Mineralocorticoid receptor in the central nervous system

Mitsuhiro Kawata, Noriyuki Morita, Keiko Yokoyama and Masafumi Morimoto

Department of Anatomy and Neurobiology Kyoto Prefectural University of Medicine

Summary

It was shown that there are two types of adrenal steroid receptors in the mammalian central nervous system(CNS): mineralocorticoid receptor and glucocorticoid receptor. We tried to produce both antibodies and examine the distribution of immunoreactive cells in the CNS. First the rat liver glucocorticoid receptor cDNA fragment which encodes 173 amino acids in the transcription modulation domain was inserted to the pGEX vector to express a rat glucocorticoid receptor-GST fusion protein in E.coli. The fusion protein was induced by IPTG specifically. After the purification of the fusion protein, the receptor protein domain was cleaved out. By immunizing with the purified protein, a polyclonal antibody was raised in a rabbit. In immunoblot analysis with the use of rat liver and brain homogenates, a single band of Mr 97kDa, the size deduced from the rat glucocorticoid receptor was detected by this antibody. In the hybridization study, fluorescein-labeled RNA probes, complementary to about 500 base pairs coding the 5' untranslated region and a part of transcription modulation domain in the rat glucocorticoid receptor cDNA were used. The expression of glucocorticoid receptor mRNA was detected in the cytoplasm and immunoreactivity was predominantly in the nucleus. The distributional pattern of mRNA in many regions of the rat brain was correlated with the distribution of immunoreactivity, but in the hippocampus and supraoptic nucleus of the hypothalamus different localization was found. Rat mineralocorticoid receptor cDNA fragment was also inserted to the pGEX vector. The fusion protein was induced by IPTG. We are now underway to produce specific antibody for mineralocorticoid receptor.