

Putting a kink in a familiar control loop

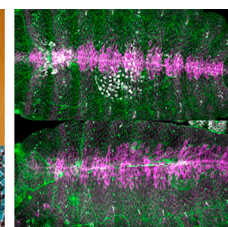
Study shows that a feed-forward signaling loop controls dorsal closure in flies.

Development doesn't always follow the plan. Ducuing et al. identify a control loop that enables *Drosophila* embryos to cope with environmental or genetic challenges and close a gap in their backs (1).

Environmental changes or inappropriate gene expression can upset the intricate cellular choreography of development. Organisms have mechanisms to buffer these disturbances and ensure that cells end up in the right places and adopt the right identities (2). During dorsal closure, two sheets of moving cells converge and seal an opening on the back of the embryo (3). Previous studies have shown that two molecules, JNK and DPP, are necessary for dorsal closure (4). If embryos lack either one, the gap remains open. DPP works by shutting down another protein, Brinker, that blocks dorsal closure. However, researchers didn't know how DPP and JNK cooperate to promote this process.

The leading edge cells at the rim of the dorsal opening are distinctive. Ducuing et al. found that DPP turns on production of three markers for these cells. Researchers had assumed that JNK was upstream of DPP in a linear pathway. To test this idea, the team created embryos that produced a dominant-negative form of JNK in bands along their bodies. DPP diffuses into these stripes from surrounding cells, so, if JNK were upstream of DPP, the three markers should still be expressed. The markers were absent, however, suggesting that DPP and JNK operate in parallel.

Both molecules, in fact, were essential for the differentiation of leading edge cells. Shutting down either JNK or DPP signaling within bands along the embryos' bodies disrupted differentiation. Then the team activated JNK or DPP. If only active JNK was present, cells outside the leading edge differentiated into leading



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(Left to right) Charlotte Keeley, Stéphane Vincent, Bertrand Mollereau, and Antoine Ducuing reveal how the proteins DPP, JNK, and Brinker work together to promote dorsal closure in *Drosophila* embryos. (Far right) In a control embryo that has just undergone dorsal closure (top), cells that were around the rim of the narrowing gap are distinctive, as indicated by the presence of the marker Jaguar (purple); green indicates cadherin. The researchers found that these cells also differentiated after deletion of the protein Brinker and the receptor for DPP (bottom).

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edge cells, but that didn't occur when only DPP was active.

These results make sense if JNK and DPP belong to a feed-forward loop. Instead of a linear pathway, the pathway containing the two molecules is triangular. JNK is part of the direct branch that stimulates genes that promote specialization of leading edge cells. DPP resides in the indirect branch. After JNK switches it on, DPP spurs leading edge cells to differentiate by targeting Brinker.

Feed-forward loops are good at filtering out noise such as mistimed gene activity. To determine whether the JNK-DPP loop performs that function, Ducuing et al. studied *puckered* mutants—in which JNK is randomly and inappropriately activated—that also lacked Brinker. The intensity of JNK activity varies from cell to cell, and the researchers observed that cells exposed to a weak JNK signal, which could be extraneous, were less likely to produce one of the leading edge markers than were cells exposed to a strong signal, which is more likely to be “real.” Brinker therefore gives cells the ability to discriminate between weak and strong JNK signals.

The JNK-DPP loop also helps ensure that dorsal closure withstands environmental challenges. At 25°C, leading edge cells differentiated normally in embryos lacking Brinker. But at 32°C, some cells outside the leading edge morphed into leading edge cells. The embryos couldn't prevent inappropriate differentiation when Brinker was absent.

“The study teaches us that a pathway that is well known can have different functions in patterning,” says senior author Stéphane Vincent. In this loop JNK promotes leading edge differentiation, and the DPP-Brinker branch serves as a filter, preventing differentiation unless it receives a sufficiently strong JNK signal. Feed-forward loops control other developmental steps, and DPP and JNK work together in other aspects of *Drosophila* development. But researchers don't yet know if these JNK-DPP interactions also involve feed-forward loops.

1. Ducuing, A., et al. 2015. *J. Cell Biol.* <http://dx.doi.org/10.1083/jcb.201410042>.
2. Waddington, C.H. 1959. *Nature*. 183:1654–1655.
3. Belacortu, Y., and N. Paricio. 2011. *Dev. Dyn.* 240:2379–2404.
4. Glise, B., and S. Noselli. 1997. *Genes Dev.* 11:1738–1747.

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