Rat microglia cells : their culture, isolation and phagocytic activity

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Abstrak

Microglia were isolated from mixed primary cell cultures of the cerebral cortex from 3 day old male Wistar rats. The mechanically dissociated cells were plated in a flask at a density of 107per 300 cm2 and maintained at 30°C in a 10% CO2/90% air atmosphere. After 10-14 days in culture, floating and weakly attached cells on the mixed primary cultured cell layer were isolated by gentle shaking of the flask for 3-5 min. The resulting cell suspension was transferred to plastic dishes and allowed to adhere at 37°C. To investigate the morphological change of microglia, the cells after 2 days of culture were incubated with biotinylated GSA-I-B4 (10ug/ml) at 4°C for overnight. To detect the phagocytic, isolated microglia were incubated with opsonized zymosan (20mgl/ml) for Ih at 37°C and with Giemsa's staining solution for 30 min at room temperature. The results were about 90% of attached cells were positive for OX6. Morphologically, most of the isolated microglial cells had amoeboid and rod-shaped cell bodies with no or a few thick processes. Most of these cells became amoeboid-like cells and showed a number of vacuoles in the cytosol when cultured in the presence of IFN-y+ LPS - treated cells exhibited the intense phagocytic activity against zymosan particles.