

responses of descending neurons in the mantis to motion stimuli were recorded extracellularly from the ventral nerve cord between the brain and the prothoracic ganglion. All recorded neurons showed excitatory responses to light ON and OFF stimuli. Some neurons exhibited larger excitation to a motion of gratings in preferred direction than a motion in opposite direction. Other neurons showed largest responses to looming stimuli that simulated a black square approaching to the mantis.

SEXUAL DIFFERENCE OF COLOR SENSE IN A LYCAENID *NARATHURA JAPONICA*

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Color sense of a lycaenid butterfly *Narathura japonica* was investigated by the ERG method. The compound eye was illuminated with light from a xenon lamp through a liquid fiber and also through an Ganzfeld dome. The light ranged from wavelengths of 300 to 700nm at 20nm intervals. The butterflies showed a stronger response to light at wavelengths below 500nm, both by the liquid fiber and the Ganzfeld dome methods. The latter method revealed a tendency of sexual difference, with females showing a relatively stronger response to longer wavelengths.

ULTRASONIC COURTSHIP SIGNAL IN A MOTH, *OSTRINIA FURNACALIS* II. RESPONSES OF HEARING NEURONS

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Male moths of *Ostrinia furnacalis* (Lepidoptera: Crambidae) produce acoustic signal including ultrasound when they attempt to copulate with female moths. Detection of the male-produced ultrasound affects female's acceptance of the male. The signal from *O. furnacalis* involves ultrasound with a broad range frequency (25-100 kHz) and pulse structure. Two other *Ostrinia* species close to *O. furnacalis* emitted sound with pulse structures differing from *O. furnacalis*. Next we investigated responses of hearing neurons to 10-120 kHz acoustic stimuli in these three species. Neurons of the three species responded to a broad range of frequency. No sexual or species differences were found in the neural responses. Sound levels that induce threshold responses were low around 30-80 kHz, which corresponded to acoustic signals of the three species. Roles of male ultrasound signal and female sex pheromone in mate recognition and premating isolation are discussed.

FUNCTIONAL ANALYSIS OF GRAVITY-SENSE GENES IN THE ASCIDIAN LARVA

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Newly hatched larvae of the ascidian, *Ciona intestinalis*, swim upward toward the sea surface. Laser ablation experiments have been shown that otolith is a gravity-sense organ for the upward swimming behavior. Previously, we have shown that a tyrosinase gene (*Ci-tyr*) is essential for the upward swimming behavior. Prestin is a motor protein in the outer hair cells of the cochlea, and which senses membrane potential and drives rapid length changes in outer hair cells. We isolated a cDNA clone encoding prestin (*Ci-prestin*). *In situ* hybridization analysis showed that *Ci-prestin* was expressed near the otolith and notocord. Immunostaining with the antibody against *Ci-prestin* revealed that *Ci-prestin* was localized the cell surface of the statocyte of the otolith. These results suggested that *Ci-prestin* could be involved in a gravity-sense machinery for the upward swimming behavior. We are trying to knockdown the *Ci-prestin* to investigate the function of *Ci-prestin* in the upward swimming behavior.

ORIGIN OF THE VERTEBRATE PHOTORECEPTOR CELLS

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To understand the origin of vertebrate photoreceptor cell, we have investigated the photoreceptor cells of the larva of the ascidian *Ciona intestinalis*, whose tadpole-like larvae share a basic body plan with vertebrates. 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. We identified two distinct classes of photoreceptor cells in the ocellus by immunostaining with antibodies against *Ci-opsin1* and vesicular neurotransmitter transporters. The first population has twenty outer segments inside the pigment cup of the ocellus, which is located right side of the brain; these cells are glutamatergic (ON type photoreceptor cells). The second has ten outer segments around the pigment cup; most, if not all, of the photoreceptor cells are GABAergic (OFF type photoreceptor cells). Laser ablation of the first and second populations leads to loss of the swimming behavior photoreponse of larvae, suggesting that they are responsible for the larval photoreponse. Thus, the ascidian larva has vertebrate-like glutamatergic photoreceptors and unique non-glutamatergic photoreceptors.

HIGHER SYNTHETIC ACTIVITY OF cGMP IN CONES THAN IN RODS

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The recovery of a photoreponse is much faster in cones than in rods. Because the photoreponse recovery is due to restoration of the cytoplasmic cGMP concentration, one can expect that the synthetic activity of cGMP is higher in cones than in rods. Using purified membrane preparations, we measured the guanylate cyclase (GC) activity in rods and cones. The result showed that the GC activity expressed in units per visual pigment present is about 30-fold higher in cones, which agrees with the expectation mentioned above. In rods and cones, the cytoplasmic Ca^{2+} concentration decreases in the light, and this Ca^{2+} decrease is known to activate GC to accelerate the recovery of a photoreponse. GCAPs are the members of a family of Ca^{2+} -binding proteins and are known to increase the GC activity at low Ca^{2+} concentrations. We are currently trying to measure the effect of GCAP1-3 on the GC activity in purified rod and cone membranes to know the maximum GC activity in rods and cones. Studies on the localization of these GCAPs are now in progress.

QUANTITATIVE PCR-BASED SCREENING OF THE CO₂ SENSOR PROTEIN OF THE MOSQUITO (*ANOPHELES STEPHENSI*)

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In mosquitoes, only females are blood-sucking insects and possess the CO₂ sensing ability for seeking their victims. Electrophysiological studies have demonstrated that the CO₂ reception is achieved through the CO₂ sensilla on the maxillary palps of female mosquitoes. We considered the sensory appendage protein (SAP), the anion exchanger (AE) and a kind of gustatory receptor (GR) as candidates for the CO₂ sensor protein based on the results of studies in other insects and properties of CO₂ molecules. We tried to isolate these genes and related genes from *Anopheles stephensi*. We have obtained five SAPs out of seven, two AEs out of three and all three GRs. To evaluate their suitability for the CO₂ sensor protein, we conducted quantitative PCR-based screening between the maxillary palps of male and those of female. As a result, one of the AEs has been found to exhibit female-specific expression, suggesting that the AE is the strong candidate for the CO₂ sensor protein.

IDENTIFICATION AND CHARACTERIZATION OF VISUAL PIGMENTS OF THE JUMPING SPIDER

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Some arthropods have color vision as well as vertebrates. In various insects, color visual pigments (color opsins) have been identified and their distributions in compound eyes are well investigated. On the other hand, in jumping spiders, which also have advanced color vision, there is no study on their visual pigments so far. In this study, we tried to isolate opsins from two jumping spider species, *Hasarius adansonii* and *Plexippus paykulli* by RT-PCR method to elucidate the molecular basis of the color vision of jumping spiders. We successfully obtained multiple opsin cDNAs from both *H. adansonii* and *P. paykulli*. Phylogenetic analysis including these jumping spider opsins revealed that the common ancestor of living arthropods has at least two kinds of opsin which are probably different in spectrum sensitivity. Further investigation using spider opsins as a molecular marker, will discover a novel mechanism for color vision.

A RHODOPSIN EXHIBITING BINDING ABILITY TO AGONIST ALL-TRANS-RETINAL

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Rhodopsins are the members of the family of G protein-coupled receptors that have diverged from ligand-binding receptors into photoreceptive pigments. Vertebrate rhodopsins are able to bind the inverse agonist 11-cis-retinal but are unable to bind the agonist all-trans-retinal, indicating that vertebrate rhodopsin changed its binding ability during the course of molecular evolution. Here we show that unlike vertebrate rhodopsin, amphioxus rhodopsin is still able to bind agonist all-trans-retinal in addition to its role as a photoreceptive pigment. The opsin exhibits an approx. 50-fold higher affinity for 11-cis-retinal than for all-trans-retinal, and mutational analyses revealed that Trp265 in Helix