Q - 17 - ORGANOTYPIC CULTURES OF ADULT HUMAN BRAIN: A NOVEL MODEL TO STUDY AGE-ASSOCIATED NEURODEGENERATIVE DISEASES

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INTRODUCTION: Organotypic culture from adult human brain is a powerful method to study cellular and molecular aspects of neuropathologies, as well as the effects of candidate neuroprotective drugs. Advantages of this method include the maintenance of brain structure and connectivity, and the use of tissue usually discarded during surgical procedures. However, few efforts have been made to improve the quality and consequently widening the applications of such model. OBJECTIVES: Here, our main goal was to establish an optimized protocol for organotypic cultures from adult human brain cortex obtained from patients undergoing temporal lobectomy for treatment of refractory epilepsy (Etichs Committee approval HCRP 2634/2008). MATERIALS AND METHODS: Cortical fragments (1x1x1 cm) collected at the surgical room were immediately immersed in ice-cold oxygenated buffered saline, sliced using a vibratome and cultured for up to 4 days. DISCUSSION AND RESULTS: Based on cell viability (followed by MTT assay), we have determined the best thickness of the slices. Importantly, viability was not significantly reduced until day 4. Tissue integrity and quantification of cell types, assessed by HE staining and immunohistochemistry, respectively, showed no alteration in tissue integrity after processing, along with a moderate increase in the number of astrocytes and neuronal stability along 2 days in vitro. A significant decrease in microglial cells was also observed. In addition, neuronal activity, probed by ERK phosphorylation after KCl depolarization, was present at least until day 2. We have readily detected binding of Alzheimer disease-associated beta-amyloid oligomers to cultured slices, as detected by ELISA, suggesting that the model is amenable for modeling neurodegenerative diseases. We are currently studying the functional response of cultured tissue by means of electrophysiology. CONCLUSION: In conclusion, human adult cortical organotypic cultures derived from amygdalohippocampectomy remain viable and functional for ex vivo experiments for at least 2 days after surgery.

Keywords: human brain, organotypic culture, neuropathology

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