# **Supplemental Figures**



#### Figure S1. L2 in ort-null flies does not respond to 20-ms light or dark flashes.

#### Fig. S1 Image Description

Responses of L2 in  $ort^{1}/Df(3R)BSC809$  flies lacking *ort* rescue to a 20-ms light flash (left, blue) or a 20-ms dark flash (right, red) with a 500-ms gray interleave. The solid line is the mean response and the shading is  $\pm 1$  SEM. n = number of cells (number of flies).



Figure S2. Linear filtering properties of idealized impulse responses. Fig. S2 Image Description

(A) Idealized impulse responses that approximately match those measured throughout this study. The peak amplitude of the second phase was varied to produce monophasic or biphasic responses with a specific ratio between the areas of the first and second phases; the time-to-peak of the first and second phases as well as the decay of the second phase were held constant. Response amplitude is in arbitrary units (A.U.).

(B) The magnitude across frequencies for each of the impulse responses in (A), computed as the absolute value of the Fourier transform. Note that when the second phase becomes larger than the first phase (area ratio = 2), the filter is low-pass like the monophasic impulse response.

(C) Simulated response to a 250-ms dark step off of gray computed by convolving the impulse responses in (A) with the stimulus. Note the delayed hyperpolarization produced by the area ratio = 2 impulse response.

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Figure S3. Asymmetric adaptation differentially deblurs and enhances light and dark edges. Fig. S3 Image Description

(A) Simulated responses to a motion-blurred light edge (left) and dark edge (right) for models with different degrees of asymmetric adaptation (g values). All other parameters are the same as those used to simulate L1 impulse responses in Figure 3. When g = 0, only dark drives the feedback element; when g = 1, only light drives the feedback element; when g = 0.5, both light and dark equally drive the feedback element. Note that when g = 0, the response following a dark-to-light transition is faster and stronger, but the response to a light-to-dark transition is like that of the no-feedback model. When g = 1, the response following a light-to-dark transition is faster and stronger, but the response to a dark-to-light transition is like that of the no-feedback model. When g = 0 or 1. Responses are plotted with inversion over the x-axis to facilitate comparisons with the inputs.

(B) As in (A), but with parameters used to simulate L2 impulse responses in Figure 3.



Figure S4. Half-wave rectification preserves deblurring and enhancement of edges in the L1 and L2 pathways. Fig. S4 Image Description

(A) Light (blue) and dark (red) edge inputs.

(B) Simulated responses to motion-blurred light and dark edges by an L1 pathway-like model incorporating L1-type dynamic feedback followed by a sign-inversion and half-wave rectification. Dotted lines are the responses of a no-feedback model. Thin lines are the portion of the L1 response that was eliminated by the rectification. Note the faster and stronger response to a dark-to-light transition.

(C) Simulated responses to motion-blurred light and dark edges by an L2 pathway-like model incorporating L2-type dynamic feedback followed by half-wave rectification. Note the faster and stronger response to light-to-dark transition as well as the faster response to a dark-to-light transition.



**Figure S5. Increased feedback improves deblurring at fast image speeds for horizontal motion.** Fig. S5 Image description

Violin plots of the temporal offset between the natural image and model output at high (left, 600 °/s) and low (right, 150 °/s) image speeds, using w = 0 (model without feedback), w = 12.7 (full feedback model), and w = 24 (model with increased feedback). Images were blurred with motion in the horizontal direction. Solid lines indicate the lower quartile, median, and upper quartile. Dotted gray line indicates temporal offset = 0 ms. \*\*\* p<0.001, Wilcoxon signed-rank test.



#### Figure S6. Feedback improves model predictions of L2 responses to a naturalistic stimulus when fit to those responses Fig. S6 Image Description

(A) A natural image was converted to grayscale without gamma correction and blurred based on the optics of the fly eye.

(B) Yaw velocity sequence from a trajectory of a walking fly that was used to determine the sequence of pixels in the natural image from which to draw stimulus intensity values.

(C) Full model of L2 responses to the naturalistic stimulus using parameters fitted to the naturalistic stimulus response (overlaid on the stimulus inverted across the x-axis)

(**D**) Modeled responses without feedback to the naturalistic stimulus using parameters fitted to the naturalistic stimulus response (overlaid on the stimulus inverted across the x-axis)

# **Appendix: Image Descriptions**

## **Fig. 1 Long Description**

- A. Two schematized line plots connected by an inhibitory arrow going from R1-6 to L1/L2
  - Top (R1-6): A trace begins at the x-axis, sharply increases above the axis to a large maximum peak, and then gradually decreases back down to the x-axis.
  - Bottom (L1/L2): A trace begins at the x-axis, sharply decreases below the axis to a large minimum trough, sharply increases to a small maximum peak above the x-axis, and then gradually decreases back down to the x-axis.
- B. Anatomy diagram with regions labeled lamina and medulla. The more distal lamina region includes the axonal arbors of R1-6 adjacent to the dendritic arbors of L1/L2. The more proximal medulla region includes the axonal arbors of L1 and L2. L1 has two axonal arbors (the more proximal one of which is boxed), and L2 has one axonal arbor (which is also boxed).
- C. Schematized line plot exemplifying how the stimulus-aligned voltage responses will be displayed in panels D-E and quantified in panel F. The y-axis is  $-\Delta F/F$  (in %); it is inverted so that depolarization is up and hyperpolarization is down. The x-axis is time (in seconds). A single trace begins at a baseline "0%"  $\Delta F/F$  level, goes downward to a sharp trough (1st peak), then upward to a shallower peak above baseline (2nd peak), and then gradually returns to baseline. The area between the trace and the baseline axis is labeled Phase 1 Area and Phase 2 Area, corresponding to the two "lobes" formed by the 1st and 2nd peaks, respectively. The lengths of time it takes to reach the two peaks are labeled Phase 1 t<sub>peak</sub> and Phase 2 t<sub>peak</sub>. Above this trace, there is a thin rectangular box representing the visual stimulus, with a small segment at the beginning shaded white indicating the short impulse flash, and a longer segment shaded gray indicating the mean gray interleaf period. The white segment spans approximately half the length of time it takes to reach Phase 1 t<sub>peak</sub>. (For clarity, peaks that are oriented downward in plots will be referred to as "troughs" or "minima" in image descriptions. Peaks that are oriented upward will be referred to as "peaks" or "maxima." Note that this is the opposite of the numeric minima and maxima, as negative  $\Delta F/F$  values are oriented upward and positive  $\Delta F/F$  values are oriented downward.)
- D. Two line plots showing the average responses of L1 to light and dark flashes (n = 103 cells, from 13 flies). For each plot, the stimulus schematic is displayed at the top, and the response trace is underneath, as in C. The x-axis is in time, spanning 0 to 0.5 second. The y-axis is in  $-\Delta F/F$ , spanning +8% to -4% (values provided in this description are approximate).
  - The light response begins at baseline (0%), goes downward to a sharp trough at +6%, and then goes upward back to the baseline, where it remains for the rest of the gray period of the stimulus. The width of the first trough is about twice the duration of the light flash.
  - The dark response begins at baseline (0%), goes upward to a sharp peak at -3%, goes downward past baseline to a trough at +3%, and then gradually goes upward back to baseline, reaching 0% near the end of the gray period. The width of the first peak is about twice the duration of the light flash.
- E. As in D, but for L2 (n = 68 cells, from 14 flies).
  - The light response begins at baseline (0%), goes downward to a sharp trough at +5%, goes upward past baseline to a small peak at -1%, and then gradually goes downward back to baseline, reaching the baseline around 0.15 second. The width of the first trough is about twice the duration of the light flash.
  - The dark response begins at baseline (0%), goes upward to a sharp peak at -3%, goes downward past baseline to a trough at +2%, and then gradually goes upward back to baseline, reaching baseline near the end of the gray period. The width of the first peak is about twice the duration of the light flash.
- F. Five panels of dot-and-whisker plots quantifying the impulse responses shown in D-E. Each panel has four sets of dot-and-whiskers corresponding to the mean and 95% confidence interval from L1 light, L1 dark, L2 light, and L2 dark. Approximate dot and whisker values (and their statistical comparisons) are summarized in the following table as "dot ± whiskers" or "dot ± (-lower, +upper)" when whiskers are asymmetric:

	Phase 1 Area (- %ΔF/F * s)	Phase 2 Area (-%ΔF/F * s)	Area Phase 2 / Phase 1	Phase 1 t <sub>peak</sub> (s)	Phase 2 t <sub>peak</sub> (s)
L1 light	$0.13 \pm 0.015$	$-0.005 \pm 0.01$	$0 \pm negligible$	0.017 ± (-0.008, +negligible)	No data
L1 dark	$0.045 \pm 0.01$	$0.23\pm0.03$	$5.2 \pm 0.9$	0.016 ± (-0.007, +0.01)	0.04 ± (-0.005, +0.01)
L2 light	$0.1 \pm 0.005$	$0.05 \pm 0.01$	0.050 ± negligible	0.017 ± (-0.008, +negligible)	0.04 ± (-0.005, +0.01)
L2 dark	$0.050 \pm 0.005$	$0.12\pm0.015$	$2.4 \pm 0.4$	0.016 ± (-0.007, +0.01)	0.05 ± (-0.01, +0.02)
*** (p<0.001)	L1 light & dark; L2 light & dark; L1 & L2 light	All pairs	All pairs	None	None
n.s. (p>0.05)	L1 & L2 dark	None	None	All pairs	L2 light & dark L1 & L2 dark

Back to Fig. 1

# **Fig. 2 Long Description**

Title above A-C: "L2 responses to photoreceptor input only (ort-neuron silencing)" Title above D-F: "L2 responses to photoreceptor input and recurrent feedback (L2 ort rescue)"

- A. Five circles representing photoreceptors, L2 cells, and three types of feedback neurons. There are arrows representing the following possible circuit pathways, as well as X's over some arrows representing silenced pathways under the ort-neuron silencing condition (ort driver expressing TNT):
  - Photoreceptors to L2
  - Photoreceptors to lateral feedback neurons
  - Lateral feedback neurons to L2 (X, silenced)
  - Lateral feedback neurons to downstream feedback neurons (X, silenced), then to L2
  - L2 to recurrent feedback neurons (X, silenced), then back to L2
- B. Two panels of line plots showing the average responses of L2 to light and dark flashes under the ort-neuron silencing condition. The x-axis is in time, spanning 0 to 0.5 second. The y-axis is in  $-\Delta F/F$ , spanning +8% to -4% (values provided in this description are approximate).
  - Each panel has three overlaid traces for the following genotypes:
    - ort-lexA control (n = 129 cells from 11 flies)
    - lexAop-TNT control (n = 96 cells from 9 flies)
    - $\circ$  ort-lexA, lexAop-TNT experimental (n = 116 cells from 9 flies)
  - Light impulse panel:
    - The two control traces are largely the same; both begin at 0%, go downward to a sharp trough at +5%, go upward past 0 to a small peak at -1%, and then gradually go downward back to 0, reaching 0 around 0.2 second. The width of the troughs is around twice the duration of the light flash.
    - The experimental trace begins at 0%, goes downward to a sharp trough at +6%, gradually goes upward back to 0 (without crossing 0), reaching 0 at around 0.2 second. The width of the trough is around four times the duration of the light flash.
  - Dark impulse panel: All three traces are initially largely the same; all begin at 0%, go upward to a sharp peak at -2.5%, and go downward past 0 to a trough at +2%. The control traces immediately start returning

to 0 gradually, reaching 0 at around 0.4 second. In contrast, the experimental trace perdures at +2% for around 0.2 second before gradually returning to 0, reaching 0 near the end of the gray period.

C. Five panels of dot-and-whisker plots quantifying the impulse responses shown in B. Each panel has dot-and-whiskers corresponding to the means and 95% confidence intervals from six conditions, which include light and dark responses from three genotypes: ort-lexA control (Control 1), lexAop-TNT control (Control 2), and ort-lexA, lexAop-TNT (experimental). Approximate dot and whisker values (and their statistical comparisons) are summarized in the following table as "dot ± whiskers" or "dot ± (-lower, +upper)" when whiskers are asymmetric:

	Phase 1 Area (-%ΔF/F * s)	Phase 2 Area (-%ΔF/F * s)	Area Phase 2 / Phase 1	Phase 1 t <sub>peak</sub> (s)	Phase 2 t <sub>peak</sub> (s)
Control 1, light	0.11 ± 0.01	0.05 ± 0.02	0.5 ± negligible	0.016 ± (-0.008, +negligible)	0.08 ± 0.025
Control 2, light	$0.09 \pm 0.01$	0.06 ± 0.02	0.8 ± negligible	0.016 ± (-0.008, +negligible)	0.06 ± (-0.01, +0.02)
Experimental, light	0.25 ± 0.03	0.005 ± negligible	0 ± negligible	0.025 ± negligible	No data
Control 1, dark	0.050 ± negligible	0.13 ± 0.03	3.2 ± 1.5	0.024 ± (-0.008, +negligible)	0.04 ± negligible
Control 2, dark	0.055 ± negligible	0.13 ± 0.02	3.0 ± 1	0.016 ± (- negligible, +0.008)	0.04 ± (- negligible, +0.02)
Experimental, dark	0.055 ± negligible	0.20 ± 0.03	4.4 ± 1.5	0.025 ± negligible	0.1 ± (-0.02, +0.08)
*** (p<0.001)	Light responses	Light responses	Light responses	Light responses	None
** (p<0.01)	None	Dark responses	None	None	Dark responses
n.s. (p>0.05)	Dark responses	None	Dark responses	Dark responses	None

- D. Five circles representing photoreceptors, L2 cells, and three types of feedback neurons. There are the same arrows as in A representing possible circuit pathways, but without X's, as there is no presynaptic silencing under the L2 ort rescue condition (ort null background with L2-Gal4 > UAS-ort). The L2 circle has membrane receptors representing the presence of ort receptors. The lateral feedback neuron circle has membrane receptors that are crossed out, representing the absence of ort receptors in these cells. Thus, the pathway from photoreceptors to L2 is intact, while the pathway from photoreceptors to lateral feedback neurons is blocked.
- E. As in B, but for the L2 ort rescue condition:
  - Each panel has three overlaid traces for the following genotypes: (all of which additionally include L2-Gal4, UAS-ort):
    - $\circ$  Df(3R)BSC809/+ control (n = 94 cells from 7 flies)
    - $\circ$  ort<sup>1</sup>/+ control (n = 59 cells from 5 flies)
    - $Df(3R)BSC809/ort^1$  experimental (n = 58 cells from 6 flies)

- Light impulse panel: All three traces are largely the same; all begin at 0%, go downward to a sharp trough at around +5%, go upward past 0 to a small peak at -1%, and then gradually go downward back to 0, reaching 0 around 0.15 second. The width of the first trough is around twice the duration of the light flash.
- Dark impulse panel: All three traces are initially largely the same; all begin at 0% and go upward to a sharp peak at -3%. The control traces go downward past 0 to a trough at +2%, whereas the experimental trace only goes to +1%. All three traces then gradually return to 0, reaching 0 at about 0.3 second. The width of the first peak is around twice the duration of the light flash.
- F. As in C, but quantifying the L2 ort rescue impulse responses shown in E. Each panel includes light and dark responses from three genotypes (all of which additionally include L2-Gal4, UAS-ort): Df(3R)BSC809/+ control (Control 1), ort<sup>1</sup>/+ control (Control 2), and Df(3R)BSC809/ort<sup>1</sup> (experimental).

	Phase 1 Area (-%ΔF/F * s)	Phase 2 Area (-%ΔF/F * s)	Area Phase 2 / Phase 1	Phase 1 t <sub>peak</sub> (s)	Phase 2 t <sub>peak</sub> (s)
Control 1, light	0.11 ± 0.01	$0.04 \pm 0.01$	$0.4 \pm negligible$	0.017 ± negligible	0.09 ± (-0.05, +0.4)
Control 2, light	$0.10 \pm 0.01$	$0.06 \pm 0.02$	$0.6 \pm 0.1$	0.011 ± (-0.003, +0.007)	0.05 ± negligible
Experimental, light	$0.09 \pm 0.01$	$0.04 \pm 0.02$	$0.5 \pm 0.1$	0.017 ± negligible	0.05 ± negligible
Control 1, dark	$0.050\pm0.005$	$0.14 \pm 0.02$	$3.0\pm0.5$	0.017 ± negligible	0.04 ± negligible
Control 2, dark	$0.055\pm0.01$	$0.14 \pm 0.02$	$2.5\pm0.5$	0.016 ± (-0.008, +negligible)	0.04 ± negligible
Experimental, dark	$0.060 \pm 0.005$	$0.09 \pm 0.02$	$1.5 \pm 0.3$	0.017 ± negligible	0.04 ± negligible
* (p<0.05)	None	Dark responses	Dark responses	None	None
n.s. (p>0.05)	Light and dark responses	Light responses	Light responses	Light and dark responses	Light and dark responses

Back to Fig. 2

# Fig. 3 Long Description

- A. Schematic and equations describing the recurrent feedback model:
  - Schematic: Circles and arrows representing the neural circuit simulated by this computational model. The three main elements are the visual input, L1/L2 ( $\nu$ ), and the feedback element (y). L1/L2 and the feedback element have an additional variable describing their temporal properties ( $\tau_{\nu}$  and  $\tau_{y}$ , respectively) The arrows indicate the following circuit pathways, whose strength is described by additional variables:
    - $\circ$  Visual input feeds forward to L1/L2, which feeds forward to the rest of the visual system
    - $\circ$  L1/L2 also feeds forward to the feedback element with variable strength dependent on g
    - The feedback element provides oppositely-signed feedback to L1/L2, with strength w
  - Equations:

$$\circ \quad \tau_v \dot{v} = -v - w * y + Input$$

$$\tau_{y}\dot{y} = -y + f(g, sign(v)) * v$$

$$f(g, sign(v)) = \begin{cases} 1 - g & \text{if } v > 0 \\ g & \text{if } v < 0 \end{cases}$$

- B. Four panels of line plots showing modeled L1 and L2 responses to light and dark flashes (overlaid on top of the measured impulse responses from Fig. 1D). All four sets of modeled responses overlap closely with the measured responses (except for the measured responses having more noise). There are two subtle differences in all the cases: in the modeled responses, the first peak is slightly rounder, and the second peak has an onset that is less sharp (resulting in a peak that is slightly later and smaller on the onset side).
- C. Two panels of line plots, each with eight lines, showing modeled L1 and L2 responses to motion blurred edges with varying feedback strengths (w = 0, 1, 2, 4, 8, and 12). The L1 and L2 conditions are qualitatively similar and will be described together, with noticeable differences specified. The y-axis is Activation (A.U.) spanning -1 to 1, and the x-axis is time (s) spanning 0 to 0.8 second. Values provided are approximate.
  - The input is a dotted line that starts at 0, drops down to -1 at 0.15s, and then jumps up to 0.5 at 0.62s.
  - The blurred input is a dotted line that matches the input, except the two transitions (from 0 to -1, and from -1 to 0.5) are slower and smoother, taking 0.1s to transition rather than being instantaneous.
  - The w = 0 trace is shaped similarly to the blurred input trace, but is shifted later. Like the input, it also travels from 0 to -1 to 0.5 A.U. The transition from 0 to -1 occurs at 0.18-028s, and the transition from -1 to 0.5 occurs at 0.65-0.75s.
  - For L1, the w = 12 trace initially follows the w = 0 trace, but instead of going all the way down to -1, it stops at -0.25 and turns around, reaching -0.1 at around 0.25s. It stays at -0.1 until 0.65s, and then increases to a peak of 0.9 at 0.75s.
  - For L2, the w = 12 trace is qualitatively similar but initially goes a bit further before stopping at -0.5, and takes a bit longer to get back to -0.1 at around 0.35s.
  - The remaining w values have traces that span the range between the w = 0 and w = 12 traces.
  - An arrow above both plots at around 0.65 emphasizes the difference between the traces at that time point. The larger the *w* value, the higher the activation value is at that time, showing that an increase in *w* results in a more rapid readout of the -1 to 0.5 transition due to the feedback engaged during the initial 0 to -1 transition.

# Back to Fig. 3

### **Fig. 4 Long Description**

- A. A series of eight example images depicting different stages of the computational modeling process:
  - i. Natural image (example photo of a skyline from the van Hateren dataset, cropped and in grayscale; the remaining images are all processed versions of this photo)
  - ii. Optically blurred image.
  - iii. Optically and motion blurred image.
  - iv. Thresholded natural image.
  - v. Thresholded output of the model without feedback.
  - vi. Difference between iv and v (the thresholded natural image and the thresholded output of the model without feedback).
  - vii. Thresholded output of the full model with feedback.
  - viii. Difference between iv and vi (the thresholded natural image and the thresholded output of the full model with feedback).

Note that when comparing the thresholded natural image with the two thresholded model outputs, the full model is more closely aligned with the original image than is the model without feedback.

B. Panel titled Vertical Motion, with two violin plots displaying the temporal offsets generated from the model without feedback and the full model with recurrent feedback. The x-axes are of temporal offset in milliseconds, ranging from -160 to 160. The model without feedback has quartiles at -59, -54, and -51, and a range from -98 to -

43 (rounded to the nearest digit). The full feedback model has quartiles at -30, -5, and 47, and a range from -119 to 156. These distributions are significantly different (\*\*\*, p<0.001).

C. As in B, but titled Horizontal Motion. The model without feedback has quartiles at -63, -59, and -56, and a range from -94 to -44. The full feedback model has quartiles at -42, -34, and -13, and a range from -79 to 156. These distributions are significantly different (\*\*\*, p<0.001).

Back to Fig. 4

## **Fig. 5 Long Description**

- A. Two panels labeled High Speed and Low Speed, each with three violin plots displaying the distribution of temporal offsets generated from the model without feedback, the full feedback model, and the model with increased feedback. The x-axes are of temporal offset in milliseconds, ranging from -160 to 160. The following describes the violin plots with statistics rounded to the nearest digit:
  - High speed:
    - The model without feedback has quartiles at -85, -74, and -66, and a range from -156 to -48.
    - The full feedback has quartiles at -39, -16, and 19, and a range from -156 to 156.
    - The model with increased speed has quartiles at -31, -7, and 40, and a range from -121 to 156.
    - All three distributions are significantly different from each other (\*\*\*, p<0.001).
  - Low speed:
    - The model without feedback has quartiles at -45, -42, and -41, and a range from -64 to -36.
    - The full feedback has quartiles at -21, 5, and 58, and a range from -110 to 156.
    - The model with increased speed has quartiles at -12, 18, and 76, and a range from -106 to 156.
    - All three distributions are significantly different from each other (\*\*\*, p<0.001).
- B. Four panels of line plots showing modeled L1 and L2 responses to light and dark flashes. Each panel has a trace labeled "increased feedback" overlaid on a control trace (which is from Fig. 2B). In all four panels, the control and increased feedback traces overlap closely during the first phase (the initial peak or trough). During the second phase, there is no difference in the L1 light response, but the rest of the responses have larger peak amplitudes with increased feedback. The peak amplitudes are increased by around 25%, 100%, and 50% for L1 dark, L2 light, and L2 dark, respectively.
- C. Four panels of line plots showing experimental L1 and L2 responses to light and dark flashes. Each panel has a trace labeled "CDM" overlaid on a control trace (which is from Fig. 1D-E). The x-axis is in time, spanning 0 to 0.5 second. The y-axis is in  $-\Delta F/F$ , spanning +8% to -4% (values provided in this description are approximate).
  - L1 light response: The CDM trace (n = 56 cells, 13 flies) has a trough at +4%, compared to +6% for the control. The CDM also takes longer to return to 0%, around 0.1s instead of 0.05s for the control.
  - L1 dark response: The CDM trace initially peaks at -2%, compared to -3% for the control, and the trough during the second phase is slightly larger, about 3.5% instead of 3% for the control.
  - L2 light response: The CDM trace (n = 102 cells, 7 flies) has a similar first phase compared to the control, but it has a reduced peak in the second phase, around -0.5% instead of -1% for the control.
  - L2 dark response: The CDM trace has a similar first phase compared to the control, but it has a much larger trough in the second phase, which is around +5% compared to +2% for the control.

### Back to Fig. 5

# Fig. 6 Long Description

A. Bottom: Line plot in which the y-axis is Intensity ranging from 0 to 1, and the x-axis is time ranging from 0 to 2 seconds. A single trace fluctuates smoothly throughout most of the range of light intensities over time, starting mostly at the brighter end of the range until around 0.6s, then traveling to the dimmer end of the range until around 1.1s, then returning to the brighter end of the range until the end, except for a rapid excursion to the dimmer end around 1.7s. The initial bright period has two prominent local minima and two prominent local maxima, whereas the middle dim period and the final bright period have smaller fluctuations, except for the final dim excursion. Note that panel B will specifically highlight three short segments of this stimulus: 1. The steep

transition from the initial bright period to the middle dim period, 2. A portion of the middle dim period with several small fluctuations, and 3. The rapid dim excursion toward the end of the stimulus (specifically at the steep dim-to bright transition).

Top: Rectangle with variable shading in grayscale visualizing the change in brightness of this naturalistic stimulus over time (as described in the line plot).

B. Top: Line plot with two overlaid traces and scale bars for the x- and y-axes. The x-axis is in time, spanning 2 seconds. The y-axis is in  $-\Delta F/F$  (inverted such that depolarization is up and hyperpolarization is down), spanning approximately 6%. The first trace is an inverted, normalized version of the naturalistic stimulus (the inversion allows dimming of the stimulus and depolarization of L2 to follow the same direction, and vice versa). The second trace shows the average voltage response of L2 (n = 52 cells, from 7 flies), which generally follows the trajectory of the stimulus, lagging slightly behind in time.

Bottom: Three notable segments of the stimulus and response are displayed as insets:

- In response to a rapid bright-to-dim transition, the L2 response begins to increase (depolarize) after the start of the stimulus transition, but this depolarizing excursion then peaks sooner than the end of the stimulus transition.
- In response to a period of subtle changes in brightness, L2 responds to each increase or decrease in brightness with relatively large amplitudes.
- In response to a rapid bright-to-dim stimulus transition, the L2 response begins to decrease (hyperpolarize) after the start of the stimulus transition, but this hyperpolarizing excursion then peaks sooner than the end of the stimulus transition.
- C. Line plot with three overlaid traces and scale bars for the x- and y-axes. The y-axis is in arbitrary units, spanning approximately 0.08 a.u., and the x-axis is time, spanning 2 seconds. The first trace is an inverted, normalized version of the naturalistic stimulus. The second trace is a normalized version of the measured response of L2. The third trace is the output of the full recurrent feedback model using the same parameters as for the impulse responses. The measured L2 response and the model output are well correlated ( $r^2 = 0.70$ ), and the two traces both generally follow the trajectory of the stimulus, lagging slightly behind in time.
- D. Same as C, except the third trace is the output of the model without feedback. This trace is less closely correlated with the measured L2 response ( $r^2 = 0.39$ ) than in C, and at times it lags noticeably behind not only the stimulus but also the measured L2 response.
- E. Same as C, except the third trace is the output of the convolutional model using the impulse response as a linear filter. This trace is less closely correlated with the measured L2 response ( $r^2 = 0.39$ ) than in C, and at times it lags noticeably behind not only the stimulus but also the measured L2 response. Compared to the model without feedback in D, the convolutional model has only subtle differences, following some changes in brightness more rapidly and accurately than in D, but sometimes exaggerating the amplitude of smaller changes relative to those of larger changes.

Back to Fig. 6

### **Figure S1 Long Description**

Two line plots showing the average responses of L2 to light and dark flashes in a Df(3R)BSC809/ort<sup>1</sup> control background (n = 121 cells, from 6 flies). The y-axis is in  $-\Delta F/F$ , spanning +8% to -4%. The x-axis is in time, spanning 0 to 0.5 second. In both the light and dark flash plots, the control trace remains at baseline (0%) throughout the entire duration of the stimulus.

Back to Fig. S1

### **Figure S2 Long Description**

- A. Three line plots with x-axes spanning 0 to 0.5s, and y-axes spanning around -1 to 1 A.U., schematizing impulse responses with the following waveforms:
  - Monophasic, Area Ratio = 0: The line goes from 0 to -1, and then back to 0.

- Biphasic, Area Ratio ~ 1: The line goes from 0 to -1, crosses 0 to a second peak at 0.2, and then gradually returns to 0.
- Biphasic, Area Ratio ~ 2: The line goes from 0 to -1, crosses 0 to a second peak at 0.4, and then gradually returns to 0.
- B. Three Fourier plots with x-axes spanning 0 to 40 Hz, and y-axes spanning 0 to 2 in magnitude.
  - Monophasic: The line monotonically decreases from 1.9 at 0 Hz to 0 at 40 Hz.
  - Biphasic (1:1): The line increases from 0.4 at 0 Hz to a peak of 1.8 at 5 Hz, and then decreases to 0 at 40 Hz.
  - Biphasic (2:1): The line monotonically decreases from 1.5 at 0 Hz to 0 at 40 Hz.
- C. Three line plots with simulated responses to an alternating dark and light flash, with x-axes spanning 0 to 0.5s, and y-axes spanning -4 to 4 in response amplitude. The dark flash lasts from 0 to 0.25s, and the light flash lasts from 0.25 to 0.5s.
  - Monophasic: The line increases from 0 to 2 between 0-0.05s, stays at 2 until 0.25s, and decreases back to 0 between 0.25-0.3s.
  - Biphasic (1:1): The line increases from 0 to 2 between 0-0.05s, and decreases gradually back to 0 between 0.05-0.25s. It then decreases from 0 to -2 between 0.25-0.3s, and increases gradually back to -0.2 between 0.3-0.5s.
  - Biphasic (2:1): The line increases from 0 to 2 between 0-0.05s, and decreases sharply past 0 to -2 between 0.05-0.25s. It then decreases from -2 to -4 between 0.25-0.3s, and increases to -0.5 between 0.3-0.5s.

Back to Fig. S2

## **Figure S3 Long Description**

- A. Two panels of line plots showing modeled L1 responses to a moving light or dark edge. Each panel has six lines showing modeled L1 responses to motion blurred edges with varying g values, (g = 0, 0.5, 1), modeled L1 responses without feedback, and the input stimuli. The y-axis is Activation (A.U.) spanning -1 to 1, and the x-axis is time (s) spanning 0 to 0.8 second. Values provided here are approximate.
  - Moving light edge
    - The input is a dotted line that starts at 0, increases to 1 at 0.15s, and then decreases to -0.5 at 0.62s. (This is sign-inverted from the examples in Fig. 3C.)
    - The blurred input is a dotted line that matches the input, except the two changes (from 0 to 1, and from 1 to -0.5) are slower and smoother, taking 0.1s to transition rather than being instantaneous.
    - The model-without-feedback trace and the g = 0 trace are both shaped similarly to the blurred input trace, but are shifted later. Like the input, they also travel from 0 to 1 to -0.5 A.U. The transition from 0 to 1 occurs at 0.18-028s, and the transition from 1 to -0.5 occurs at 0.65-0.75s. The g = 0 trace is slightly different at the end of the stimulus, where it stops decreasing at -0.25 and begins to return to 0, whereas the model-without feedback trace continues to decrease until it reaches -0.5 like the stimulus does.
    - The g = 1 trace differs in that it initially overlaps with the g = 0 trace, but instead of going all the way up to 1, it stops at 0.25 and goes back down, reaching 0.1 at around 0.3s. It stays at 0.1 until 0.65s, and then decreases to a trough of -1 at 0.75s.
    - The g = 0.5 trace is intermediate between the g = 0 and g = 1 traces (but is closer to the g = 1 trace).
  - Moving dark edge
    - The input is a dotted line that starts at 0, drops to -1 at 0.15s, and then jumps to 0.5 at 0.62s. (This is the same as Fig. 3C.)
    - The blurred input is a dotted line that matches the input, except the two changes (from 0 to -1, and from -1 to 0.5) are slower and smoother, taking 0.1s to transition rather than being instantaneous.
    - The model-without-feedback trace and the g = 1 trace are both shaped similarly to the blurred input trace, but are shifted later. Like the input, they also travel from 0 to 1 to -0.5 A.U. The transition from 0 to 1 occurs at 0.18-028s, and the transition from 1 to -0.5 occurs at 0.65-0.75s. The g = 1 trace is slightly different at the end of the stimulus, where it stops decreasing at -0.25

and begins to return to 0, whereas the model-without feedback trace continues to decrease until it reaches -0.5 like the stimulus does.

- The g = 0 trace differs in that it initially overlaps with the g = 1 trace, but instead of going all the way down to 1, it stops at 0.25 and goes back up, reaching -0.1 at around 0.3s. It stays at -0.1 until 0.65s, and then increases to a peak of 1 at 0.75s.
- The g = 0.5 trace is intermediate between the g = 1 and g = 0 traces (but is closer to the g = 0 trace).
- An arrow above both plots at around 0.65 emphasizes the difference between the traces at that time point, where the impact of the variably engaged feedback levels is apparent. By that time point, the light edge has engaged feedback when g = 1 (or 0.5), altering their responses to stimuli at 0.65s relative to the model without feedback. In contrast, the dark edge responses show evidence of engaged feedback when g = 0 (or 0.5).
- B. Same as A, but for L2. The input, blurred input, and model-without-feedback traces are the same as in A. The model-without-feedback trace still overlaps with the g = 0 trace for the light edge, and the g = 1 trace for the dark edge. The g = 0.5 trace is also still intermediate between the g = 0 and g = 1 traces. An arrow is also above both plots at around 0.65 to emphasize the difference between the traces at that time point, where the impact of the variably engaged feedback levels is apparent.
  - Moving light edge: The g = 1 trace stops increasing a bit higher than in A, at 0.5 (instead of 0.25). After the second transition, it decreases to a larger trough of -1.2 (instead of -1).
  - Moving dark edge: The g = 0 trace stops decreasing a bit lower than in A, at -5 (instead of -0.25). After the second transition, it increases to a larger peak of 1.2 (instead of 1).

Back to Fig. S3

# Figure S4 Long Description

The x-axis of these plots is time (s) spanning 0 to 0.8s. The y-axis of these plots is Activation (A.U) spanning -1 to +1.

- A. Input: Line plot showing the same moving light edge and moving dark edge stimuli as in Fig. S3 overlaid together. The light edge starts at 0, increases to +1, and then decreases to -0.5. The dark edge starts at 0, decreases to -1, and then increases to +0.5.
- B. L1 Pathway: Line plot showing modeled L1 responses to the stimuli in A. Responses greater than zero are shown as more saturated/darker than responses less than zero to distinguish between responses that are known to be relevant to downstream neurons after half-wave rectification has occurred.
  - Dotted lines show responses from the model without feedback. The light response transitions from 0 to +1, remains at +1, and then transitions to -0.5. The dark response transitions from 0 to -1, remains at -1, and then transitions to +0.5. The saturated (>0) and unsaturated (<0) portions of this set of responses are symmetric.
  - Solid lines show responses from the full model with feedback. The light response transitions from 0 to +1, decreases slightly to +0.8, and then transitions to -0.25, following a similar time course as the dotted no-feedback light response. The dark response transitions from 0 to -0.4, turning around sooner than the no-feedback dark response and staying at -0.2; it then increases faster and more strongly to +0.8 compared to the no-feedback dark response. The saturated (>0) and unsaturated portions of this set of responses is not symmetric, and notably the feedback model of the saturated portion has a faster response at 0.65s than the no-feedback model.
- C. L2 Pathway: Line plot showing modeled L2 responses to the stimuli in A. Responses greater than zero are shown as more saturated/darker than responses less than zero to distinguish between responses that are known to be relevant to downstream neurons after half-wave rectification has occurred.
  - Dotted lines show responses from the model without feedback, which are sign inverted relative to the L1 pathway in B. The light response transitions from 0 to -1, remains at -1, and then transitions to +0.5. The dark response transitions from 0 to +1, remains at +1, and then transitions to -0.5. The saturated (>0) and unsaturated (<0) portions of this set of responses are symmetric.
  - Solid lines show responses from the full model with feedback. The light response transitions from 0 to 0.75, increases gradually to -0.4, and then transitions to +0.5, following a faster time course than the no-

feedback light response. The dark response transitions from 0 to +0.5, turning around sooner than the no-feedback dark response and staying at +0.1; it then increases faster and more strongly to -1 compared to the no-feedback dark response. The saturated (>0) and unsaturated portions of this set of responses is not symmetric, though the feedback model of both portions have faster responses at 0.65s than the no-feedback model.

Back to Fig. S4

### **Figure S5 Long Description**

Two panels labeled High speed and Low speed, each with three violin plots displaying the distribution of temporal offsets generated from the model without feedback, the full feedback model, and the model with increased feedback. The x-axes are of temporal offset in milliseconds, ranging from -160 to 160. The following describes the violin plots with statistics rounded to the nearest digit:

- High speed:
  - $\circ$  The model without feedback has quartiles at -92 , -80, and -71, and a range from -156 to -55.
  - The full feedback has quartiles at -53, -45, and -28, and a range from -149 to 156.
  - The model with increased speed has quartiles at -43, -34, and -16, and a range from -131 to 156.
  - All three distributions are significantly different from each other (\*\*\*, p<0.001).
- Low speed:
  - $\circ$  The model without feedback has quartiles at -47, -45, and -44, and a range from -57 to -38.
  - The full feedback has quartiles at -32, -25, and -3, and a range from -68 to 156.
  - The model with increased speed has quartiles at -27, -16, and 17, and a range from -50 to 156.
  - All three distributions are significantly different from each other (\*\*\*, p<0.001).

Back to Fig. S5

#### **Figure S6 Long Description**

- A. Series of three images. The first is a panoramic color photo of forest scenery, consisting mostly of tree trunks at various distances. The second is the same photo after removing gamma correction and converting to grayscale. The third is a further modified version of the photo after applying optical blurring.
- B. Line plot in which the y-axis is yaw velocity (degrees per second) ranging from -400 to 400, and the x-axis is time (seconds) ranging from 0 to 2. A single trace fluctuates smoothly throughout most of the range of yaw velocities over time, beginning at approximately -50 degrees/second at time 0. The trace has four prominent peaks at approximately (-250, 0.2), (400, 0.6), (-200, 1.1), (300, 1.7), with several smaller local extrema in between the four peaks.
- C. Line plot with three overlaid traces and scale bars for the x- and y-axes. The y-axis is in arbitrary units, spanning approximately 0.08 a.u., and the x-axis is time, spanning 2 seconds. The first trace is an inverted, normalized version of the naturalistic stimulus (see Fig. 6A for description). The second trace is a normalized version of the measured response of L2. The third trace is the output of the full recurrent feedback model after fitting it to the naturalistic stimulus. The measured L2 response and the model output are closely correlated ( $r^2 = 0.90$ ), and the two traces both generally follow the trajectory of the stimulus, lagging slightly behind in time.
- D. Same as C, except the third trace is the output of the model without feedback. This trace is less closely correlated with the measured L2 response ( $r^2 = 0.56$ ) than in C, and at times it lags noticeably behind not only the stimulus but also the measured L2 response.

Back to Fig. S6