

Lecture 5. MCB 141. Dorsal-ventral patterning of the *Drosophila* embryo.
Reading: Chapter 21, Molecular Biology of the Gene, 7th Edition.

1. The differential activation of the Toll receptor leads to a broad Dorsal nuclear gradient in the precellular *Drosophila* embryo, approximately 90 min following fertilization. This Dorsal gradient works at about the same time as the Bicoid gradient, but instead of regulating differential gene expression along the anterior-posterior axis, it controls gene activity across the dorsal-ventral axis.

2. High levels of the Dorsal gradient activate a target gene called Snail within ventral regions that form the mesoderm. Snail is essential for EMT, epithelial-mesenchyme transitions, and is specifically required for the invagination of the mesoderm during gastrulation. Mutant embryos that lack Snail fail to gastrulate.

3. Low levels of the Dorsal gradient activate another target gene, short gastrulation (Sog), in lateral regions of the embryo that form the neurogenic ectoderm. This tissue ultimately produces the central nervous system of the adult fly. In principle, both high and low levels of the Dorsal gradient can activate Sog expression in both the mesoderm and neurogenic ectoderm. However, the Sog enhancer contains binding sites for both Dorsal and Snail, which functions as a transcriptional repressor to exclude Sog expression in the mesoderm and thereby restricts its activity to the neurogenic ectoderm.

4. The third Dorsal target gene that is critical for dorsal-ventral patterning is called Dpp. In principle, Dpp can be activated throughout the embryo by ubiquitous maternal activators. However, dpp is repressed in both ventral and lateral regions of the early embryo by the Dorsal gradient. Dorsal mainly functions as an activator, but can mediate repression when it binds next to specialized AT-rich elements (TTYNTTSAT). (Unlike short-range Snail and Gap repressors, the Dorsal/AT elements function as long-range silencers. For example, Snail represses stripe 2 expression in the ventral mesoderm when its binding sites are placed within the stripe 2 enhancer. In contrast, the Dpp silencer element can repress stripe 2 activity in ventral regions even when it maps far away from the stripe 2 enhancer.)

5. Sog encodes a secreted protein that is related to the vertebrate Chordin inhibitor of BMP signaling. You will hear quite a lot about BMP signaling later in the course. The Sog inhibitor completely excludes DPP activity in the neurogenic ectoderm. So, the combination of transcriptional repression by the Dorsal gradient and protein inhibition by Sog ensures that Dpp activity is excluded from the future CNS and restricted to the dorsal ectoderm.

6. Name that phenotype. There are mutants that completely lack the Dorsal nuclear gradient. What do the resulting embryos look like? There is no Snail expression and so there is no mesoderm. There is no Sog expression and so there is no neurogenic ectoderm. Dpp is no longer restricted to the dorsal ectoderm, but instead is expressed throughout the embryo, thereby converting all three major tissues, the mesoderm, neurogenic ectoderm, and dorsal ectoderm, into dorsal ectoderm.

7. Let's briefly consider gastrulation. Snail is activated in approximately 1,000 cells comprising the embryonic mesoderm. These cells undergo coordinated invagination at about 3 hours after fertilization. Once the mesoderm enters the blastocoel it undergoes migration along the inner surface of the lateral ectoderm. This migration is promoted by interactions of FGF8 (Thisbe) expressed in the neurogenic ectoderm and the FGF receptor (Heartless) expressed in the mesoderm.

8. The mesoderm spreads as a monolayer along the inner neurogenic ectoderm and the lateral-most cells enter the dorsal ectoderm. These cells receive the localized Dpp signal and activate genes such as tinman, which are essential for the differentiation of the dorsal mesoderm forming the heart.

9. Why is the FGF receptor called heartless? Predict the consequences of restricting snail expression to the ventral-most 12-14 cells rather than its normal pattern of expression, which encompasses 18-20 cells including ventral-lateral regions.