Advancing Human Brain Modeling with a Novel Microglia-Containing Organoid System: Validation and Strategies to Overcome Current Limitations

Sara Ferro[1], Anna Cascio[1,2], Paolo Vaccari[1] , Stefania Giussani[3], Oliver Harschnitz[3], Veronica Krenn[1,4,5]

E-mail: sara.ferro@unimib.it

[1] Department of Biotechnology and Bioscience, University of Milan-Bicocca, Milan, Italy

[2] DIMET PhD School, Dept. of Medicine and Surgery, University of Milan – Bicocca, Milan, Italy

[3] Neurogenomics Research Centre, Human Technopole, Milan, Italy

[4] Early Career Fellow, Human Technopole, Milan, Italy

[5] Milan Center for Neuroscience (NeuroMI), Italy

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Abstract: Human Organoids have revolutionized the study of brain development. By recapitulating the cellular composition and morphogenesis of the developing human brain, these systems now enable dissection of cellular and molecular mechanisms in a human setting with unprecedented accuracy. Their use and translational relevance is currently hindered by the lack of microglia, the resident immune cells of the brain, which play a pivotal role in sculpting the neural landscape, beyond their immune surveillance capacity. Increasing evidence links microglial dysfunction to neurodevelopmental disorders, including autism spectrum disorders. To overcome this limitation, we established a microglia-enriched brain organoid platform capable of recapitulating the main developmental stages of human microglia in vitro, including their yolk sac origin and colonizing the neural tube during early development. We show that yolk sac-like microglia progenitors, known as erythromyeloid progenitors, derived from hPSCs successfully incorporated into brain organoids and in situ reach a microglia-like identity. Furthermore, morphological and transcriptomic analyses demonstrate that these cells undergo a time-dependent in vivo-like maturation towards microglia identity, validating this in vitro platform for modelling human microglial development.

Using this platform, we show that TGFβ1 supplementation is critical for microglial survival and maturation and identified a crucial role of TGF-β signaling in promoting the emergence of molecular signatures related to metabolic reprogramming and immune sensing, pointing to a role of these processes in human microglia maturation. We will discuss ongoing strategies for functional validation of these effects, by establishing metabolic and immune challenges assays in microglia-containing organoids, as well as to validate a direct effect of TGF-β on microglia using a genetic model of TGF-β signalling deficiency.

Finally, we will present our ongoing efforts to optimize the production of microglia-containing organoids by increasing the efficiency of microglial progenitor generation. These improvements are expected to reduce cost and labor requirements, ultimately making this platform more accessible for experimental use.