Stopped-Flow Fluorescence Studies on Binding Kinetics of Neurotoxins with Acetylcholine Receptor.

TOSHIYA ENDO, MAMORU NAKANISHI, SHOEI FURUKAWA, FRANCIS J. JOUBERT, NOBUO TAMIA and KYozo HAYASHI*

Acetylcholine receptor from *Narke japonica* electroplax exhibits a fluorescence change upon binding of snake neurotoxins. This fluorescence change primarily arises from the conformational change of the acetylcholine receptor and reflects the binding process of the toxin with the receptor. The rate constants for the binding of the neurotoxins to the receptor show surprisingly a wide range of distribution. Examination of the relationship between the rate constants of fluorescence change of the short neurotoxins and their amino acid sequences, thermal stability, hydrogen-deuterium exchange behavior, overall net charge, etc. reveal the positive charges on the side chains and overall net charge of the neurotoxin govern the magnitude of the binding rate.

Catecholamines Induce an Increase in Nerve Growth Factor Content in the Medium of Mouse L-M Cells.

YOSHlKO FURUKAWA, SHOEI FURUKAWA, EIJIRO SATOYOSHI and KYozo HAYASHI*

L-M cells, a mouse fibroblast cell line, synthesized and secreted a nerve growth factor (NGF) which was identical to those properties of β-NGF of the mouse submaxillary gland. Treatment of the cells with either norepinephrine or epinephrine in the range of 0.05-0.2 mM for 24h resulted in a 3-20-fold increase in NGF content in the medium of the L-M cells. The rate of incorporation of [3H] leucine into trichloroacetic acid-insoluble materials was essentially unchanged during the treatment. These results suggested that norepinephrine and epinephrine stimulated the *de novo* synthesis and secretion of NGF protein. Evidence is also presented to indicate that the effects of the drugs are not mediated by adrenergic receptors.

An Enzyme-Linked Immunosorbent Assay for Antibodies against Saline-Soluble Muscle Components in Myasthenia Gravis.

SAEKO AKAZAWA, ISAO KAMO, YOSHlKO FURUKAWA, EIJIRO SATOYOSHI and KYozo HAYASHI*

An enzyme-linked immunosorbent assay (ELISA) system has been developed for measuring antibodies against rat skeletal muscle components solubilized with phosphate-buffered saline. With this assay, 53.8% of sera from patients with myasthenia gravis (MG) was positive. No sera from patients with neurological disorders other than MG gave positive values. No correlation between titers of antibody against the muscle components and those of anti-acetylcholine receptor antibody (r=0.01) was found. These results indicate that our ELISA system is useful for diagnosing myasthenia gravis.