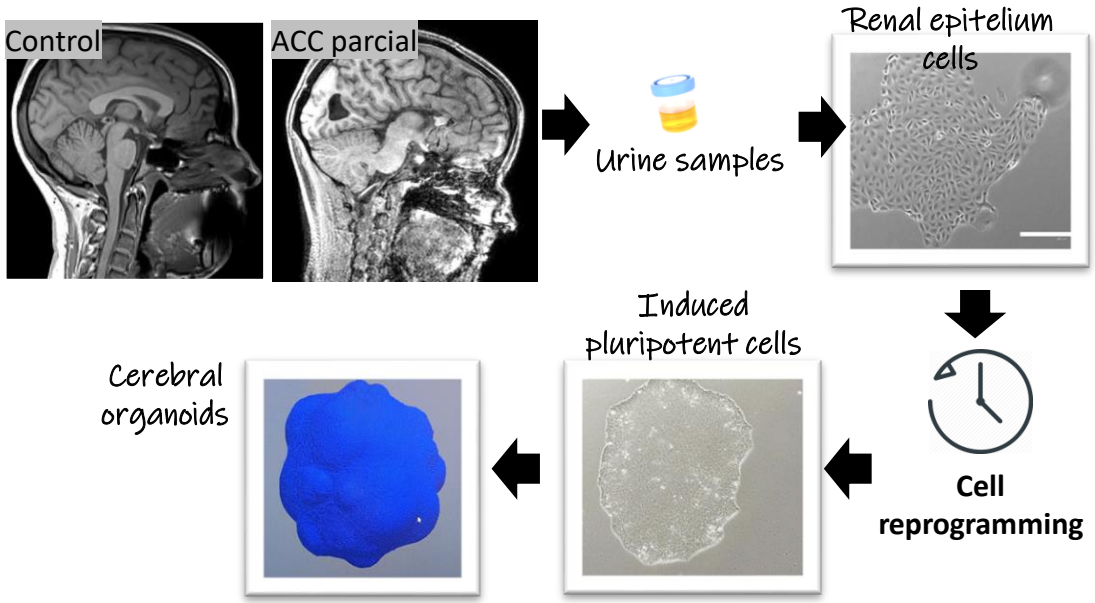
MORPHOLOGICAL ANALYSIS OF INDUCED PLURIPOTENT STEM CELLS DERIVED CEREBRAL ORGANOIDS FROM CALLOSUM DYSGENESIS PATIENTS AND CONTROL SUBJECTS

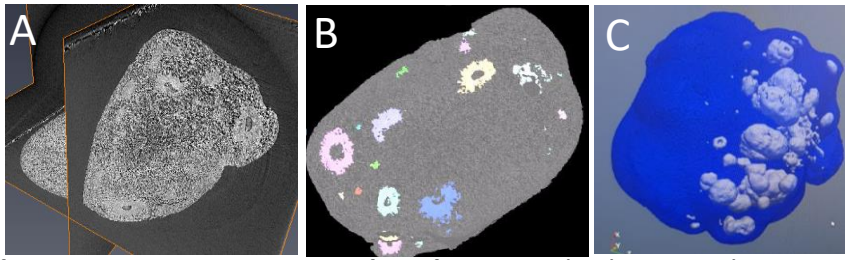
Livia Goto-Silva[1], Caroline V.F. Lima[1], Carlos S. B. Dias[2], Mariana P. Stelling[3], Simone C. Cardoso[4], Steven K. Rehen[4], Fernanda Tovar-Moll[1].

1- D'Or Institute for Research and Education; 2- Brazilian Synchrotron Light National Laboratory; 3- Federal Institute for Education; 4- Federal University of Rio de Janeiro.

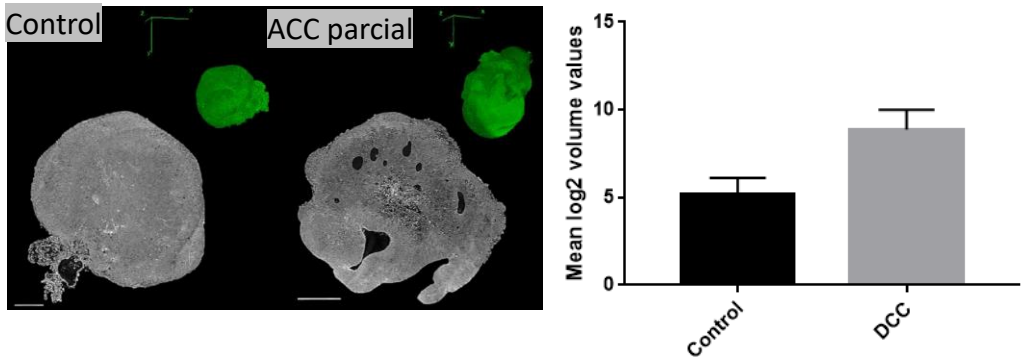
Dysgenesis of the corpus callosum (DCC) is a neurodevelopmental disease in which the corpus callosum (CC), the largest commissure in the brain, is absent or reduced. Although not having part or all the CC, DCC patients do not show a typical inter-hemisphere disconnection syndrome. This phenomenon has remained as a neurological paradox for decades and it is still subject of studies to which our team has intensively contributed. The aim of our group is to understand the mechanisms of plasticity in normal and DCC brains. Our approach is to use brain organoids as an ex-vivo model to investigate the molecular and cellular basis of DCC. Here we characterized morphological changes in DCC organoids using microtomography.



Obtaining cerebral organoids from subjects in the study: urine from patients and controls (T1 image) was collected. Urine cells were cultivated and reprogrammed to pluripotency using Sendai vírus. Cerebral organoids were produced following a published protocol (Goto-Silva & Ayad, 2019)



Organoid processing, acquisition and analysis. Cerebral organoids were stained with osmium tetroxide, dehydrated and embedded in paraffin. Acquisition covered an angular range of 180 degrees to compose a sinogram (1024 projections - exposure time of 1 s/projection). Reconstruction used the raft algorithm. Images A-D: 45 day control organoid. A) orthogonal view, arrows: rosettes (neurogenic regions) ; B) segmentation of rosettes; C) rosette volume.



DCC organoids show increase in rosette volume. Volume of rosettes was measured in control and DCC organoids at 25 days.

Conclusion: Our results are promising to the analysis of morphological features comparing DCC patients and controls. Increase in rosette volume suggests an increase in proliferative cells in patients' organoids that is being investigated using complementary techniques.

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