Title: **Neonatal glial culture systems for in vitro modelling of the selected perinatal neurological disorders in physiologically normoxic conditions**

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Abstract:

Developing nervous system is extremely sensitive to influence of the extracellular clues. Even temporal alteration in tissue homeostasis or pathophysiological conditions might results in serious aberration in the central nervous formation and functioning, leading to long lasting neurological disorders. Preventing or curing the resulted disorders require the detailed knowledge about mechanisms of the cell response to the stimuli and its interactions with the neighbouring cells in the tissue microenvironment. To address this issue we have elaborated protocol of culturing rat neonatal glial cells (astrocytes, oligodendrocytes and microglia) in physiologically normoxic conditions, i.e. in 5 % oxygen concentration. The cells are cultured in the strictly controlled, minimal media, without addition of any supplements which could potentially modulate fundamental cell processes. The neonatal glial cells are to be cultured as monofractions or as co-cultures and could be used for creating the selected disease-in-a-dish models of perinatal disorders, like for instance neonatal asphyxia, neuroinflammation or hypoglycemia. Accordingly, in our studies on the in vitro model of perinatal asphyxia, evoked by temporal deprivation of oxygen and glucose (OGD), the response of each type of glial cells was shown to be different. Obtained data indicate OGD significantly ($p < 0.05$) enhance proliferation of astrocytes (by nearly 77 %) and oligodendrocyte progenitors (by approximately 62 %), as well as hampers their differentiation to myelinating cells. Contrary, neither the proliferation nor morphology of microglia was affected by the insult. In conclusion, the elaborated method of culturing the neonatal glial cells in physiologically relevant conditions might be useful for studies of basic mechanisms of cell response in the selected models of perinatal neurological disorders.

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