Department of Basic Medical Sciences

Division of Neuronal Network

神経ネットワーク分野

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Our major research interest is the molecular mechanisms of higher brain functions in mammals such as emotion, and learning and memory. We are especially focusing on the roles of functional molecules localized in synapses, for instance, neurotransmitter receptors, signal transduction molecules and adhesion molecules, in neuronal information processing. We are examining receptor functions, synaptic transmission and plasticity, and their roles in the whole animal with electrophysiological, biochemical, molecular genetic and behavioral approaches.

1. LMTK3 deficiency causes pronounced locomotor hyperactivity and impairs endocytic trafficking

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LMTK3 belongs to the LMTK family of protein kinases that are predominantly expressed in the brain. Physiological functions of LMTK3 and other members of the LMTK family in the central nervous system remain unknown. In this study, we performed a battery of behavioral analyses using *Lmtk3*$^{-/-}$ mice and showed that these mice exhibited abnormal behaviors, including pronounced locomotor hyperactivity, reduced anxiety behavior, and decreased depression-like behavior. Concurrently, the dopamine metabolite levels and dopamine turnover rate were increased in the striata of *Lmtk3*$^{-/-}$ mice compared with wild-type controls. In addition, using cultured primary neurons from *Lmtk3*$^{-/-}$ mice, we found that LMTK3 was involved in the endocytic trafficking of *N*-methyl-D-aspartate receptors, a type of ionotropic glutamate receptor. Altered membrane traffic of the receptor in *Lmtk3*$^{-/-}$ neurons may underlie behavioral abnormalities in the mutant animals. Taken together, our data suggest that LMTK3 plays an important role in regulating locomotor behavior in mice.

2. The glutamate receptor GluN2 subunit regulates synaptic trafficking of AMPA receptors in the neonatal mouse brain

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The N-methyl-D-aspartate receptor (NMDAR) plays various physiological and pathological roles in neural development, synaptic plasticity and neuronal cell death. It is composed of two GluN1 and two GluN2 subunits, and in the neonatal hippocampus, most synaptic NMDARs are GluN2B-containing receptors, which are gradually replaced with GluN2A-containing receptors during development. Here, we examined whether GluN2A could be substituted for GluN2B in neural development and functions by analyzing knock-in (KI) mice in which GluN2B is replaced with GluN2A. The KI mutation was neonatally lethal, although GluN2A-containing receptors were transported to the post-synaptic membrane even without GluN2B and functional at synapses of acute hippocampal slices of postnatal day 0 (P0), indicating that GluN2A-containing NMDARs could not be substituted for GluN2B-containing NMDARs. Importantly, the synaptic α-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid receptor (AMPAR) subunit GluA1 was increased, and the transmembrane AMPAR regulatory protein (TARP) and synaptic Ras-GTPase activating protein (SynGAP), which are both involved in AMPAR synaptic trafficking, were increased and decreased, respectively, in KI mice, whereas calcium/calmodulin-dependent protein kinase IIα (CaMKIIα) was not involved in the increase of GluA1. Although the regulation of AMPARs by GluN2B has been reported in cultured neurons, we showed here that AMPAR-mediated synaptic responses were increased in acute KI slices, suggesting differential roles of GluN2A and GluN2B in AMPAR expression and trafficking *in vivo*. Taken together, our results suggest that GluN2B is essential for the survival of animals and that the GluN2B-GluN2A switching plays a critical role in synaptic integration of AMPARs through regulation of GluA1 in the whole animal.

3. The active zone protein CAST regulates synaptic vesicle recycling and quantal size

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Synaptic efficacy is determined by various factors, including the quantal size, which is dependent on the amount of neurotransmitters in synaptic vesicles at the presynaptic terminal. It is essential for stable synaptic transmission that the quantal size is kept within a constant range and that synaptic efficacy during and after repetitive synaptic activation is maintained by replenishing release sites with synaptic vesicles. However, the mechanisms for these fundamental properties have still been undetermined. We found that the active zone protein CAST played pivotal roles in both presynaptic regulation of quantal size and recycling of endocytosed synaptic vesicles. In CAST knockout mice, miniature excitatory synaptic responses were increased in size and synaptic depression after prolonged synaptic activation was larger, which was attributable to selective impairment of synaptic vesicle trafficking via the endosome in the presynaptic terminal mediated by Rab6. Therefore, CAST serves as a key molecule that regulates dynamics and neurotransmitter contents of synaptic vesicles in the excitatory presynaptic terminal.

Publications

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