

Gradients and Regulatory Networks of Wnt Signaling in *Hydra* Pattern Formation

Thomas W. Holstein

Department for Molecular Evolution and Genomics Heidelberg University

The Wnt/ β -catenin pathway plays an important role in axial patterning during metazoan development. Wnt genes are expressed at the site of the blastopore in cnidarian and many bilaterian embryos organizing the oral-aboral body axis. How this Wnt-expressing organizer evolved in metazoan evolution and how it is regulated is largely unknown.

We study organizer formation in two cnidarian models, in *Hydra* and *Nematostella*. Our data indicate that the *Hydra* head organizer (and the *Nematostella* blastoporus organizer) represents an organizer ancestral to those in bilaterians. Previously, we identified the cnidarian Wnt genes revealing a genetic complexity similar to chordates (Hobmayer et al 2000, Kusserow et al 2005, Lengfeld et al 2009). The kinetics of Wnt gene expressions during head formation in intact and regenerating polyps indicate that a cascade of consecutive Wnt activation with Wnt3 at the top of the cascade. Recombinant HyWnt3 can also rescue a head regeneration deficient mutant strain reg-16. In order to understand the maintenance of Wnt signaling in the Hydra head organizer we used Hy β -Catenin and HyWnt3::EGFP transgenic animals. Using *Hydra* transgenesis, we investigated cis-regulatory sequences of Hydra Wnt3 (HyWnt3). We generated transgenic Hydra with a HyWnt3 promoter driving eGFP reporter gene expression in the head organizer. We found that an autocatalytic regulatory network maintains the *Hydra* head organizer, which includes a feedback loop of Wnt signaling and TCF binding sites in the Wnt promoter. Repressor elements in the Wnt promoter are required to suppress ectopic Wnt expression outside of the head organizer.

Our data point to the importance of transcriptional regulation, which localizes the Wnt signaling center to a specific region along the body axis. With respect to these new data, we will discuss the molecular nature of the *Hydra* axial patterning mechanisms and the evolutionary conserved character of the organizer.