effects of icv administration of these peptides on C on A response were examined. Icv injection of urocortin or leptin produced a marked decrease in C on A response. The suppressive effect of both urocortin and leptin was abolished by pretreatment with chlorisondamine or propranolol, but not by adrenalectomy. The suppressive effect of leptin was completely abolished either by surgical severing of the splenic nerves or by icv injection of an antibody against CRH, but only partially by an anti-urocortin antibody. Additionally, the suppressive effect of urocortin was prevented by the anti-CRH antibody, while that of CRH was not prevented by the anti-urocortin antibody. These results suggest that urocortin, CRH and leptin are important neuropeptides involved in the hypothalamic control of peripheral immune functions such as stress-induced immunosuppression. The suppressive effect of leptin is mediated through the activation of the CRH (urocortin)-sympathetic nervous system.

3) To elucidate possible involvement of leptin, CRH and urocortin in the stress-induced immunosuppression, the effects of respective antibodies on splenic lymphocyte proliferation after footshock stress were examined. Icv injection of an antibody against CRH completely abolished the stress-induced suppression of the C on A response. However, antibodies against leptin and urocortin showed no effect. Moreover, the serum leptin level was decreased by immobilization and footshock stress. These results suggest that the central CRH, but not urocortin and leptin, is a factor regulating the peripheral immune activity through sympathetic nerve activation under stress-conditions.

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SYMPATHETIC REGULATION OF HEPATIC INTERLEUKIN EXPRESSION DURING NON-INVASIVE STRESS

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IL-1 and IL-6 are the major cytokines produced in the liver, which are involved in some specific responses in this organ during the acute phase of inflammation. Recently, evidence has accumulated suggesting that the production of IL-1 and IL-6 is also increased during various non-invasive stress such as immobilization and exercise. In this thesis, to

elucidate the mechanisms of the stress-induced production of IL-1 and IL-6, I examined mRNA expression of these cytokines *in vivo* in the rats exposed to oscillation stress, and also *in vitro* using primary cultured hepatocytes and non-parenchymal liver cells. I especially focused on a possible regulatory role of catecholamines secreted from sympathetic nerves and the adrenal gland during stress.

I first examined mRNA expression of IL-1β and IL-6 in the liver after subjection to oscillation stress in rats. Thirty-minute subjection to oscillation stress increased IL-1β and IL-6 mRNA expression in the liver. Prior treatment of rats with gadolinium chloride, which eliminates macrophages, prevented the stress-induced IL-1β expression. Either adrenalectomy or treatment of guanethidine, a blocker of norepinephrine release in the sympathetic nerve endings, partially attenuated the stress-induced IL-1β mRNA response, but the combined treatment completely blocked it. Injection of β-adrenergic receptor antagonist (propranolol) also suppressed the stress-induced IL-1β response. Essentially the same results were obtained for IL-1β mRNA expression in the spleen. These results suggest that oscillation stress induces IL-1β mRNA expression in the liver and spleen, probably in Kupffer cells and splenic macrophages, and that stress-induced IL-1β expression is elicited by catecholamines released from sympathetic nerve terminals and the adrenal gland.

Previously, I demonstrated in mice that immobilization stress induced IL-6 mRNA expression in the liver in parallel with an elevation of plasma IL-6 level, and that IL-6 immunoreactivity was present more in hepatocytes than in non-parenchymal liver cells. Thus, to clarify the relationship between IL-1 and IL-6 in the liver cells during non-invasive stress, I next examined mRNA expression of IL-1β and IL-6 in vitro using primary cultured hepatocytes and non-parenchymal liver cells isolated from rats. When cultured *in vitro*, both types of cells expressed IL-6 mRNA. IL-6 mRNA expression in hepatocytes, but not in non-parenchymal liver cells, was increased when the cells were treated with NE. The stimulatory effect of NE was inhibited by combined use of α-and β-adrenergic receptor antagonists. IL-6 mRNA expression in hepatocytes was also increased by incubating with the culture medium of non-parenchymal liver cells treated with NE. The effect of the medium was blocked by an IL-1 receptor antagonist. Moreover, exogenous IL-1β stimulated IL-6 mRNA expression in hepatocytes. IL-1β was present in the medium of non-parenchymal liver cells and increased by NE-treatment. All these results suggest that NE released from sympathetic nerve terminals during stress directly increases IL-6 mRNA expression in hepatocytes and indirectly through IL-1β production from non-parenchymal liver cells.

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