Precision and reproducibility of the *Drosophila* segmentation gene network under Bicoid dosage perturbations

Feng Liu[1], Alexander H. Morrison[2], and Thomas Gregor[3]

***Short Abstract* — We have developed a quantitative experimental approach to investigate how the early *Drosophila* segmentation gene network responds to Bicoid (Bcd) dosage changes. Using a fly line library of constructs expressing Bcd-GFP with varying dosages, we measured *in vivo* Bcd-GFP gradients and the positions of several representative segmentation patterning markers. We show that the reproducible generation of the Bcd gradients does not involve feedback regulation on Bcd protein expression. Additionally, a fixed threshold model cannot achieve the precision of the patterning markers. Instead, only a mechanism reliant on either additional maternal inputs or on a non-local read-out of the Bcd concentration would be in agreement with our data.**

***Keywords* — segmentation gene network, precision, reproducibility, embryogenesis, *Drosophila*, Bicoid.**

I. Introduction

During *Drosophila* embryogenesis, the morphogen Bicoid (Bcd) forms a gradient and directs patterning along the anterior-posterior axis via a hierarchical segmentation gene network in a concentration dependent manner[1, 2]. Previous studies have indicated that the high degree of precision and reproducibility of the segmentation patterning is established by reproducible control of the Bcd gradient concentration and the precise response of the downstream target genes [3, 4]. However, it is still unclear what the underlying mechanism for reproducible generation of Bcd gradients is and how the precise response of the segmentation gene network is achieved [5, 6]. The most basic model is that there is negative feedback regulation on the Bcd protein expression and the response of the network is a local read-out of the Bcd concentration at a fixed threshold [7]. To test this model, we have developed a quantitative approach to investigate how the network responds to controlled perturbations of the Bcd dosage.

II. Results

We generated a set of fly lines expressing Bcd-GFP transgenes with Bcd dosages ranging from half to triple the wild type dosage in increments as small as 10%. *In vivo* Bcd-GFP gradients and the positions of several representative downstream segmentation patterning markers were measured, including boundaries of profiles of *hunchback* mRNA and protein, peaks of the even-skipped protein profiles and *in vivo* cephalic furrow locations. We demonstrate an absolute linear relationship between the nuclear Bcd-GFP protein concentration and the *Bcd-GFP* DNA insertions, suggesting that the reproducibility of the Bcd gradient is unlikely to involve feedback regulations. Upon Bcd dosage perturbation, the average shift of the segmentation patterning markers is only about half of the amount predicted from a fixed threshold model. Furthermore, given the measured Bcd gradient noise, the positional variances of the segmentation markers are also only about half of the predicted amounts based on a fixed threshold model. Finally, the dependence of the Bcd gradient on embryo length is not sufficient to generate the scaling of the segmentation markers. All of these findings indicate that the absolute Bcd concentration of a given nucleus is not enough to determine a unique cell fate, and that the cell's interpretation of that concentration is dependent on a more global context.

III. Conclusion

We conclude that of the many possible mechanisms for generating precise and reproducible Bcd-dependent read-out markers, the only ones that are viable with our results must rely on either additional maternal inputs or on a non-local read-out of the Bcd concentration. A quantitative model for the latter is presented.

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[1]Physics Department, Princeton University, Princeton, NJ, 08544, E-mail: fl2@princeton.edu

[2]Department of Molecular biology, Princeton University, Princeton, NJ, 08544. E-mail: ahmorris@princeton.edu

[3]Physics Department, and Lewis-Sigler Institute for Integrative Genomics, Princeton University, Princeton, NJ, 08544. E-mail: tg2@princeton.edu

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