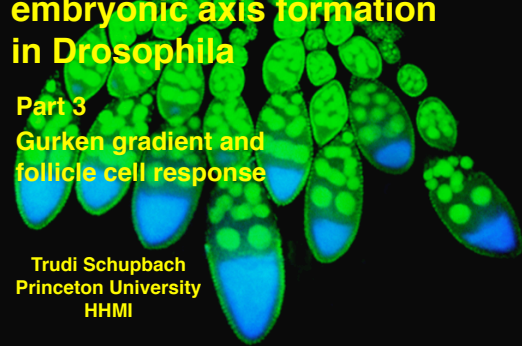


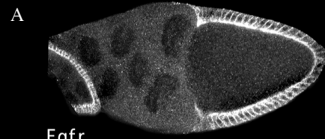
Maternal control of embryonic axis formation in *Drosophila*

Part 3 Gurken gradient and follicle cell response

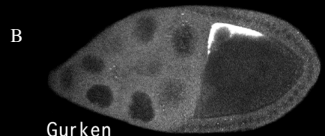
Trudi Schupbach
Princeton University
HHMI



Protein expression pattern of EGF receptor and its ligand, Gurken



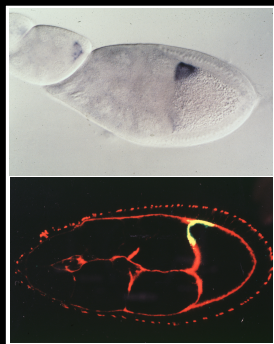
Egfr



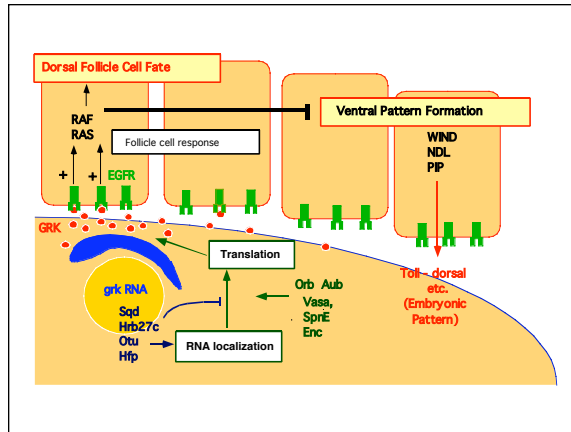
Gurken

A: from B.Z. Shilo: Sapir et al (1998) Development

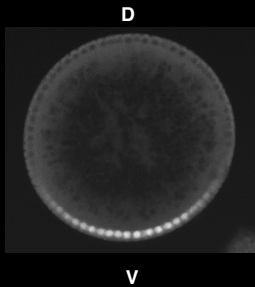
Gurken RNA and protein are localized in the oocyte



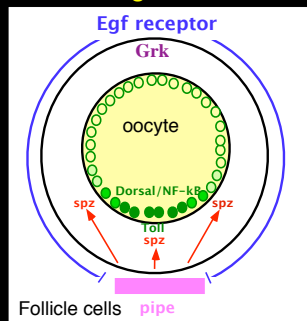
Neuman-Silberberg & Schupbach, 1993, 1996



A nuclear gradient of Dorsal protein determines the DV axis of the embryo

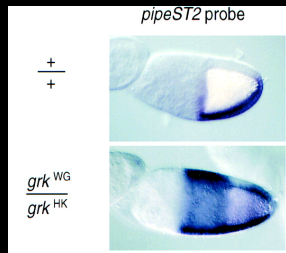


A signaling pathway activated through Pipe generates the gradient of nuclear Dorsal/NF- κ B



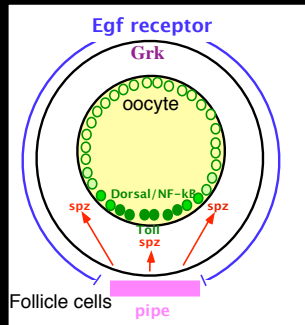
KV Anderson and colleagues, D Stein and colleagues

The Gurken signal represses *pipe* expression in the dorsal follicle cells

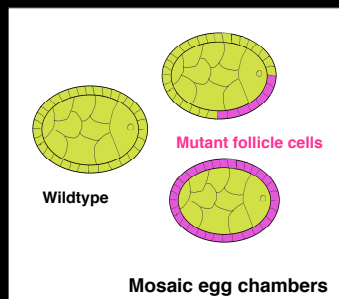


Sen et al. Cell, 95, 1998

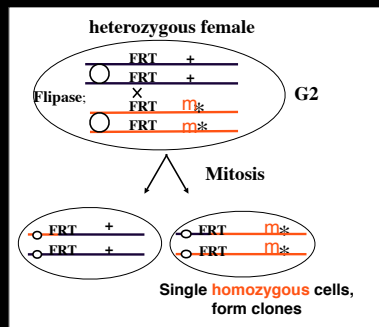
Is the repression of *pipe* by Egfr activity direct or indirect?



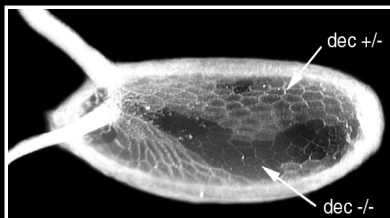
Genetic screen for mutations acting in the follicle cells, downstream of EGFr activation



Using mitotic recombination to generate homozygous mutant patches (clones) of cells

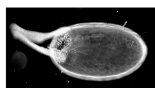
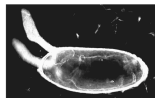


Mosaic egg shell created by mitotic recombination in follicle cells during oogenesis



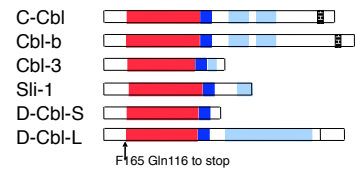
Laura Nilson

New mutations isolated in a mosaic follicle cell screen on 3L

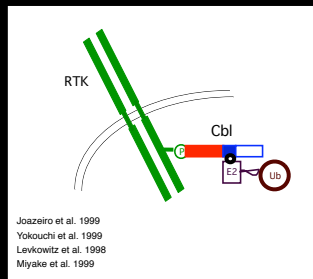
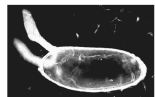


Li-Mei Pai

F165 is a mutation in the Drosophila Cbl gene



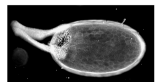
- Phosphotyrosine binding
- Ring finger/Ubiquitin-protein ligase
- Proline rich domain
- Leucine zipper

Cbl proteins downregulate activated Receptor Tyrosine Kinases, such as the EGF receptor**Cbl mutations result in dorsalized phenotypes**

F 165

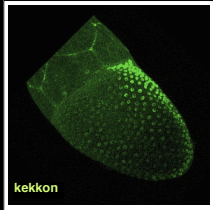


G 192



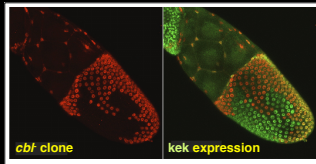
G 547

***kekkon* is an EGFr response gene**



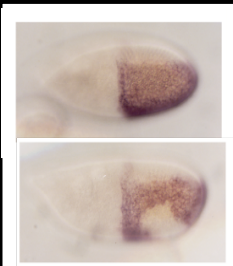
In wildtype, Kekkon is expressed on the dorsal side of the egg chamber

In *cbl* mutant cells, *kekkon* is misexpressed on the ventral side of the egg chamber



Pai, Barcelo & Schupbach, 2000

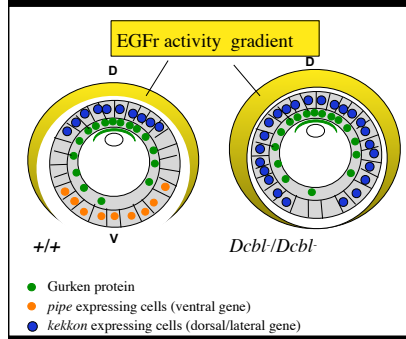
Pipe expression is repressed in *cbl* mutant clones on the ventral side of the egg chamber



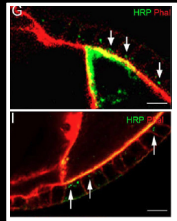
wildtype

cbl mutant clone

The direct repression of *pipe* expression by Egfr implies a wider distribution of Gurken and EGFr



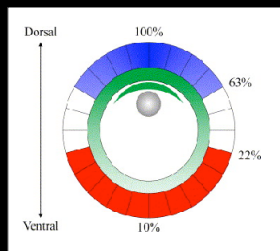
A Grk-HRP fusion protein can be detected on the ventral side of the egg chamber



Li-Mei Pai, (Chang et al.
Development, 2008)

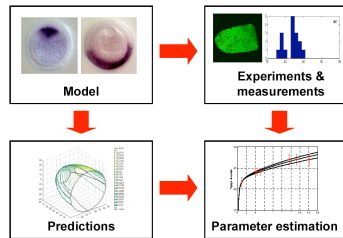
Based on these results, we already conclude that Gurken is a long-range molecule and thus supporting the proposed mechanism that Gurken signaling directly establishes the dorsal-ventral axis of the egg. Currently, we are trying to quantify further these results and perform measurements to hopefully bracket the parameter values.

Quantitative analysis of the Gurken gradient



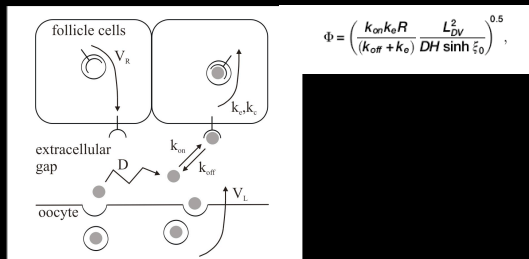
Goentoro, Reeves, Kowal, Martinelli, Schupbach, Shvartsman, Dev Cell (2006)

Inferring the shape of Gurken gradient



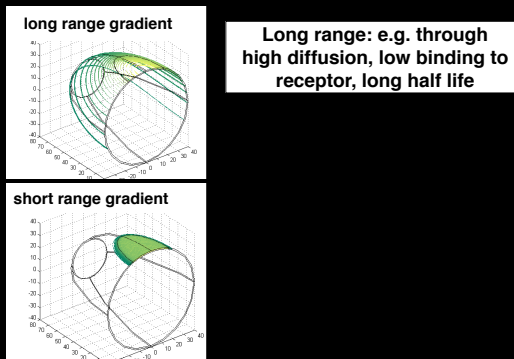
Lea Goentoro, Stanislav Shvartsman

Model of the inductive signal



None of the molecular and cellular parameters are known

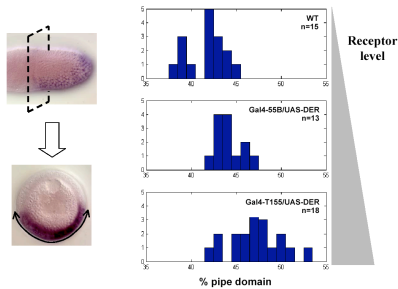
Spatial distribution of secreted Gurken



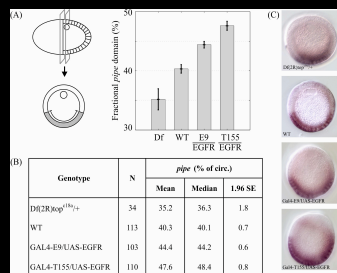
The analysis shows that,

When Da is low, that is when reaction is slow compared to diffusion, secreted Gurken can travel far from the source of secretion.

When Da is relatively high, and in this case when reaction is comparable to diffusion, most Gurken molecules secreted from dorsal side are captured immediately by nearby receptors.

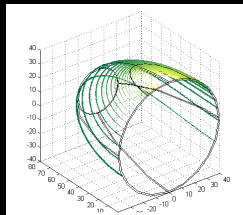
Measuring expansion in *pipe* domain

Testing the model



Goentoro, Reeves, Kowal, Martinelli, Schupbach, Shvartsman, Dev Cell (2006)

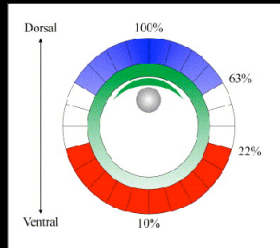
Conclusion



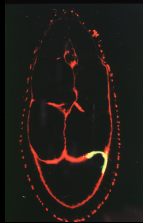
Gurken is a long-range signal

Based on these results, we already conclude that Gurken is a long-range molecule and thus supporting the proposed mechanism that Gurken signaling directly establishes the dorsal-ventral axis of the egg. Currently, we are trying to quantify further these results and perform measurements to hopefully bracket the parameter values.

Quantitative analysis of the Gurken gradient



Goentoro, et al. (2006)



RNA Localization

*Angela Jaramillo
*Jennifer Goodrich
*Nicole Clouse
*Amanda Norvell
Scott Ferguson

Translational Control, *encore, half pint*

*Cheryl Van Buskirk

Mathematical Modelling

*Lea Goentoro

Collaborators:

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E. Gavis, Princeton U.

Gurken-Egfr signal

*Shira Neuman-Silberberg
*Siegfried Roth
*Jim Price

Follicle Cell Response

Yan Yan
Natalie Deneff
Yi Sun
*Li-Mei Pai
*Laura Nilson

Technical help

Gail Barcelo

* former lab members