animals isolated from U.K. were destroyed by cold stimuli, 2°C for 48 hours. By contrast, most of wild-type animals can survive for 48 hours at 2°C, after cultivation at 15 degree. Detailed temperature-changes experiments revealed that the cold-resistance is established by temperature memory at adult stage. We measured cold-resistance in various mutant animals defective in sensoryneurons. Abnormal enhancement of cold-resistance is found in the mutant animals defective in TAX-4 cGMP-gated channel, which is expressed in 10 pairs of sensoryneurons including thermo-sensoryneuron AFD and chemo-sensoryneurons. Developmental defect of AFD that is caused by mutation in OTX transcriptional factor did not affect cold-resistance. These results suggest that cold-resistance is controlled by sensory neurons excepting AFD, and that sensoryneurons transmit inhibitory signaling for cold resistance. To determine molecular pathway for cold resistance, we referred previous DNA microarray analysis (Sugi et al., Nature neurosci., 2011). So far, at least 10 genes such as endonuclease, tubulin kinase and phosphatase PP1 are involved in cold-resistance.

3PT202 Inhibitory effect of glutamate-stimulated astrocytes on action potentials evoked by bradykinin in cultured dorsal root ganglion neurons

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The patch-clamp and Ca^{2+} -imaging techniques have revealed that astrocytes have dynamic properties including ion channel activity, neurotransmitter release, such as adenosine triphosphate (ATP) and glutamate. Our previous study demonstrated that free nerve endings of cultured neuron from dorsal root ganglion (DRG) are activated by bradykinin (BK). We study whether ATP and glutamate can modulate the BK-response of the neuron cultured with the astrocyte in the mouse DRG, and enunciate the role of the astrocyte in the nociceptive signal transmission using patch-clamp technique. The astrocytes were identified using fluorescent antibody, anti-GFAP. The membrane potential of the astrocyte was about -39 mV. The application of glutamic acid (GA) to the bath evoked the opening of two kinds of Cl^- channel of the astrocyte membrane with unit conductance of about 380 pS and 35 pS in the cell-attached mode, respectively. ATP application evoked the opening of two kinds of K^+ channel of the astrocyte with unit conductance of about 60 pS and 29 pS, respectively. Application of BK to the neuron evoked an action potential (spike). The BK-application of concomitantly with ATP increased the frequency of BK-evoked spike of the neuron. Stimulation of BK with GA inhibited the BK-evoked spike. The application of furosemide, a potent cotransporter ($\text{Na}^+ - \text{K}^+ - \text{Cl}^-$) inhibitor, prior to the stimulation of BK with GA blocked the inhibition of the spike. It is thought that the inhibition of the spike is related to Cl^- movement from the astrocyte.

3PT203 マウス脳ホモジネートにおける一酸化窒素 (NO) の測定 Nitric oxide measurement in mice brain homogenate

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It is known that nitric oxide (NO) plays important roles in a number of physiological processes over various animal species of vertebrate and invertebrate, such as neurotransmission, vasodilation and immune response. Especially in recent years, relationship between nitric oxide (NO) and aging has been received a lot of attention. However down-regulation of NO signaling during aging process is strongly suggested, NO dynamics in brain have not been studied in detail. Therefore, in this study, NO production in mice brain homogenate was measured using NO-specific electrode. Brain homogenates (cerebral cortex, cerebellum and hippocampus) were prepared from male wild-type mice (8 and 120 weeks). After oxidation current of the NO electrode stabilized in the saline, each brain homogenate was added for estimation of basal NO production. To confirm specificity of the electrode, brain homogenate was stimulated with calcium ion (Ca^{2+}). When brain homogenate was added, NO concentration increased and was stabilized at constant level. Ca^{2+} accelerated the basal NO production in a concentration-dependent manner. However concentration of basal NO production did not so different in region and age, Ca^{2+} -induced NO production in aged mice significantly decreased in every region compared with young mice. From these results, NO production from mice brain homogenate was successfully measured with the NO-specific electrode. We also examined that aging induced down-regulation of Ca^{2+} /NO signaling in mice brain.

3PT204 線虫 AIY 介在神経細胞での局在した神経活動：蛍光イメージングによる研究

Localized neural activities in AIY interneuron of *Caenorhabditis elegans*: fluorescent imaging study

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The neural activities of subcellular regions are important for understanding the relationship between the cellular morphology and the function in single neurons. From this view, AIY interneuron of *Caenorhabditis elegans* has a very unique feature of Ca^{2+} dynamics; Ca^{2+} change is observed only on the specific region of neurites, but not on soma during odor and temperature stimulation. To investigate this dynamics in subcellular regions of AIY in detail, we visualized the membrane potential and showed that its dynamics is different from the Ca^{2+} one.

For visualization of neural activities on specific neurons in *C. elegans*, we have developed a specific micro-fluidic device to fix the worm. The worm was applied to 10^{-4} dilution of isoamyl alcohol as the controlled odor stimulation during 50 sec. The genetic Ca^{2+} indicator (YC3.60) expressed in AIY has revealed the subcellular neural activities to the external environmental change. To compare membrane potential change with the dynamics, we also used voltage-sensitive fluorescent protein (VSFP) 2.42 (Akemann et al., 2010). VSFP2.42 revealed that the odor stimulation depolarized membrane potential both on the neurite and soma. In addition, the membrane potential showed oscillation with attenuation of the amplitude during odorant stimulation. These results suggest that AIY has the diversity of biophysical characteristics in the single cell and it is necessary for understanding its function.

3PT205 コルチコステロンは、シナプス局在の GR-kinase 系を介して、海馬神経シナプスを増やす

Corticosterone induced GR/kinase network-driven rapid spinogenesis in rat hippocampus

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Modulation of hippocampal synaptic plasticity by corticosterone (CORT) has been attracting much attention, due to its importance in stress responses. Many studies have investigated the chronic effect of CORT, but rapid effect of CORT in hippocampus is unclear. Here, we investigated effect of CORT, a major glucocorticoid in rat, on density and morphology of dendritic spine which is essential for memory storage processes, in adult male rat hippocampus. The application of 1 μM CORT induced a rapid increase in the total density of spines of CA1 pyramidal neurons within 1 h from 0.98 spines/ μm to 1.24 spines/ μm . CORT increased the density of large-head (0.5-1.0 μm) and middle-head spines (0.4-0.5 μm), but not small-head (0.2-0.4 μm) spines. Co-presence of RU486, an antagonist of glucocorticoid receptor (GR), completely abolished the effect of CORT. Blocking ionotropic glutamate receptors with MK-801 inhibited the CORT effect. This result suggests that Ca^{2+} homeostasis via NMDA receptors is necessary to CORT-induced spinogenesis. Blocking various kinases shows that CORT enhances spinogenesis via MAPK, PKA, PKC, PI3K and calcineurin signaling pathways. In particular, the contribution of Erk MAPK and PKA is essential. Inhibition of protein synthesis reduced the CORT-induced spinogenesis but inhibition of transcription. These results indicate that CORT drives the signaling pathway including synaptic GR and multiple kinase pathways in hippocampal CA1 neurons. (Komatsuzaki et al., 2012 PLoS-ONE)

3PT206 $\text{IP}_3/\text{Ca}^{2+}$ シグナリングによる GABA 作動性シナプスの制御 Regulation of GABAergic synapses by $\text{IP}_3/\text{Ca}^{2+}$ signaling

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The number of GABA-A receptors (GABA_AR) clustering at the inhibitory synapse is a crucial determinant of GABAergic synaptic transmission efficacy. Ca^{2+} influx in response to excitatory neuronal activity results in the dispersal of synaptic GABA_AR clusters. In addition to Ca^{2+} influx, Ca^{2+} release from the intracellular Ca^{2+} store also plays various roles in neurons. However, the impact of Ca^{2+} release on the inhibitory synapse remains to be elucidated. Here, we