ELECTROPHYSIOLOGICAL ANALYSIS OF HONEY BEE AUDITORY MECHANISM

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Honey bees develop sound communication representing the waggle dance. The various types of sound generated has been analyzed, however the question "How do honey bees hear the sound?" has not been addressed so far. We then conducted the electrophysiological analysis to identify honey bee auditory organs, and to record the sound evoked potential (SEP) of auditory nerves in response to various types of sound. Furthermore, we also measured the velocity and displacement of antennal vibration generated by sound. We did the same analysis on Bumble bees that do not exhibit dance communication. We have found that honey bee auditory organs locate at the Jonston's organ of antennae, and the antennal vibration and SEP is maximum around 250 Hz sound. The similar results are obtained with Bumble bees, suggesting that both bees have the auditory nerves with similar properties.

THE ANALYSIS OF MOTOR SYSTEM OF THE ASCIDIAN LARVA

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The ascidian larva is an excellent model for studies of the functional organization of vertebrate neural circuits due to its remarkably simple central nervous system (CNS) with about 100 neurons. Cholinergic and GABAergic neurons are critical neurons for locomotion of many animals. We successfully visualized the cholinergic and GABAergic neurons in the larval CNS with antibodies against vesicular acetylcholine (Ci-AchTP) and GABA transporter (Ci-vGAT), respectively. Ci-vAchTP positive neurons were located in the brain vesicle and the visceral ganglion. Five pairs of cholinergic neurons in the visceral ganglion extended their axons posteriorly and innervated the most anterior muscle cells in the tail. Ci-vGAT positive neurons were located in the brain vesicle, visceral ganglion, and anterior nerve cord. Two distinct pairs of Ci-vGAT positive neurons, bilaterally aligned along on the anterior nerve cord, extended their axons anteriorly and connected to axons of the contralateral cholinergic neurons. These results suggest that the neural network comprising cholinergic motor neurons and inhibitory interneurons is a CPG producing a rhythmic movement of the tail.

VISUALIZATION OF VISUAL PATHWAY IN ASCIDIAN LARVAE

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Ascidian larvae of Ciona intestinalis change their photic behavior during the course of development. Newly hatched larvae show no response to a light stimulus at any intensity. At 4 h after hatching, larvae were induced to start to swimming upon the cessation of illumination, and to stop swimming upon the onset of illumination. In order to follow the neural networks from the photoreceptor cells to muscle, the visual pathways of the larva were visualized by expressing a WGA transgene in the photoreceptor cells under the control of the arrestin gene (Ci-arr) promoter. In order to discriminate the first-order neurons from the higher-order neurons a transgene construct containing both sequences of *Ci-arr-WGA* and *Ci-arr-EGFP* was introduced into fertilized eggs by electroporation. Immunofluorescence labeling revealed that WGA and GFP transgene products were expressed in the photoreceptor cells and WGA transgene products were transported along their axons and transsynaptically to the second neurons in the brain vesicle and then to the fourth pair motor neurons of five pair neurons in the visceral ganglion.

FACILITATION BY P26OLF OF MEMBRANE AGGREGATION ACTIVITY OF ANNEXIN I AND II

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A Ca^{2+}-binding protein, p26olf, was found in bullfrog (Rana catesbiana) olfactory epithelium. To study the physiological role of p26olf, we tried to identify the target protein(s) of p26olf, and found that annexin I, II, and V are the candidates that specifically bind to p26olf at high Ca^{2+} concentrations. Because these annexins are known to activate membrane aggregation, and therefore possibly repair of membranes, we examined the effect of p26olf on the activity of each of these annexins. Using recombinant annexin I, II and V, we monitored liposome aggregation with light scattering measurements. The result showed that p26olf facilitated the activities of annexin I and II significantly but affected that of annexin V hardly. The effective Ca^{2+} concentration for annexin II was about ten times lower than that for annexin I. Because these annexins were found to co-localize with p26olf in the olfactory cilia, the result suggested that p26olf facilitates repair of olfactory cilia membranes by binding with annexin I or II depending on the cytoplasmic Ca^{2+} concentration.

SUCROSE INDUCES INCREASE OF NITRIC OXIDE CONCENTRATION IN SUGAR RECEPTOR CELL OF BLOWFLY, *PHORMIA REGINA*

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Our group has previously reported that exogenous nitric oxide (NO) can imitate sugar in generation of spikes from sugar receptor cells in *Phormia regina*, which suggest that NO is a signaling molecule in sugar receptor cells. However, it was unclear whether NO is produced in the sugar receptor cells or not. To further investigate NO producing cell, we tried to measure intracellular NO directly with NO-sensitive fluorescent dye, DAF-2 DA, in the cultured taste receptor cells from pupa of *P. regina* that specifically responded to their stimulants (sugar, salt and bitter). Pulse(10 sec) of sucrose induced $[NO]_i$ in some of the cells. In addition, we observed that pulses of sucrose induced $[NO]_i$ and $[Ca^{2+}]_i$ under $[Ca^{2+}]_o$ free condition, which suggests that increase of NO in the cell does not require extracellular Ca^{2+}.

NITRIC OXIDE-REGULATED TASTE TRANSDUCTION MECHANISM IN SUGAR RECEPTOR CELLS OF THE BLOWFLY, *PHORMIA REGINA*

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A taste organ in flies, intensively used as a model organism to study taste transduction, is a hair-shaped unit housing four functionally differentiated taste receptor cells. Three of them preferentially respond to sugars, salts and water, respectively. In the sugar receptor cells of the blowfly, *Phormia regina*, intracellular cGMP has been suggested as a second messenger. Cyclic GMP is produced by guanylyl cyclase (GC), the soluble form of which is activated by nitric oxide (NO). To explore a possible function of NO in the sugar receptor cells, we examined the effects of a scavenger of NO and inhibitors of NO synthase and soluble GC on the electrophysiological responses of the sugar receptor cells. Each of the scavenger and inhibitors, when introduced from the tip of a taste sensory unit into the taste receptor cells by the aid of sodium deoxycholate, suppressed the responses of the sugar receptor cells. These results suggest that NO is produced in the sugar receptor cells and activates soluble GC in the same cells, participating in the sugar taste transduction. This study was partially supported by PROBRAIN, Japan to T.N.

SPEED VERSUS ACCURACY TRADE-OFF OF PATH INTEGRATION IN A SUBSOCIAL BUG, *PARASTRACHIA JAPONENSIS*

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A subsocial bug, *Parastrachia japonensis* transports host drupes to its nymph-containing burrow. Since the bug left its burrow, it searched tortuously until it had encountered a drupe. After the bug found one, it took the shortest route back to its burrow. When the bug was displaced to another position, it walked straight toward the fictive burrow and then showed the searching behavior at the vicinity of the fictive burrow. Those results indicate that *P. japonensis* orients to its burrow using path integration.

During the busy provisioning season, the homing bug sometimes encounters a competitor of the same species which tries to steel its conveying drupe. In this occasion, the homing bug showed the escaping behavior to walk faster than the safety conditions. We measured the walking speed and the accuracy of the homing orientation in the field. When the bug encountered a competitor, its walking speed was increased, whereas the accuracies of its orientation; especially the distance accuracy, was decreased. Those results indicate there is a trade-off between speed and accuracy in the path integration, and the distance information is easily influenced by the escaping behavior.

RESPONSES OF DESCENDING NEURONS IN THE PRAYING MANTIS TO MOTION STIMULI

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The praying mantis is a predatory insect that detects its prey by vision. The mantis captures its prey with rapid grasping movements of forelegs. According to the spatial position of the prey, the direction of foreleg movements is controlled by motor neurons in the prothoracic ganglion. Spatial information of the prey position is translated by descending neurons that connects the brain and the prothoracic ganglion. To clarify the neural representation of spatial information in the central nervous system of the mantis,