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. Functional coupling of the metabotropic glutamate receptor, inositol triphosphate receptor and L-type Ca^{2+} channel in mouse CA1 pyramidal cells

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Activity-dependent regulation of calcium dynamics in neuronal cells can play significant roles in the modulation of many cellular processes such as intracellular signaling, neuronal activity and synaptic plasticity. Among many calcium influx pathways into neurons, the voltage-dependent calcium channel (VDCC) is the major source of calcium influx, but its modulation by synaptic activity has still been under debate. While the metabotropic glutamate receptor (mGluR) is supposed to modulate L-type VDCCs (L-VDCCs), its reported actions include both facilitation and suppression, probably reflecting the uncertainty of both the molecular targets of the mGluR agonists and the source of the recorded calcium signal in previous reports. In this study, using subtype-specific knockout mice, we have shown that mGluR5 induces facilitation of the depolarization-evoked calcium current. This facilitation was not accompanied by the change in single-channel properties of the VDCC itself; instead, it required the activation of calcium-induced calcium release (CICR) that was triggered by VDCC opening, suggesting that the opening of CICR-coupled cation channels was essential for the facilitation. This facilitation was blocked or reduced by the inhibitors of both L-VDCCs and inositol triphosphate receptors (IP_3Rs). Furthermore, L-VDCCs and mGluR5 were shown to form a complex by coimmunoprecipitation, suggesting that the specific functional coupling between mGluR5, IP_3Rs and L-VDCCs played a pivotal role in the calcium-current facilitation. Finally, we showed that mGluR5 enhanced VDCC-dependent long-term potentiation of synaptic transmission. Our study has identified a novel mechanism of the interaction between the mGluR and calcium signaling, and suggested contribution of mGluR5 to synaptic plasticity.

2. Dysfunction of the RAR/RXR signaling pathway in the forebrain impairs hippocampal memory and synaptic plasticity

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Retinoid signaling pathways mediated by retinoic acid receptor (RAR)/retinoid X receptor (RXR)-mediated transcription play critical roles in hippocampal synaptic plasticity. Furthermore, recent studies have shown that treatment with retinoic acid alleviates age-related deficits in hippocampal long-term potentiation (LTP) and memory performance and, furthermore, memory deficits in a transgenic mouse model of Alzheimer's disease. However, the roles of the RAR/RXR signaling pathway in learning and memory at the behavioral level have still not been well characterized in the adult brain. We here show essential roles for RAR/RXR in hippocampus-dependent learning and memory. In the current study, we generated transgenic mice in which the expression of dominant-negative RAR (dnRAR) could be induced in the mature brain using a tetracycline-dependent transcription factor and examined the effects of RAR/RXR loss. The expression of dnRAR in the forebrain down-regulated the expression of RARβ, a target gene of RAR/RXR, indicating that dnRAR mice exhibit dysfunction of the RAR/RXR signaling pathway. Similar with previous findings, dnRAR mice displayed impaired LTP and AMPA receptor-mediated synaptic transmission in the hippocampus. More importantly, these mutant mice displayed impaired hippocampus-dependent social recognition and spatial memory. However, these deficits of LTP and memory performance were rescued by stronger conditioning stimulation and spaced training, respectively. Finally, we found that pharmacological blockade of RARα in the hippocampus impairs social recognition memory. From these observations, we concluded that the RAR/RXR signaling pathway greatly contributes to learning and memory, and LTP in the hippocampus in the adult brain.

3. NMDAR2B tyrosine phosphorylation is involved in thermal nociception

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Previous studies found that the NMDA receptor-mediated signaling regulates thermal nociception, though the underlying molecular mechanism remains unclear. The GluN2B subunit of the NMDA receptor is tyrosine-phosphorylated, Tyr-1472 being the major phosphorylation site. In this study, we have found that homozygous knock-in mice that express a Tyr-1472-Phe mutant of GluN2B display defects in the nociceptive response in the hot plate test. Expression of the neurotensin receptor subtype 2 (NTSR2), which is relevant to the regulation of thermal nociception, is decreased in the amygdala of GluN2B Tyr-1472-Phe knock-in mice. In addition, NTSR2-mediated c-fos induction is impaired in the amygdala of these mice. These data suggest that Tyr-1472 phosphorylation on GluN2B is involved in thermal nociception through regulating the NTSR2 mRNA expression in the amygdala.

Publications

Nomoto, M., Takeda, Y., Uchida, S., Mitsuda, K., Enomoto, H., Saito, K., Choi, T., Watabe, A.M., Kobayashi, S., Masushige, S., Manabe, T. and Kida, S. Dysfunction of the RAR/RXR signaling pathway in the forebrain impairs hippocampal memory and synaptic plasticity. *Molecular Brain* 5: 8, 2012.

Delawary, M., Tezuka, T., Kiyama, Y., Yokoyama, K., Wada, E., Wada, K., Manabe, T., Yamamoto, T. and Nakazawa, T. NMDAR2B tyrosine phosphorylation is involved in thermal nociception. *Neurosci. Lett.* 516: 270-273, 2012.

Kato, H.K., Kassai, H., Watabe, A.M., Aiba, A. and Manabe, T. Functional coupling of the metabotropic glutamate receptor, $InsP_3$ receptor and L-type Ca^{2+} channel in mouse CA1 pyramidal cells. *J. Physiol. (Lond.)* 590: 3019-3034, 2012.

Manabe, T. Analysis of synaptic plasticity with the slice patch-clamp recording technique. In *Patch Clamp Techniques: From Begging to Advanced Protocols*. Edited by Okada, Y. (Springer, Tokyo) pp 147-157, 2012.

渡部文子，真鍋俊也．神経② 神経可塑性．イラストで徹底理解するシグナル伝達キーワード事典．山本雅，仙波憲太郎，山梨裕司編集．羊土社．pp 266–273，2012.