Heat Shock Protein 60 in Corpora Amylacea

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Heat shock protein 60 representation in the corpora amylacea of the brain was investigated in five different neurological diseases. In the cases with cerebral infarct, amyotrophic lateral sclerosis, multiple sclerosis, acute disseminated encephalomyelitis and primary tumors of the nervous system the corpora amylacea showed similar appearance with strong HSP-60 positivity in all investigated disorders at the predilection sites. In the inflammatory diseases, besides corpora amylacea, several cellular elements exhibited HSP-60 immunostaining too. In these cases, the widespread HSP-60 immunoreactivity associated with relative moderate corpora amylacea production as compared to other diseases. From this contradiction we concluded the corpora amylacea participate in the cellular stress reaction but stress protein synthesis certainly is not the primary event in corpora amylacea formation. In the development of the corpora amylacea the incipient process is most probably degenerative in nature, which later on is accompanied by stress protein synthesis and slow growing of these round structures designated for a protective role in the brain. However, the role of the stress protein synthesis in the corpora amylacea formation and growth was not unequivocally answered in this study. It is necessary to perform further comparative investigations of the stress protein representation and corpora amylacea formation in different diseases which may help in discovering useful pathogenetic data and the biological role of this degenerative structure. (Pathology Oncology Research Vol 7, No 2, 140–144, 2001)

Introduction

The cells of the nervous system produce peculiar materials with special morphology in the elderly, and especially in some degenerative disorders. These structures are known as Bunina- Hirano- Lewy-bodies, Rosenthal fibres, Corpora amylacea (CA), Alzheimer’ neurofibrillary tangles, etc. The source and the role of these structures has not yet been quite clearly explained. Since these inclusions or degenerative products usually develop in the ageing brain and in some kinds of chronic degenerative brain diseases it is plausible to assume that repetitive, or chronic local cellular stress reactions might participate in their origin and development.

Heat shock proteins (HSP) are important members of the substances synthesised in the cells under stress conditions. These proteins are ubiquitous and, they are also present in the cells of the nervous system under normal conditions. Several reports have accumulated during the last few years about their central nervous system representation and speculation arose about their significance. Numerous publications analysed the relationship between the stress proteins and the degenerative products mentioned above. We also investigated the heat shock protein positivity in different neurological diseases and observed that the CA were strongly labeled by HSP-60 antibody. We thought it might be worthy to demonstrate these data and that we may be able to draw some conclusions from the findings.

Materials and Methods

The cases to be studied were selected from the brain autopsies of the Laboratories of Neuropathology of the Departments of Neurology, University Hospital Pécs, further from the University Hospital of Tatabánya and Miskolc. The brains were removed in 48 hours after death. Formalin fixation and paraffin embedding was established after the macroscopical neuropathological investigation in
every cases. The histological diagnosis was based on routine neuropathological staining and different impregnation methods including haematoxylin-eosin, PAS, Klüver-Barrera, Wölcke stainings and Gallyas-astrocyte, Hortega-microglia, Bielschowsky-axon impregnations as well as GFAP immunocytochemistry. The HSP-60 immunohistochemical reactions were performed in cases of the following disorders: 1) Cerebral infarct. 2) Acute disseminated encephalomyelitis. 3) Multiple sclerosis. 4) Glioblastoma multiforme. 5) Amyotrophic lateral sclerosis. Five cases were selected from every group for examination and randomly assorted.

The HSP-60 immunohistochemical reactions were performed by peroxidase method. The 5 µm sections were incubated in bovine serum albumin then 3% hydrogen peroxide was applied. HSP-60 monoclonal antibody (Sigma H 4149) was administered as primary antibody in 1:250 dilution for 2 hours, only higher Butter solution was used for the negative control sections. After the primary antibody, biotinylated secondary antibody was added which was followed by the ExtrAvidin-Peroxidase reaction. The reaction was visualised by addition of an AEC substrate mixture. The monoclonal antibody used in our studies reacts with the 383-447 amino acids of the human HSP 60 but it does not exhibit cross reaction with the stress protein of bacterial origin. The sections from different neurological diseases were investigated for HSP-60 immunoreactivity and the HSP-60 representation was analysed as a possible marker of degeneration and CA formation.

**Results**

The results demonstrated unequivocal HSP-60 positivity of the CA in all samples (Figure 1). There were no significant differences found between the intensity of the reactions. While there was a pronounced HSP 60 positivity in the CA the immunohistochemical reaction was relatively weak in other structures of the brains, except the cases of inflammatory diseases. It was possible to detect punctuate HSP-60 representation in the astrocytes, and cytosolic HSP-60 positivity in the neurons, oligodendrocytes and inflammatory cells (lymphocytes) in the cases of multiple sclerosis and disseminated encephalomyelitis.