In terms of circuit function, additional new questions arise from the study at hand. Does the presence of supernumerary L4 branches have any consequence on the total number of L4 branches innervating a single cartridge? Furthermore, is synaptic specificity affected in Dscam2 mutants?

Importantly, these novel mutant flies could help to better understand the role of L4 cells and their tiling of the lamina in visual processing. The lamina is known to be involved in motion detection and L4 and L2 neurons belong to the so-called OFF pathway. A recent study showed that L4 neurons play a key role motion processing, resolving previous contradictory studies on its function (Meier et al., 2014). It was also found that visual circuit miswiring in the absence of Dscam2 leads to specific and conditional differences in visual acuity (Bosch et al., 2015). Strikingly, in three of the behavioral paradigms tested, Dscam2 mutants showed an opposing response as wild-type flies. Therefore, disrupted modularity in the Dscam2-deficient optic lobe leads to aberrant cross-wiring of visual columns resulting in deficits in light and motion detection. The new Dscam2 and Dscam4 mutant lines generated by Tadros et al. (2016) should help to assess and model the function of L4 dendritic tiling in visual processing.

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Can You Hear Me Now?

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Auditory communication is central to the social interactions of many animals. In fruit flies, males sing to court females. Coen et al. (2016) demonstrate that males can dynamically adjust the loudness of their songs according to the distance to a female.

The animal kingdom abounds with examples of strange mating rituals. Bowerbirds decorate elaborate nests; blue-footed boobies kick up their brilliant blue feet and dance; many humans post carefully curated representations of their lives online and wait for prospective mates to swipe right. Fruit flies, too, engage in elaborate courtship rituals that involve, among other things, male flies serenading females with songs. Central to all of these rituals is communication: prospective mates evaluate the suitability of a potential partner by the sensory cues they provide. As a result, careful calibration and delivery of these signals is critical to successful courtship. How does one implement an effective signaling strategy that maximizes one's chances of mating? The fruit fly brain is relatively simple, and powerful genetic tools and experimental techniques facilitate dissection, manipulation, and quantification of neural circuit activity and behavioral responses. Thus, song production by male flies provides an ideal model for investigating the neural underpinnings not only of courtship rituals in particular, but also of complex sensorimotor transformations in general. In this issue of Neuron, Coen et al. describe a new level of sophistication in how male flies tune their songs in response to female movements.

Fly songs, produced by the extension and vibration of a wing, are composed of two modes: sine song, a low-frequency humming, and pulse song, a train of high-frequency, high-amplitude pulses separated by inter-pulse intervals (IPIs)
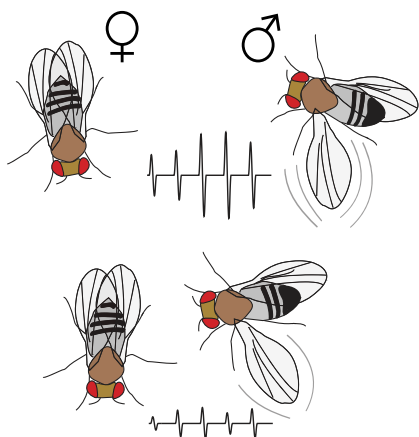
Figure 1. Male Fruit Flies Dynamically Adjust Pulse Song Amplitudes as a Function of Female Distance
Traces illustrate the structure of male song produced by extension and vibration of the wing, with the amplitude of the pulses being greater when the female is further away (top pair) and smaller when the female is closer (bottom pair).

of characteristic length (Yamamoto and Koganezawa, 2013). Song structure varies among species and is one of the central cues that allow females to identify conspecifics. Females will not mate with males that sing the wrong song, and experienced males will only sing to conspecific, virgin females. A growing body of work has begun to clarify the mechanisms by which males identify receptive females and initiate singing (Clowney et al., 2015; Clyne and Miesenböck, 2008; Kohatsu and Yamamoto, 2015; Kohatsu et al., 2011; Shirangi et al., 2013; von Philipsborn et al., 2011; Zhou et al., 2015). The song pathway begins with approximately 20 neurons, so-called P1 neurons, located in the protocerebrum, which activate descending neurons and the central pattern generators in the thorax that drive singing (von Philipsborn et al., 2011; Yamamoto and Koganezawa, 2013). Activation of these neurons is necessary and sufficient for singing (von Philipsborn et al., 2011), and in vivo calcium imaging has demonstrated P1 activity correlates with male courtship preferences (Clowney et al., 2015; Kohatsu et al., 2011). Remarkably, olfactory, gustatory, visual, and auditory sensory pathways converge on P1 neurons (Clowney et al., 2015; Kohatsu and Yamamoto, 2015; Kohatsu et al., 2011; Zhou et al., 2015). Olfactory and gustatory

circuits provide feedforward excitatory and inhibitory input onto P1 (Clowney et al., 2015), and the relative balance of excitation and inhibition is thought to allow males to discriminate among prospective mates. For example, female pheromones result in net excitation of P1 neurons, whereas male pheromones result in net inhibition. Visual motion cues are not normally sufficient to induce persistent courtship (Agrawal et al., 2014), but upon activation of P1 neurons, either optogenetically or by a brief presentation of female pheromones, presentation of moving dot stimuli induces persistent song production and tracking behavior (Kohatsu and Yamamoto, 2015). Altogether, this suggests that P1 neurons are command neurons that initiate and maintain song production in response to multimodal sensory input, controlling whether males court.

To woo or not to woo, however, is not the only question. Courtship is costly, both in terms of energy input and time commitment, and thus males would do well to adjust their courting strategies in response to feedback from females so as to maximize their chance of success. Indeed, variability in song structure, previously attributed to neural noise, can in fact be largely accounted for by the dynamic sensory experience of the male (Coen et al., 2014), suggesting that male fruit flies modify their songs in real time to more effectively court. Building on this result, here Coen et al. (2016) demonstrate that male flies modulate the amplitude of their pulse songs as a function of the distance to the female, singing louder when females are father away (Figure 1). This phenomenon, *amplitude modulation with distance* (AMD), has clear ethological relevance across species in various contexts and had previously only been identified in humans and song birds (Brumm and Slater, 2006; Zahorik and Kelly, 2007).

AMD is a sensorimotor transformation that requires that a male fly first estimate the distance between himself and the female, and second, adjust the amplitude of his singing such that he sings louder when the female is farther away. How do male flies estimate female distance? AMD in humans and song birds is thought to be due to visual input, and previous work has demonstrated that *Drosophila*

can identify the closer of two targets using visual cues (Schuster et al., 2002). Coen et al. show that AMD is abolished in blind flies, and thermal activation of the P1 command neurons allowed tethered flies to perform AMD in response to moving black squares presented on a screen, even in the absence of other sensory cues. Thus, visual cues are necessary and sufficient for AMD. Male flies are also capable of AMD when the visual stimulus is restricted to a single eye, indicating that a monocular mechanism, rather than binocular stereopsis, is involved in distance estimation. The two major mechanisms for monocular distance estimation are motion parallax, in which self-motion causes distant objects to move across the visual field more slowly than closer objects, and object expansion, where approaching objects get larger on the retina. However, male self-motion is not required for AMD, and silencing neurons in the visual system that are known to contribute to motion vision or loom sensation did not abolish AMD. Thus, the neural circuits underlying distance estimation during AMD remain to be elucidated.

Once males estimate female distance, how do they adjust the amplitude of their pulse songs? Male flies continue to perform AMD during courtship even when P1 neurons are activated. This suggests that, in contrast to other sensory pathways, the visual signals that control AMD impinge upon the circuitry that controls song production downstream from P1 neurons. Indeed, thermal activation of downstream neurons—thoracic dPR1 neurons—causes a reversal in the sign of AMD, with males singing louder when females are closer, and activation of putative central pattern generator neurons—vPR6 neurons—abolished AMD altogether. This suggests that while the balance of excitatory and inhibitory input from multiple sensory pathways onto P1 neurons may gate song production, additional sensory pathways, including visual pathways involved in female distance estimation, may control the pattern and amplitude of the song. Moreover, Coen et al. found that song pattern and amplitude are controlled separately, as there is no correlation between pulse song amplitude and the IPI. Two primary groups of muscles control wing

movements in *Drosophila*. The direct flight muscles attach to the base of the wing and have been implicated in controlling the temporal structure of song (Shirangi et al., 2013). The indirect flight muscles attach to the thorax and are important for power generation during flight (Moore et al., 2000). Coen et al. found a strong correlation between indirect flight muscle spiking activity and pulse amplitude, but no correlation between spiking activity and IPI. Together, these results suggest that the indirect flight muscles control pulse amplitude, whereas the direct flight muscles independently control song timing.

Although the ability to modulate song intensity with distance makes intuitive sense, it remains to be seen how AMD contributes to mating success. Regardless, these studies reveal an intriguing level of sophistication to courtship song, with male fruit flies modulating their tune following a control strategy that was previously thought to be the exclusive province of larger, more complex vertebrate brains. Future studies in the fly will enable this strategy to be understood at the circuit and algorithmic level, shedding new light on how animal communication is shaped by interactions between individuals.

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Recurrent Feedback Loops in Associative Learning

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In this issue of *Neuron*, Gao et al. (2016) report on a little-studied feedback pathway from the cerebellar nuclei back to the cerebellar cortex. They find that it contributes to associative conditioning and execution of learned movements, highlighting a role for local feedback loops in the brain.

The brain, like other computational devices, often uses feedback to fine-tune and optimize its computations. For example, iterative computational circuits frequently sample their own output to estimate error in their intended outcome and use this information to improve their computational accuracy in the next iteration. In addition to the traditional global feedback loop that originates from the final output of the circuit, local feedback loops also offer computational benefits.

Consider, for example, a computational device made of a number of discrete operational stages, each of which transforms the information it receives to generate its own output. In such a case, local feedback loops, embedded in each operational stage, offer tremendous flexibility and specificity for modulation of the overall computational capacity and accuracy of the device. In many cases, local feedback mechanisms are indispensable for circuit optimization, for example, when the computation performed at each stage is not linear, or when additional (external) inputs are incorporated at each operational stage. In addition to providing a mechanism for error minimization, feedback can also be used to alter the gain of a circuit, or as a plasticity mechanism for learning.

In artificial computational devices, feedback mechanisms work at both the local and global level. It is therefore