Human neural organoids for the mechanistic study of neural tube defects. The example of folate receptor 1.

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ABSTRACT

Neural tube defects (NTDs) are the second most common and serious birth defects that occur during early pregnancy when the neural tube fails to form and close. It is estimated that more than 260,000 pregnancies are affected by NTDs. Epidemiological studies have shown that a significant proportion of NTDs are folic acid sensitive bur remarkably, the mechanisms by which folate prevents neural tube defects and why there are folate-sensitive and folate insensitive NTDs are unclear. Folate has been identified as a critical nutrient in preventing neural tube defects (NTDs), prominent birth defects both in incidence and devastating consequences to fetal and infant health, by unclear mechanisms. NTDs occur when the neural tube fails to close at four weeks of pregnancy.

We have developed a neural tube organoid model derived from human induced pluripotent stem cells (hiPSCs) and used it for the elucidation of the mechanism of action of folate/folate receptor 1 (FOLR1) during neural tube formation by generating folate receptor 1 knockdown or knockout neural organoids. The use of hiPSCs and organoids to investigate the mechanisms of NTDs is innovative. The bioengineering of hiPSC-derived, self-organizing, 3D neural organoids allows for easy manipulation while mimicking the neural tube structure in a remarkable way. This human cell-based in vitro 3Dmodel for neural tube formation is characterized by expression of the neural stem cell marker Sox2 and presence of structures with tubular morphology. FOLR1 expression and discover localizes to apicolateral regions of neural cells surrounding the lumen of newly formed neural tube-like structures. FOLR1 knockdown (FOLR1 KD) impairs formation of neural tubes in neural organoids, resulting in structures that do not exhibit the characteristic basal displacement of nuclei surrounding the lumen suggestive of deficient neural cell apical constriction. FOLR1-deficient cells fail to apico-basally elongate, resulting in aberrant tubular structure. FOLR1 KD-induced phenotype is rescued by incubating 3D human neural cell-based cultures from day 0with 50 μM pteroate, a folate precursor that binds to FOLR1 but cannot participate in metabolic pathways or be converted to folate by eukaryotic cells.[1]

Our work suggests a non-metabolic role of folate, separate from its role as a vitamin, through FOLR1, where FOLR1 depletion results in reduced Ca2+ dynamics and accelerated apical endocytosis, cell adherens junction turnover and failure of neural plate cells to apically constrict.

REFERENCES

[1] Olga A Balashova*, **Alexios A Panoutsopoulos***, et al.2024 Nature Communications, doi: 10.1038/s41467-024-45775-1