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. We are also trying to elucidate fundamental aspects of psychiatrical and neurological disorders using model animals.

1. SNAP-25 phosphorylation at Ser187 regulates synaptic facilitation and short-term plasticity in an age-dependent manner

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Neurotransmitter release is mediated by the soluble NSF attachment protein receptor (SNARE) complex, but the role of its phosphorylation has scarcely been elucidated. Although protein kinase C (PKC) activators are known to facilitate synaptic transmission, there has been a heated debate on whether PKC mediates facilitation of neurotransmitter release through phosphorylation. One of the SNARE proteins, SNAP-25, is phosphorylated at the residue serine-187 by PKC, but its physiological significance has been unclear. To examine these issues, we analyzed mutant mice lacking the phosphorylation of SNAP-25 serine-187 and found that they exhibited reduced neurotransmitter release probability and enhanced presynaptic short-term plasticity, suggesting that not only the release process, but also the dynamics of synaptic vesicles was regulated by the phosphorylation. Furthermore, it has been known that the release probability changes with development, but the precise mechanism has been unclear, and we found that developmental changes in release probability of neurotransmitters were regulated by the phosphorylation. These results indicate that SNAP-25 phosphorylation developmentally facilitates neurotransmitter release but strongly inhibits presynaptic short-term plasticity via modification of the dynamics of synaptic vesicles in presynaptic terminals.

2. CDKL5 controls postsynaptic localization of GluN2B-containing NMDA receptors in the hippocampus and regulates seizure susceptibility

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Mutations in the *Cyclin-dependent kinase-like 5 (CDKL5)* gene cause severe neurodevelopmental disorders accompanied by intractable epilepsies, i.e. West syndrome or atypical Rett syndrome. Here, we report generation of the *Cdkl5* knockout mouse and show that CDKL5 controls postsynaptic localization of GluN2B-containing N-methyl-D-aspartate (NMDA) receptors in the hippocampus and regulates seizure susceptibility. *Cdkl5* -/Y mice showed normal sensitivity to kainic acid; however, they displayed significant hyperexcitability to NMDA. In concordance with this result, electrophysiological analysis in the hippocampal CA1 region disclosed an increased ratio of NMDA/α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptor-mediated excitatory postsynaptic currents (EPSCs) and a significantly larger decay time constant of NMDA receptor-mediated EPSCs (NMDA-EPSCs) as well as a stronger inhibition of the NMDA-EPSCs by the GluN2B-selective antagonist ifenprodil in *Cdkl5* -/Y mice. Subcellular fractionation of the hippocampus from *Cdkl5* -/Y mice revealed a significant increase of GluN2B and SAP102 in the PSD (postsynaptic density)-1T fraction, without changes in the S1 (post-nuclear) fraction or mRNA transcripts, indicating an intracellular distribution shift of these proteins to the PSD. Immunoelectron microscopic analysis of the hippocampal CA1 region further confirmed postsynaptic overaccumulation of GluN2B and SAP102 in *Cdkl5* -/Y mice. Furthermore, ifenprodil abrogated the NMDA-induced hyperexcitability in *Cdkl5* -/Y mice, suggesting that upregulation of GluN2B accounts for the enhanced seizure susceptibility. These data indicate that CDKL5 plays an important role in controlling postsynaptic localization of the GluN2B-SAP102 complex in the hippocampus and thereby regulates seizure susceptibility, and that aberrant NMDA receptor-mediated synaptic transmission underlies the pathological mechanisms of the CDKL5 loss-of-function.

Publications

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