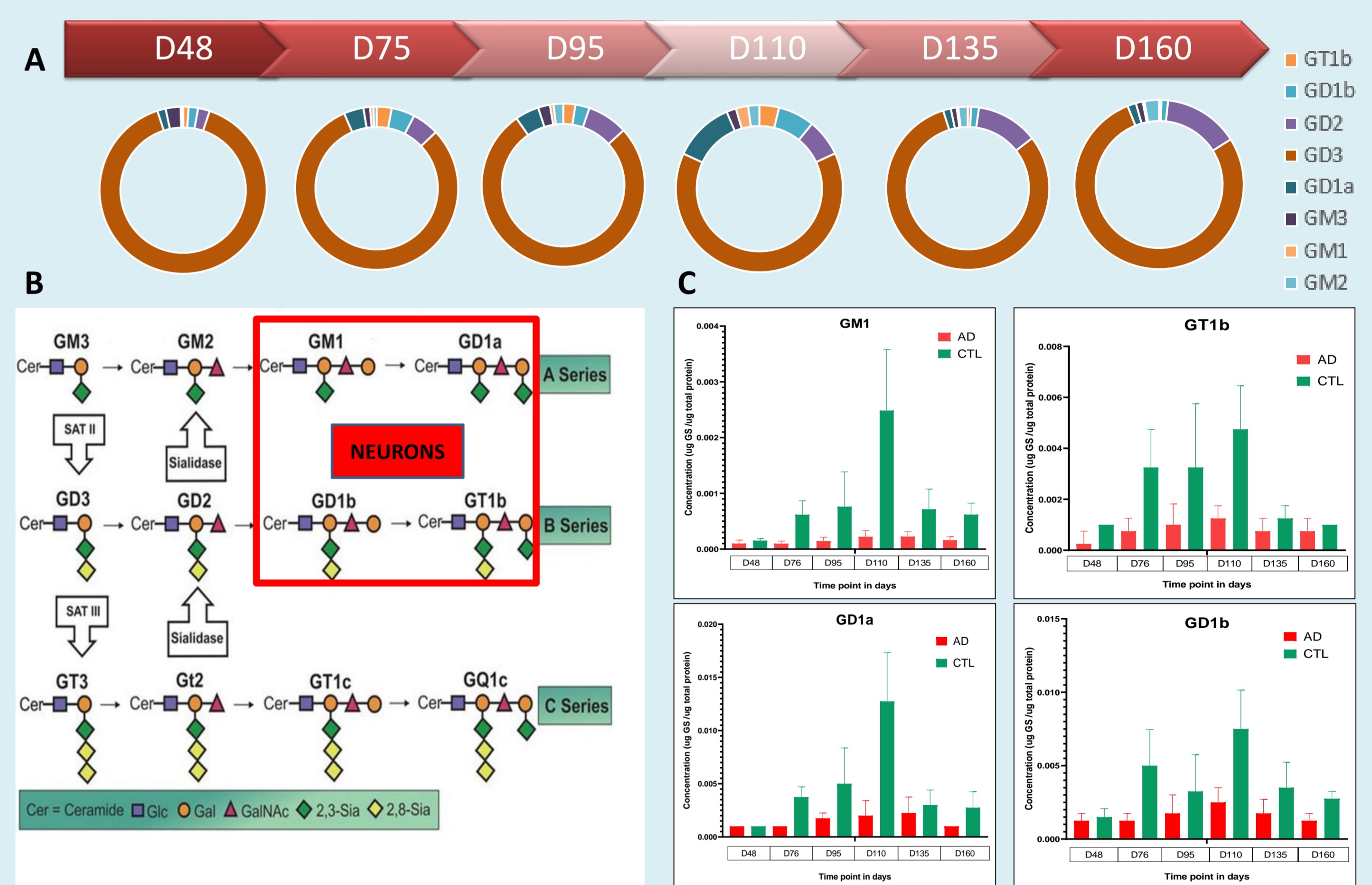


Figure 3: A. The pie chart shows the predominance of simpler gangliosides such as GD3 and GM3 at the early developmental phase in healthy organoids and then gradual decline as the organoids are aged, while giving room to complex GS's – GM1, GD1a, GD1b, and GT1b.

B. GS synthesis pathways. The GS's enclosed in the box are the ones available on the neuronal membrane [4].

C. Levels of the complex GS's. The organoids had the maximum level of these GS's by 110 days, after which it declined in older organoids. The decline in the levels of these GS's was even more pronounced in the AD (red) organoids compared to control organoids (green).

CONCLUSIONS

- We observed an increase in the GS's concentration in developing organoids, which, however, declined in old organoids.
- In AD organoids, the GS's amount was lower than that of control organoids which could indicate the importance of membrane lipids in the regular neuronal function.
- CO's were able to portray developmental changes in a short period which, highlights their role as an efficient and appropriate cerebral model system.

References:

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