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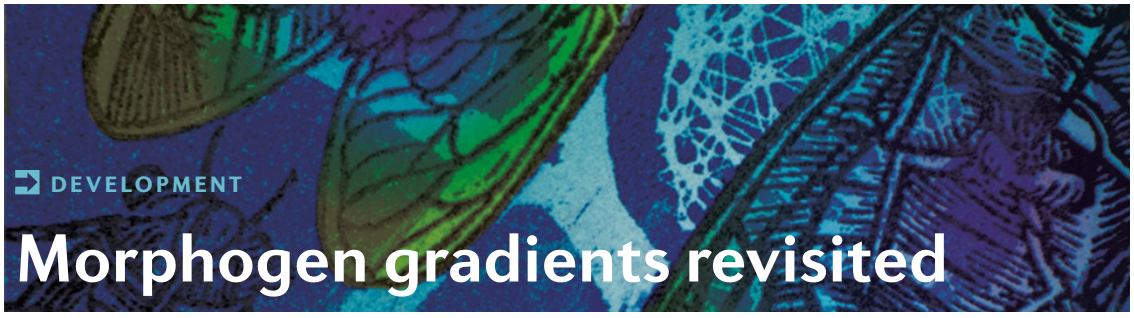

DEVELOPMENT

Morphogen gradients revisited

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> patterning and growth can proceed without an instructive gradient of secreted Wg

WNTs are secreted signalling proteins that control patterning and growth during development. They spread from where they are synthesized, forming a gradient and activating target gene expression at a distance in a concentration-dependent manner. However, the actual requirement for WNTs to spread had not been directly tested. Now, Vincent and colleagues show that a membrane-tethered version of Wingless (Wg), which is the main WNT in *Drosophila melanogaster*, can also control development, as its expression leads to viable flies that produce normally patterned appendages of almost the right size.

Using homologous recombination, the authors replaced the endogenous *wg* gene with one that expresses a fusion protein comprising the type 2 transmembrane protein Neurotactin and Wg (NRT–Wg). This enabled them to generate flies that expressed membrane-tethered Wg at physiological levels in the absence of the wild-type form. Interestingly, these flies were viable and developed normally patterned appendages and cuticular structures — the wings were only slightly smaller than in control animals.

The role of WNT gradients has been most extensively studied in *D. melanogaster* wing imaginal discs, which are small pockets of embryonic epithelial cells that give rise to fully patterned wings. Normally, Wg is produced throughout the prospective wing field in early larval stages to establish the wing primordium and then becomes restricted to a narrow strip of cells at the dorsoventral boundary. It is thought that high Wg signalling activity near the dorsoventral boundary activates the *senseless* target gene and that further away from this boundary, low-level Wg signalling (due to decreasing levels of spreading Wg) activates more sensitive target genes such as *vestigial*, *Distal-less* and *frizzled*. Surprisingly, the authors observed that in flies expressing only NRT–Wg, *senseless* expression was the same as in wild-type flies and that the expression of the other three target genes dropped more sharply (in relation to the distance from cells expressing NRT–Wg) than in controls. However, this did not seem to affect adult wing patterning.

So, how does NRT–Wg activate genes at a distance, given that it cannot spread? The authors used flies in which *Nrt–Wg* gene expression could be switched off to confirm that the protein is not unusually stable and that NRT–Wg-expressing cells do not release any functional Wg. Indeed, analysis of mosaic imaginal discs of NRT–Wg flies that contained groups of interspersed *wg*-mutant cells confirmed that NRT–Wg does not activate *Distal-less* expression beyond adjoining cells, whereas control experiments showed that wild-type Wg activates the same target gene over a distance of several cell diameters. However, by carefully analysing the expression of *wg* by fluorescence *in situ* hybridization, they found that *wg* is transcriptionally active throughout the wing primordium until later in development than previously thought, persisting during a critical period of patterning and growth. Thus, this could provide a local source of low-level Wg protein. Moreover, it was also observed that all cells of the prospective wing blade are derived from cells that express *wg* before its expression becomes restricted to the dorsoventral boundary.

Although *wg* transcription persists further in development than previously thought, an additional mechanism is required to sustain growth and patterning without secreted Wg. Vincent and colleagues observed that the expression of some target genes, such as *vestigial* and *Distal-less*, persists when signalling is prematurely terminated. Although the mechanisms underlying this 'memory' of earlier signalling remain to be defined, this suggests that target gene expression in the prospective blade does not require Wg spreading from the dorsoventral boundary.

Although the authors find that there is a slight delay in the overall rate of larval development and decreased fitness, this study indicates that patterning and growth can proceed without an instructive gradient of secreted Wg. This might prompt researchers to revisit the requirement for the long-range spreading of other WNTs in development.

Kim Baumann

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