Figures and figure supplements

A common directional tuning mechanism of *Drosophila* motion-sensing neurons in the ON and in the OFF pathway

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Figure 1. Receptive fields and responses to apparent motion stimuli of T5-cells. (a) The Hassenstein-Reichardt model incorporates PD enhancement only, realized by a multiplication. Left: Responses to individual light pulses (‘Flicker’) delivered at the two different positions. The responses are shifted.

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Figure 1 continued

according to the stimulus sequence used for the subsequent apparent motion stimuli. Middle: Responses of the model to apparent motion stimuli in preferred (upper row) and null direction (lower row, thick line = measured response, thin line = linear expectation, that is sum of responses to the single light pulses). Right: Nonlinear response component defined as the difference between measured response and linear expectation. (b) same as (a) but for a Barlow-Levick model. This model incorporates ND-suppression only, realized by a division. (c) Average responses of five T5 cells to flicker stimuli (stimulus size: 5 degree) delivered to different optical columns. In order to average the responses of different flies, the response patterns were aligned and normalized with respect to the maximum response (central column) and shown in a false color code. (d) Same as (c) but for T4-cells. Data represent the mean of 10 T4-cells from 10 flies (from Haag et al., 2016). (e) Responses of T5 cells to stimuli presented to the central column and simultaneously to one of the columns of the two surrounding rings. As in c, the responses of different flies were aligned with respect to the column eliciting the maximum response when stimulated individually and normalized to it. Depending on the location, simultaneous stimulation of a second column led to either a suppressed (blue colors) or an enhanced (red colors) response compared to the exclusive stimulation of the central column. The suppression is stronger on the null side of the T5 cells. Data represent the mean of 6 T5 cells from 6 different flies. (f) Same as e) but for T4-cells. Data represent the mean of T4-cells from 8 flies (from Haag et al., 2016).

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Figure 1—figure supplement 1. (a) Time course of the response of T5 to flicker stimuli (duration 470 ms) delivered to ommatidia 1–7. Each flicker stimulus was repeated three times. (b) Same data as Figure 1c,d but plotted as a bar histogram representing the mean ± SEM. The receptive field of Figure 1—figure supplement 1 continued on next page.
Figure 1—figure supplement 1 continued

T5-cells turned out to be broader than the receptive field of T4-cells. T5 cells responded to stimulation of ommatidia 2–7 with an average of 83% response amplitude, T4 with an average of 63%. The distributions are significantly different (t-test p-value: 0.013). The responses to stimulation of ommatidia 8–19 elicited similar responses in both cell types (T5: 48%; T4: 44%; t-test p-value: 0.49).

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Responses of T4-cells and T5 cells to stimuli presented to the central column and simultaneously to one of the columns of the two surrounding rings. Same data as Figure 1e,f but plotted as a bar histogram representing the mean ± SEM. Responses of different flies were normalized to the column eliciting the maximum response (column 1 in scheme). Both cell types show significantly smaller responses in the dorsal region (columns 19, 8, 9) than in the ventral region (columns 13, 14, 15) of the receptive field (T4: dorsal 55%, ventral 86%, t-test p-value 0.00005; T5: dorsal 64%, ventral 118%, t-test p-value 0.0008).

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Figure 2. Apparent motion stimuli between adjacent cartridges. (a) Response of a single T5 cell recorded in a single sweep to two-step apparent motion stimuli. The schematic to the left shows the position of the two stimuli (blue and green shading). Left: Responses to individual light pulses

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('Flicker') delivered at the two different positions. The responses are shifted in time according to the stimulus sequence used for the subsequent apparent motion stimuli. Middle: Responses of T5 to apparent motion stimuli in preferred and null direction (thick line = measured response, thin line = linear expectation, that is sum of responses to the single light pulses). Right: Nonlinear response component defined as the difference between measured sequence response and linear expectation. The responses are the mean obtained from $n = 3$ stimulus repetitions. (b) Two-step apparent motion stimuli were shown at three different positions in the receptive field of T5 cells. The stimulus consisted of light off pulses positioned on one column for 472 ms, immediately followed by a light off pulse for 472 ms to the upper, neighboring cartridge. The same stimuli were repeated along the opposite direction. (c) Nonlinear response component, that is the difference between sequence response and the sum of the responses to the individual pulses, as a function of time for a stimulus size of 5 degree. Apparent motion stimuli delivered to the upper two cartridges resulted in a null direction suppression and no preferred direction enhancement. Apparent motion stimuli in the lower cartridges did not lead to a deviation from the linear expectation. For all three stimuli no preferred direction enhancement could be found. Data represent the mean ± SEM in 6 T5-cells measured in 6 different flies. (d) Same as d, but with a stimulus size of 8 degree. In contrast to the results for a smaller stimulus size, we found preferred direction enhancement for stimulation of the lower and the central pair of columns. Data shows the mean ± SEM in 10 T5-cells measured in 8 different flies. (e) Responses of T4 (open circles) and T5 cells (closed squares) to apparent motion stimuli in preferred (red colors) and null direction (black colors) between the two central columns 0 and 1. Compared to the responses of T4, T5 responses to two-pulse sequences along the preferred (PD) and the null (ND) direction are shifted to larger stimulus sizes. Data represent the mean values ± SEM of 10 T5 cells measured in 5 flies and of 7 T4 cells in 4 different flies, respectively. Black asterisks represent statistically significant (t-test, $p$-value<0.05) differences for null-direction responses of T4-cells and T5-cells, red asterisks for preferred direction responses. (f) Nonlinear response components of T4 and T5 cells. Same dataset as in Figure 1e.
Figure 3. A common mechanism for direction selectivity in all four subtypes of T4 and T5 cells. (a) Pictograms indicating the stimulus positions and the preferred and null-direction of the respective layer. (b) Nonlinear response components of T4-cells to apparent motion stimuli in different layers of the
Figure 3 continued

lobula plate. For T4 cells projecting to all four layers, preferred direction enhancement and null direction suppression are found to be spatially distributed within the receptive field such that enhancement is found on the preferred side while suppression is predominant on the null side of the receptive field. Data represent the mean ± SEM of 6, 8, 7 and 9 T4 cells (from layer 1–4). (c) Nonlinear response components of T5 cells to apparent motion stimuli in different layers of the lobula plate. Data represent the mean ± SEM of 8, 5, 6 and 13 T5 cells (from layer 1–4).

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Figure 4. Directional tuning and selectivity of T4 and T5 cells. (a) Example of directional tuning to grating motion as determined by the vector sum of responses to grating motion along four cardinal directions. All neurons within each layer have almost identical preferred directions. (b) Histogram of preferred directions within all four layers. Clear peaks appear at the four cardinal directions. Data were obtained from 5 different flies. (c) Direction selectivity within each layer, as defined by the difference between the preferred and null direction responses, divided by the sum. Data represent the mean ± SEM obtained from 5 flies (same data set as in b). (d) Same as c, but flies were stimulated by ON and OFF edges, respectively. Data represent the mean ± SEM obtained from 3 flies.

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