SELECTIVE STAINING OF ASTROGLIA AND MICROGLIA BY SEQUENTIAL COMBINATION OF HISTOCHEMICAL AND IMMUNOCYTOCHEMICAL MARKERS.

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Simultaneous demonstration of astrocytes and microglial cells was achieved by the combined use of glial specific histochemical and immunocytochemical markers. The study was made on adult Wistar rats fixed by intracardiac perfusion with 4% paraformaldehyde and 5% sucrose in 0.1 M cacodylate buffer (pH 7.4). Vibratome (30 µm) including including the hippocampus were obtained and processed for histochemical demonstration of 1) nucleoside-diphosphatase (NDPase) or 2) purine-nucleoside phosphorylase (PNPase). In addition, the sections were processed a) for immunocytochemical detection of the astroglial marker glial fibrillary acidic protein (GFAP) or b) for OX-42 immunoreactivity.

Sections sequentially processed for NDPase and OX-42 immunoreactivity showed a population of double labelled cells with a morphology like microglial cells. Brain sections reacted for both NDPase and GFAP contained two differently stained populations of cells: NDPase positive cells identified as microglial cells and GFAP positive astroglial cells. No double labelled, NDPase-GFAP positive cells, were observed. Sections reacted sequentially for PNPase and OX-42 showed that most OX-42 positive cells (microglia) were PNPase positive, although some OX-42 positive, PNPase negative cells were also observed. Many PNPase positive, OX-42 negative cells, were observed and morphologically identified as astrocytes, and brain sections reacted for both PNPase and GFAP showed that GFAP positive astrocytes always were positive for PNPase. In these sections, cells negative to GFAP, but positive to PNPase had the morphology of microglial cells.

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