Histaminergic signaling underlies the early stages of color vision in the butterfly, *Papilio xuthus*

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Abstract

Spectrally opponent responses, i.e. wavelength-dependent response-polarity inversions, have been observed at the level of photoreceptors quite frequently in butterflies. The opponent photoreceptors show fast-onset hyperpolarization when stimulated with specific wavelengths. Similar fast, on-transient hyperpolarizing responses that are found in the insect second-order visual neurons (lamina monopolar cells, LMCs), are mediated by histamine-gated chloride channels. In the lamina of *Papilio*, the photoreceptors are not only presynaptic to LMCs, but also pre- and post-synaptic to other photoreceptors. Thus, I hypothesized that histaminergic sign-inverting synapses exist between different spectral receptors as a mechanism for the spectral opponency in butterfly photoreceptors. I conducted immunolocalization on two candidates of histamine-gated chloride channels, PxHCLA and PxHCLB, in the visual system of the butterfly *Papilio xuthus*. The anti-PxHCLA labeling was associated with the plasma membrane of non-photoreceptor neurons that are postsynaptic to photoreceptors, suggesting that PxHCLA is located in the LMCs. The anti-PxHCLB labeling overlapped with photoreceptor axons, indicating the PxHCLB is expressed at the inter-photoreceptor synapses. I also analyzed physiological properties of these channels by whole-cell patch-clamp using those expressed in cultured cells. The patch-clamp experiments confirmed that both PxHCLA and PxHCLB are indeed activated by histamine and permeate chloride ions. Using sharp microelectrode recording in discontinuous single-electrode current-clamp mode, I have shown that the responses of both LMCs and spectrally opponent photoreceptors could be reversed when the membrane potential was close to the equilibrium potential of chloride ions. To understand how the histaminergic synapses mediate signal processing in the early visual system, I recorded responses of LMCs and photoreceptors in the *Papilio* lamina. Regarding the spectral properties, LMCs seem to integrate chromatic signals from the photoreceptors in the same ommatidium. It suggests that PxHCLA channels most likely implement the primary visual processing from photoreceptors to secondary neurons. On the other hand, several classes of spectrally opponent photoreceptors were recorded. The recorded opponent responses are well explained by the ommatidial type-specific inter-photoreceptor (sign-inverting) synaptic connections via PxHCLB. The findings in this thesis bring a closer view on the underlying neural circuits of color processing at the initial visual stage in *Papilio*.