Drosophila Wnt/Fz Pathways

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Wnts [also known as Wingless (Wg)] are a family of conserved signaling molecules involved in a plethora of fundamental developmental and cell biological processes, such as cell proliferation, differentiation, and cell polarity. Dysregulation of the pathway can be detrimental, because several components are tumorigenic when mutated and are associated with hepatic, colorectal, breast, and skin cancers. First identified in the fruit fly *Drosophila melanogaster* as a gene family responsible for patterning the embryonic epidermis, the *Wnt* gene family, including *Wg*, encode secreted glycoproteins that activate receptor-mediated signaling pathways leading to numerous transcriptional and cellular responses. The main function of the canonical Wg pathway is to stabilize the cytoplasmic pool of a key mediator, beta-catenin [β-catenin, known as Armadillo (Arm) in fruit flies], which is otherwise degraded by the proteasome pathway. Initially identified as a key player in stabilizing cell-cell adherens junctions, Arm is now known to also act as a transcription factor by forming a complex with the lymphoid enhancer factor (LEF)/T cell–specific transcription factor (TCF) family of high mobility group (HMG)-box transcription factors. Upon Wnt/Wg stimulation, stabilized Arm translocates to the nucleus, where, together with LEF/TCF transcription factors, it activates downstream target genes that regulate numerous cell biological processes.

Description

This record contains information specific to the Drosophila Wnt/Fz Pathways.

The study of the *Drosophila* Wnt homolog Wingless (Wg) has provided a unique paradigm for the genetic analysis of the Wnt signaling pathway (abbreviated here as Wnt/Wg). Mutations in *Wg* were identified in screens for embryonic and adult phenotypes. Phenotypic similarity and epistasis analyses have placed several other genes, such as *Armadillo* (*Arm*), *Dishevelled* (*Dsh*), *zeste white 3* (*zw3*), *Drosophila Axin* (*Daxin*), *pygopus* (*pygo*), *legless* (*lgs*), and *Pangolin* (*Pan*) in the same pathway. The *Drosophila* genome encodes seven Wnt proteins, four Frizzled (Fz) receptors, and two adenomatous polyposis coli (APC) proteins, but contains only a single copy of each of the genes *Dsh*, *Arm*, *Pan*, and *Zw3* (*1*).

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In the absence of Wg signaling, the cytoplasmic concentration of Arm (the *Drosophila* homolog of β-catenin) is kept low through constitutive phosphorylation by the kinase Zw3 [the *Drosophila* homolog of glycogen synthase kinase-3β (GSK-3β)] and subsequent degradation by the ubiquitin pathway. Upon exposure to Wg, the signal is relayed through the Wingless receptor, Frizzled-2 (Fz2) and a transmembrane coreceptor called Arrow, which is the *Drosophila* homolog of low-density lipoprotein-related-protein 5 and 6 (LRP5/6). The association of Wg, Fz2, and Arrow leads to the activation of the signal transduction cascade presumably by the membrane recruitment and activation of Dsh, which eventually leads to the phosphorylation and consequent inactivation of Zw3. The increased protein concentration then allows Arm to form a complex with the transcriptional repressor Pan [a transcription factor of the T cell–specific transcription factor (TCF) family], and thereby release the Pan-dependent repression of Wg target genes.

The Wnt/Wg pathway can also be activated by inhibiting negative regulators, such as GSK-3β, APC, or Axin, that promote β-catenin degradation, or by introducing activating mutations in β-catenin that renders it incapable of interacting with the degradation complex, thus stabilizing the cytosolic pool. Wnt/Wg signaling can also be activated through an alternative "noncanonical" pathway that may lead to protein kinase C (PKC) and c-Jun N-terminal kinase (JNK) activation, resulting in calcium release and cytoskeletal rearrangements, respectively. See the Wnt/Ca^{2+}/cyclic GMP Pathway, which describes Wnt signaling through PKC (*2*).

The canonical Wg signaling pathway, which is described by this Wnt/Fz pathway (Fig. 1), has been well studied during embryonic and larval development (*3*). The extracellular distribution of Wg during embryonic and imaginal disc development has been used as a model system to dissect gradient formation of extracellular signaling molecules. Wg is required for patterning and cell fate decisions in the embryonic epidermis. One of the experimental strategies used to dissect the organization of signaling pathways is genetic epistasis analysis, which allows one to order signaling components in a hierarchical pathway. For example, Noordermeer *et al*. (*4*) analyzed the embryonic segment polarity phenotypes in double mutants generated by expressing Wg in various mutant backgrounds. Ectopic expression of Wg leads to an expanded Engrailed (En) expression domain, which can be suppressed by removing the activity of genes that are required downstream for the reception and intracellular transmission of the signal. Epistasis experiments showed that no expansion of En in response to ectopic Wg occurs in the absence of either Dsh or Arm activity. These observations suggest that Dsh and Arm act downstream of Wg reception and are required for the intracellular transduction of the signal.

Wg is required during both early and late stages of *Drosophila* development (*3*, *5*, *6*). In the embryo, Wg is in-

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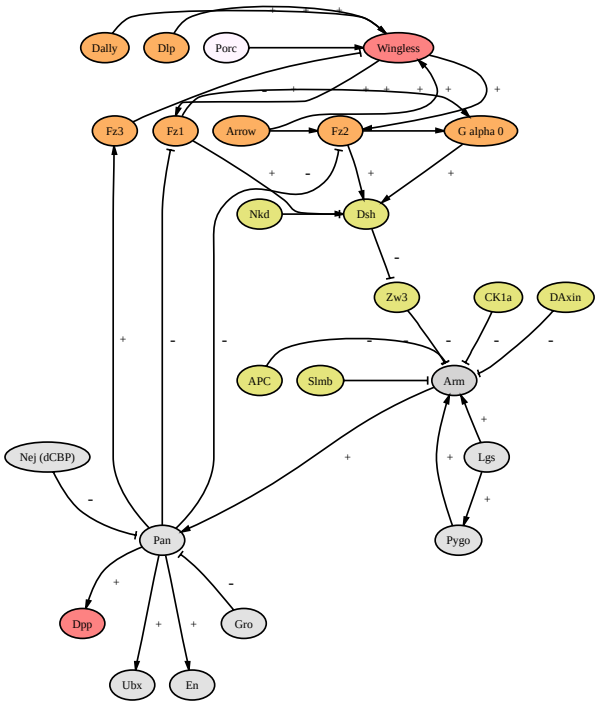

Fig. 1. Pathway image captured from the dynamic graphical display of the information in the Connections Maps available 18 April 2005. This version of the pathway includes the latest data from (*14*). For a key to the colors and symbols and to access the underlying data, please visit the pathway (http://stke.sciencemag.org/cgi/cm/stkecm;CMP_6459).

volved in segmentation and patterning of the embryonic epidermis, and in the development and patterning of the *Drosophila* eye and head structures, nervous system, midgut, and malpighian tubules (*7*). During larval development, Wg is required for development of imaginal discs, in particular for dorsoventral patterning of the leg and wing, bristle specification, and eye development.

Wg signaling in the *Drosophila* eye and wing has been efficiently used to screen for modifiers of the signal transduction cascade (*8*–*11*). For example, overexpression of both Wg and a constitutively activated form of Arm in the eye results in a cell death-associated "small eye" phenotype, whereas ectopic activation of the pathway in the wing gives rise to the formation of extra sensory wing margin bristles. In contrast, inhibition of Wg signaling in the wing by either overexpression of a dominant-negative form of the Wg-receptor Fz2 or that of E-cadherin (a cell adhesion molecule critical for adherans junction formation that also physically interacts with Arm and hence sequesters it from its signaling role in the nucleus) leads to loss of wing margin bristles and wing notch phenotypes. Several new pathway members, as well as modifiers, of the Wg pathway have been identified by conducting enhancer-suppressor screens for the above-mentioned phenotypes in the eye or wing, including the genes encoding Pygo and Lgs, which are critical for the transduction of the Wg signal. For an historic representation of the pathway before these new elements were added, see Figure 2.

The Wnt/Wg signaling pathway is a complex pathway, and two recent reports have added to the understanding of new mechanisms and regulators of the pathway. First, Tolwinski *et al*. (*12*), have suggested a new mechanism (and perhaps a branch) for activation of the Wnt/Wg pathway: They provide evidence that it is the physical proximity of the coreceptor, Arrow, and the Wg receptor, Fz2, that triggers the activation of the signal transduction cascade (*12*). The research revealed that Arrow recruited Axin to the membrane by physically interacting with it, thereby causing its degradation. The degradation of Axin subsequently leads to the stabilization of Arm and activation of the Arm-Pan-mediated target genes. Importantly, this process of Axin degradation was independent of Zw3 activity, which was shown previously to be critical for the activation of the Wg pathway. Second, Katanaev *et al*. (*13*) provided evidence for the involvement of heterotrimeric guanine nucleotide-binding proteins (G proteins) in the Wg signal transduction pathway in flies (*13*). They demonstrate the role of $G\alpha_0$ subunit in transducing signals from Frizzleds and provide evidence for the involvement of G proteins in both the canonical Wg-Arm-mediated signaling and the planar cell polarity (PCP) pathway, which also involves Wg. This is the first such evidence for the role of G proteins in Wnt/Fz signaling in flies.

Most recently, to gain a global understanding of the Wnt/Wg pathway and to identify novel players, DasGupta *et al*. (*14*) have undertaken a whole-genome RNAi-mediated screen in *Drosophila* clone8 cells. In this genome-wide screen for regulators of the Wnt/Wg pathway, several new candidate genes were identified that positively or negatively regulate Wg/Wnt

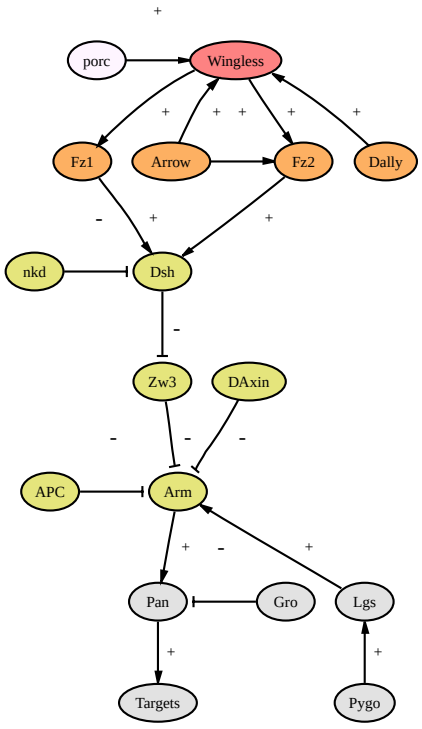

Fig. 2. Historic pathway image captured from the dynamic graphical display of the information in the Connections Maps available 22 February 2005. This version of the pathway does not include the most recent data. For a key to the colors and symbols and to view the most current information, please visit the pathway (http://stke.sciencemag.org/cgi/cm/stkecm;CMP_6459).

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signaling [see Table 1 in the supplementary tables (http://stke.sciencemag.org/cgi/content/full/sigtrans;CMP_6459/DC1)]. More than 50% of the candidate genes identified in the screen have identifiable vertebrate orthologs, and mutations in several human homologs are directly related to known human diseases (see Table 2 in the supplementary tables). Functional studies for several candidate genes have been conducted in mammalian cultured cells and in zebrafish to demonstrate the applicability of the *Drosophila* screen to vertebrate and mammalian systems.

Pathway Details

URL: http://stke.sciencemag.org/cgi/cm/stkecm;CMP_6459
Scope: Specific
Organism: invertebrates: arthropods: insects: *Drosophila*
Canonical Pathway: Wnt/beta-catenin Pathway (http://stke.sciencemag.org/cgi/cm/stkecm;CMP_5533)

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