

Drosophila melanogaster:

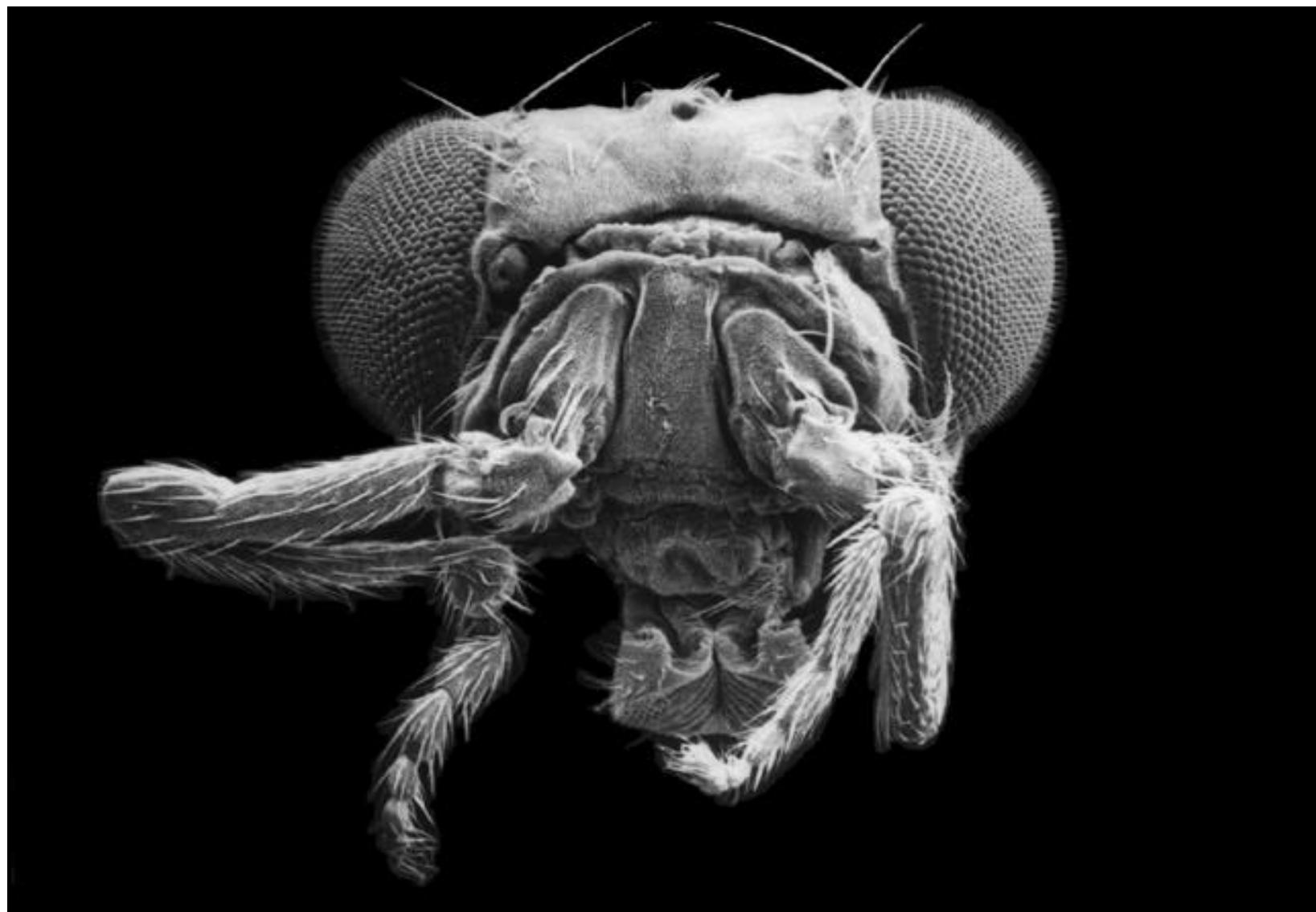
pattern formation

2019

Patrick Callaerts, Ph.D.

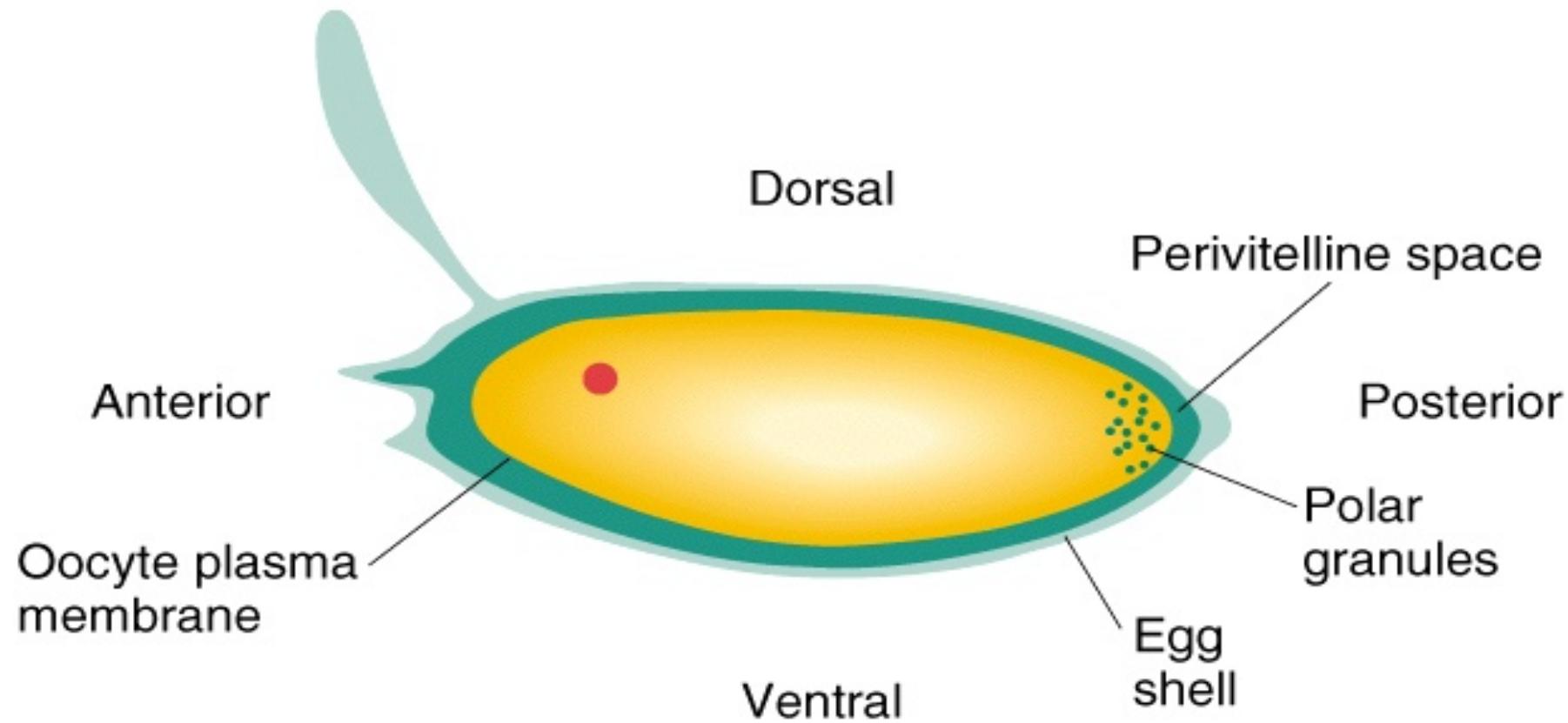


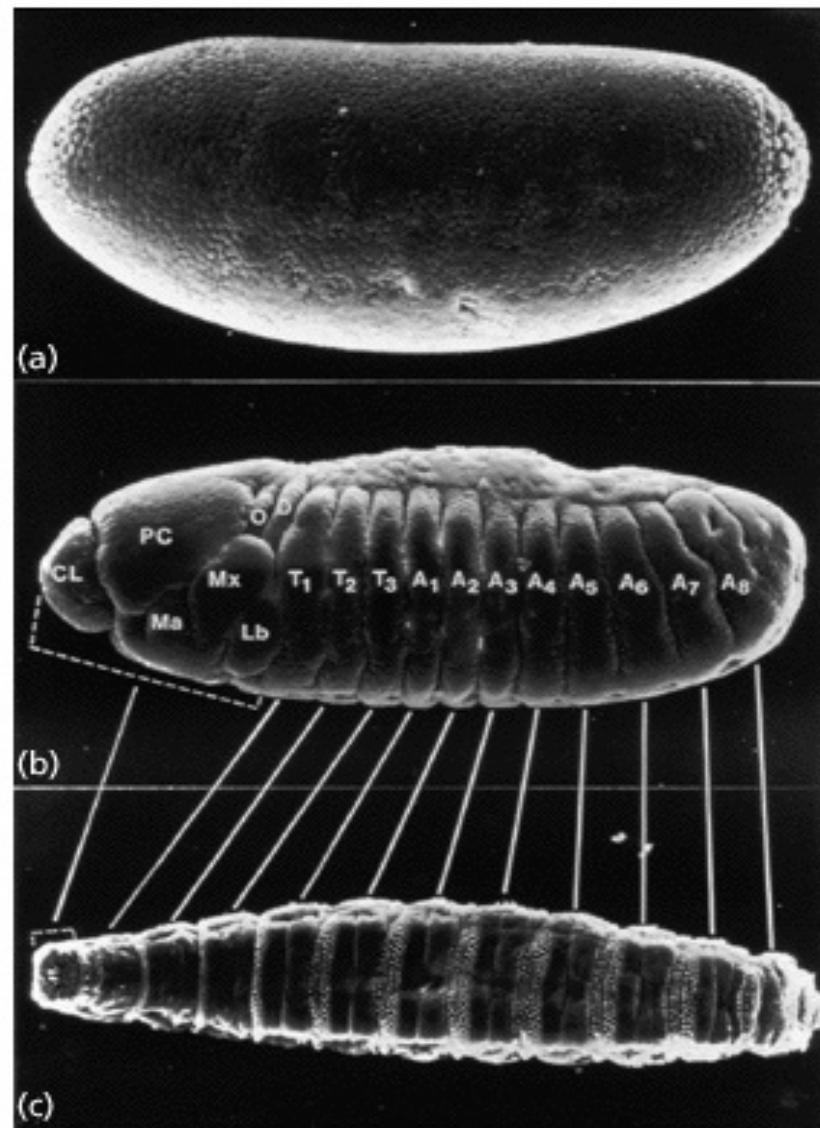




Overview

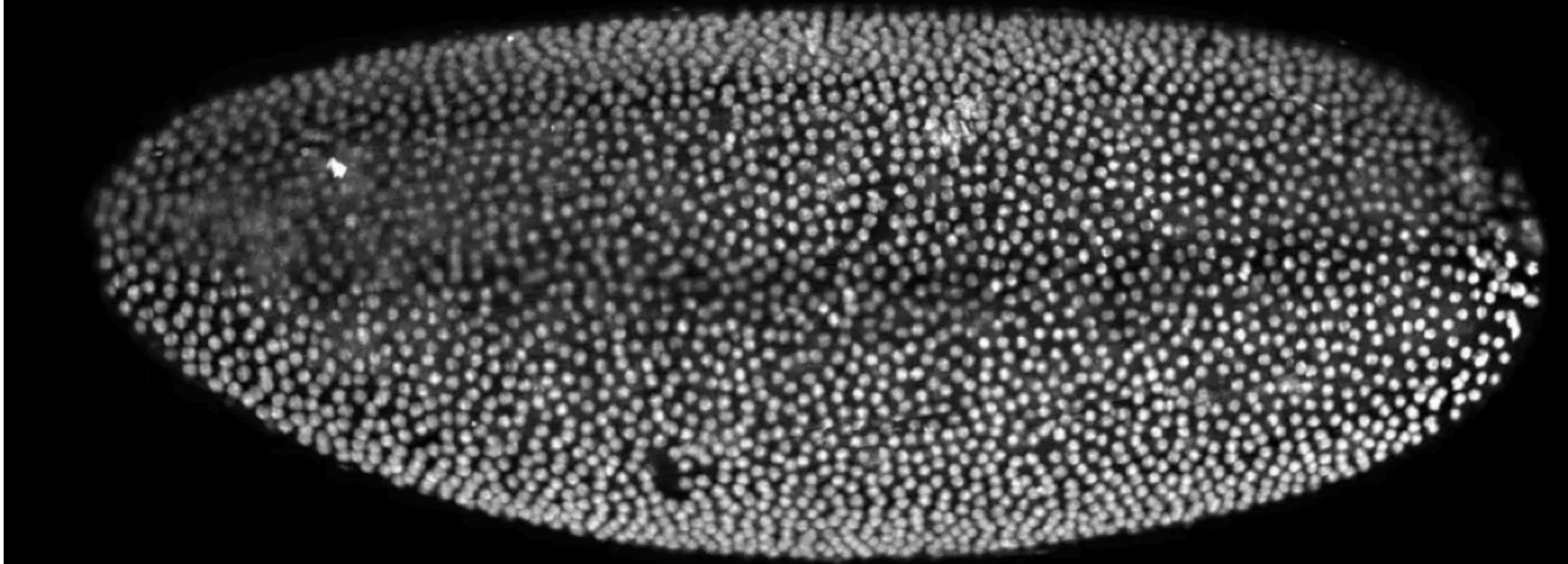
- Maternal gene gradients
- Segmentation genes - converting gradients into stripes
- Homeotic genes





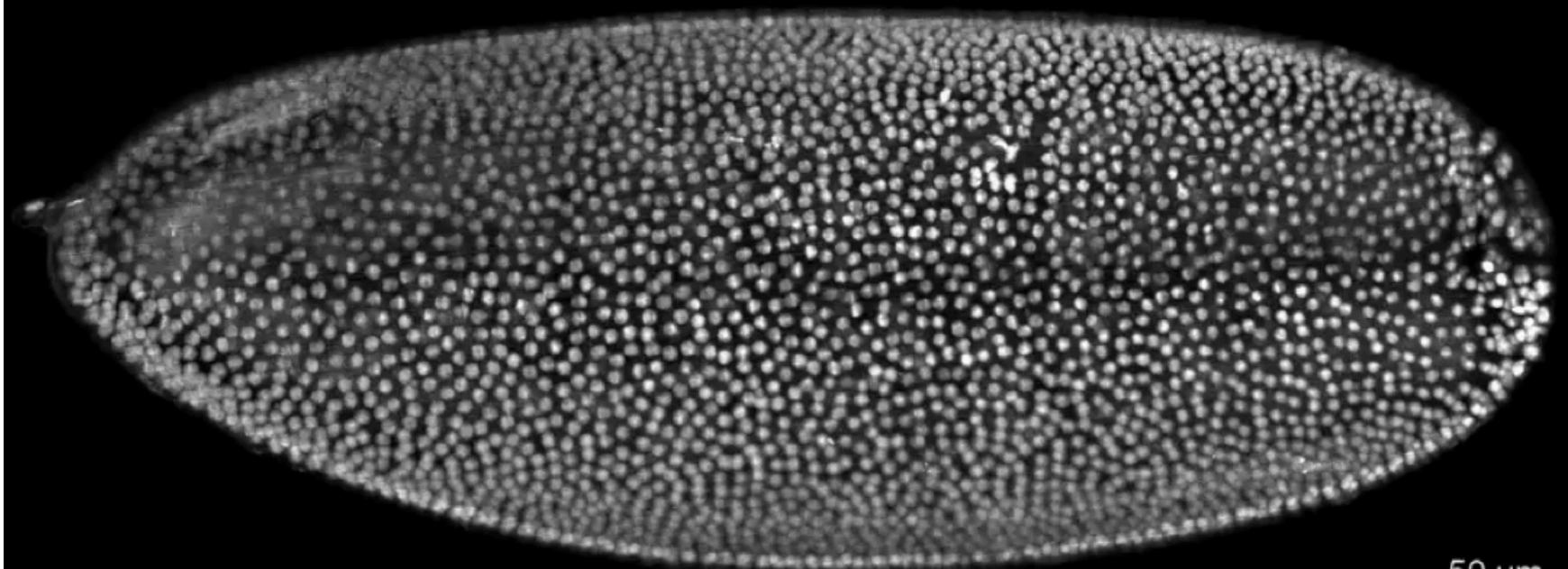
ventrolateral view

02:00:00

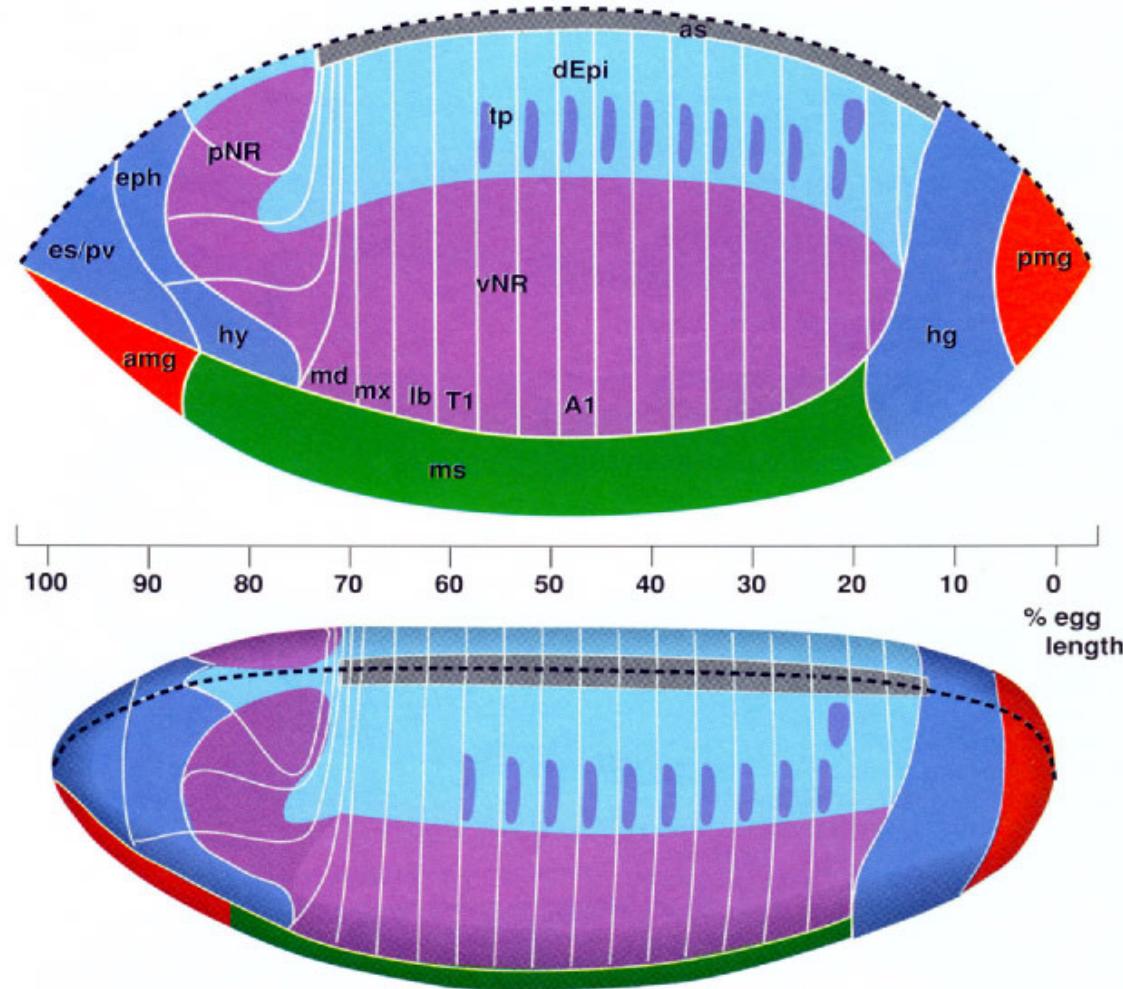


dorsolateral view

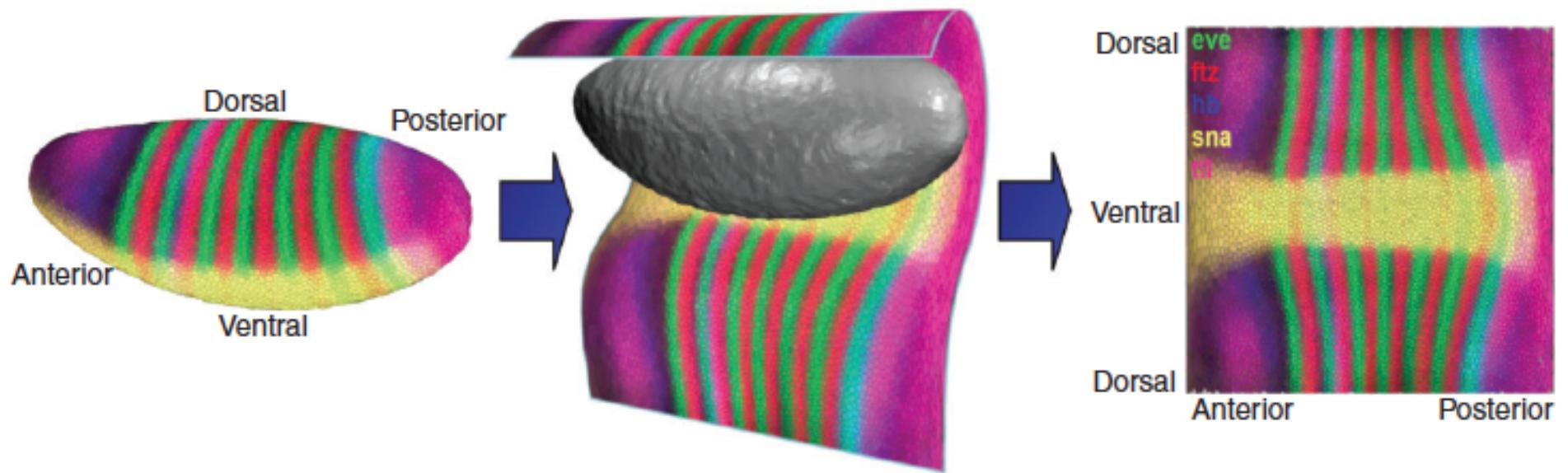
50 μm



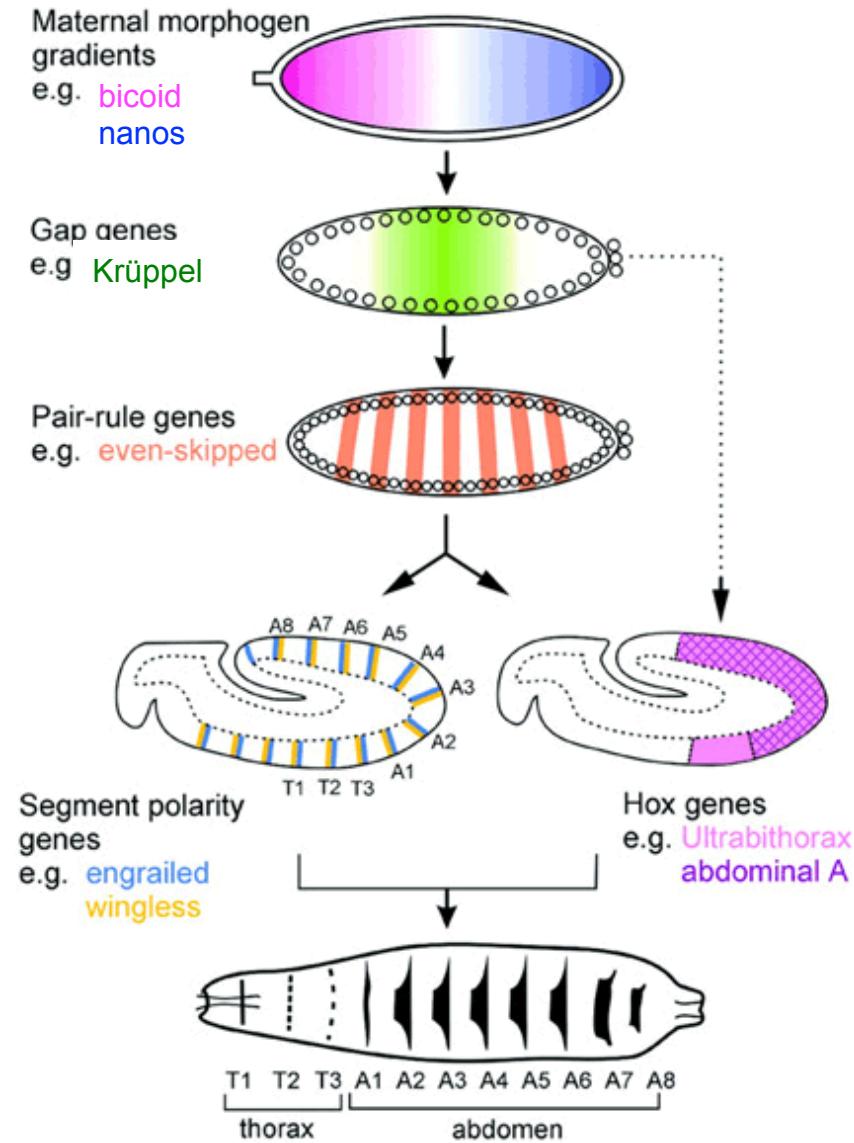
Blastoderm: fate maps



Blastoderm: the molecular basis of fatemaps



Patterning the embryo

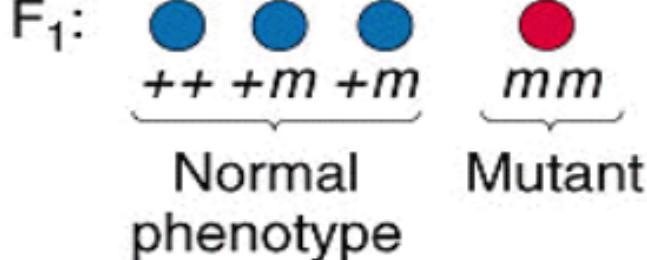


Maternal effect genes

(a) Zygotically acting genes

The F₁ phenotype due to zygotic genes is determined solely by the F₁ genotype.

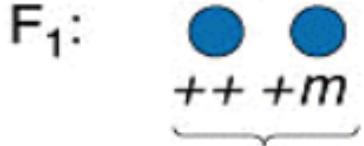
$$P: +m\text{♀} \times +m\text{♂}$$



(b) Maternal-effect genes

The F₁ phenotype due to maternal-effect genes is determined solely by the phenotype of P maternal parent.

$$P: +m\text{♀} \times ++\text{♂}$$



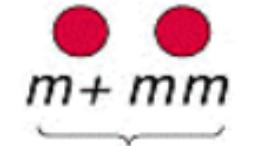
$$+m\text{♀} \times mm\text{♂}$$

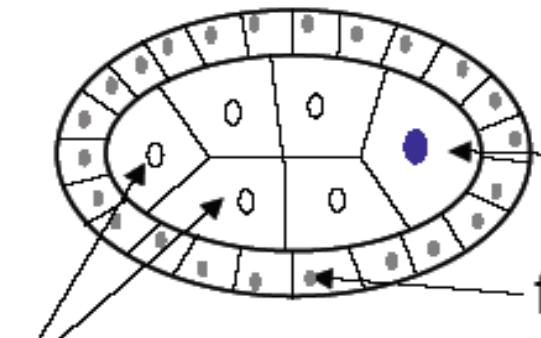


$$mm\text{♀} \times ++\text{♂}$$



$$mm\text{♀} \times m+\text{♂}$$

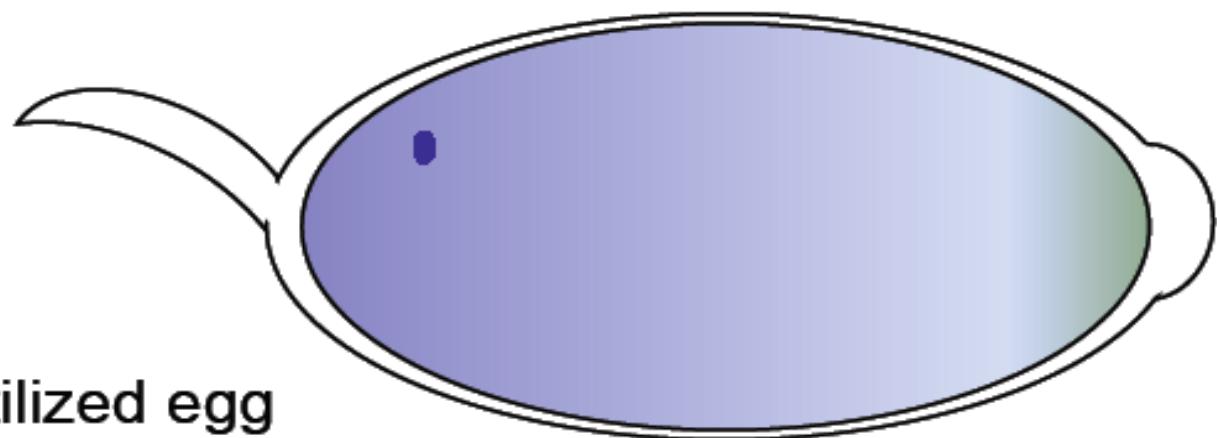
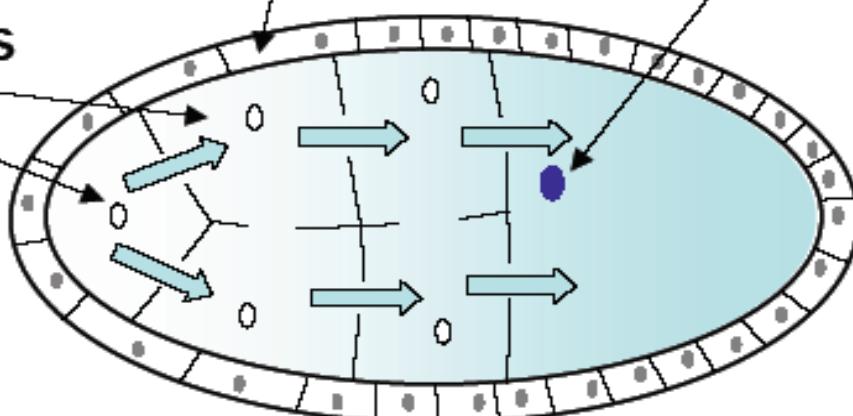




nurse cells

follicle cells

oocyte nucleus



laid fertilized egg

Patterning the embryo

Four Independent Genetic Regulatory Systems Specify the Anteroposterior and Dorsal-Ventral Axes

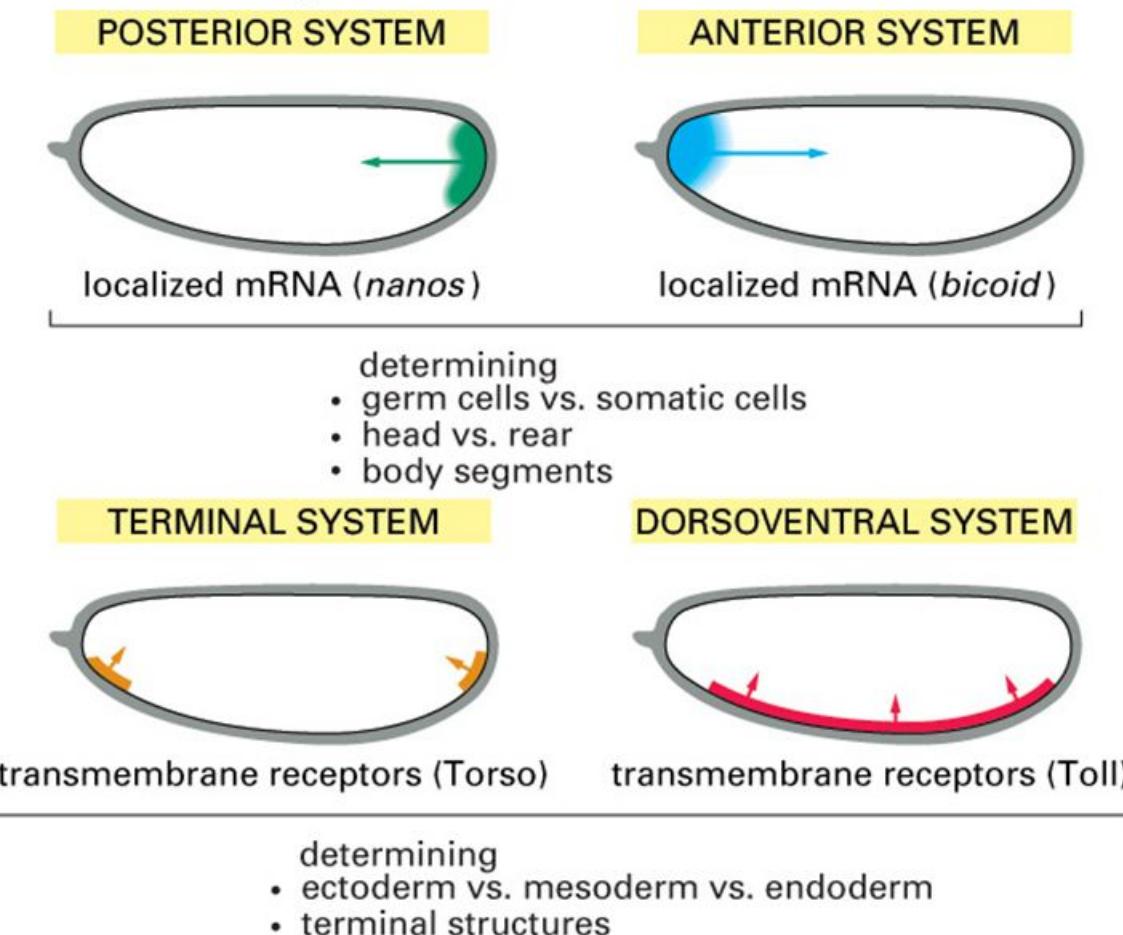
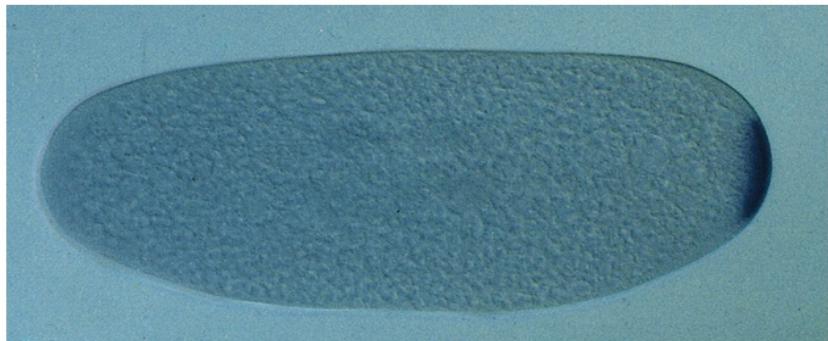
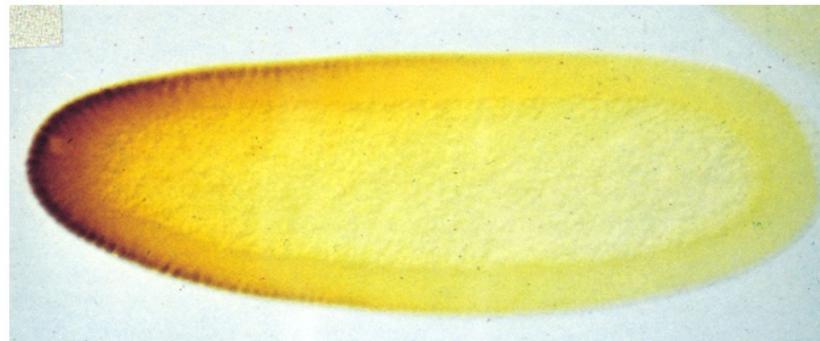


Figure 21–31. Molecular Biology of the Cell, 4th Edition.

Bicoid (Bcd)

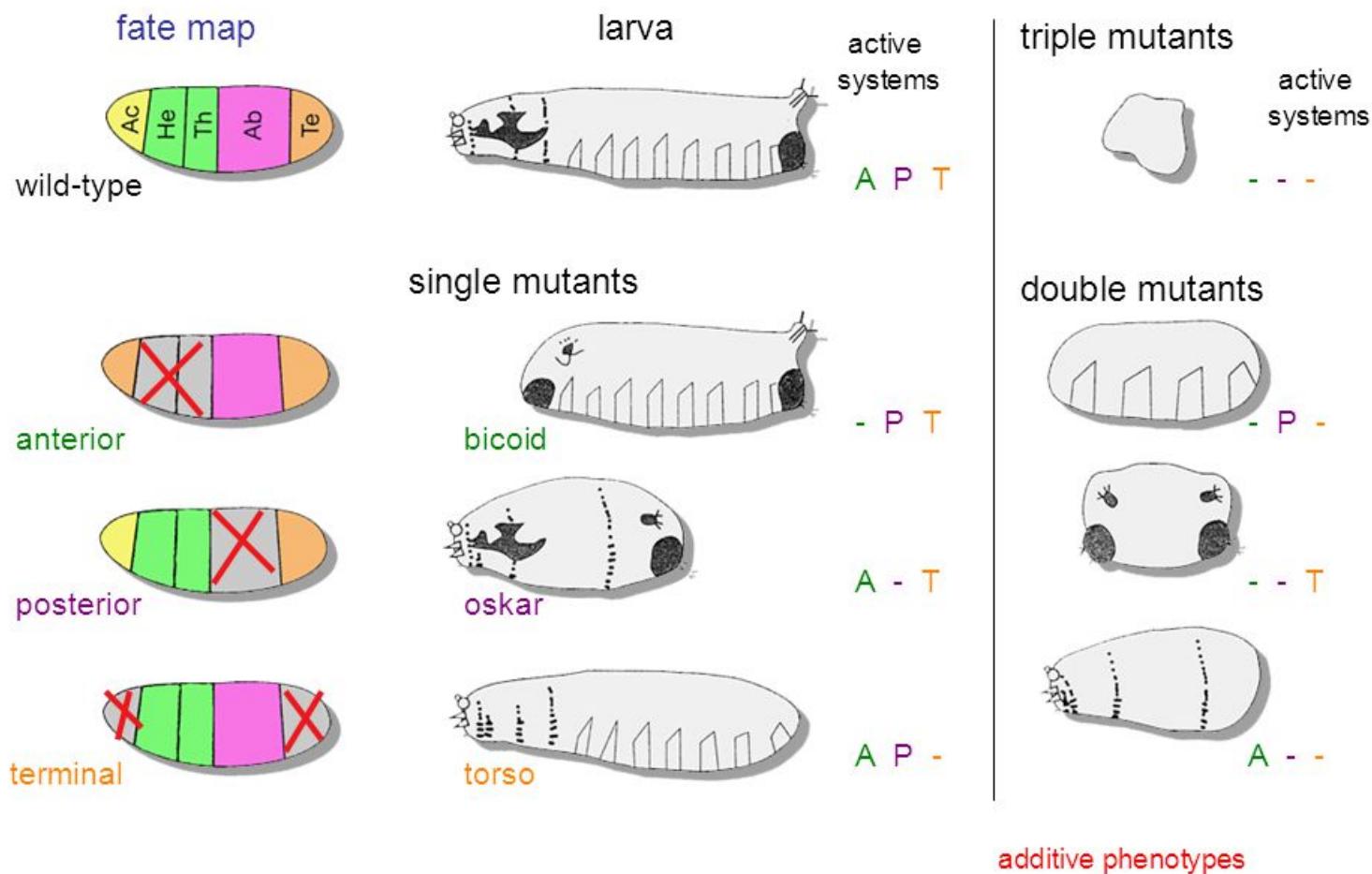


Nanos (nos)

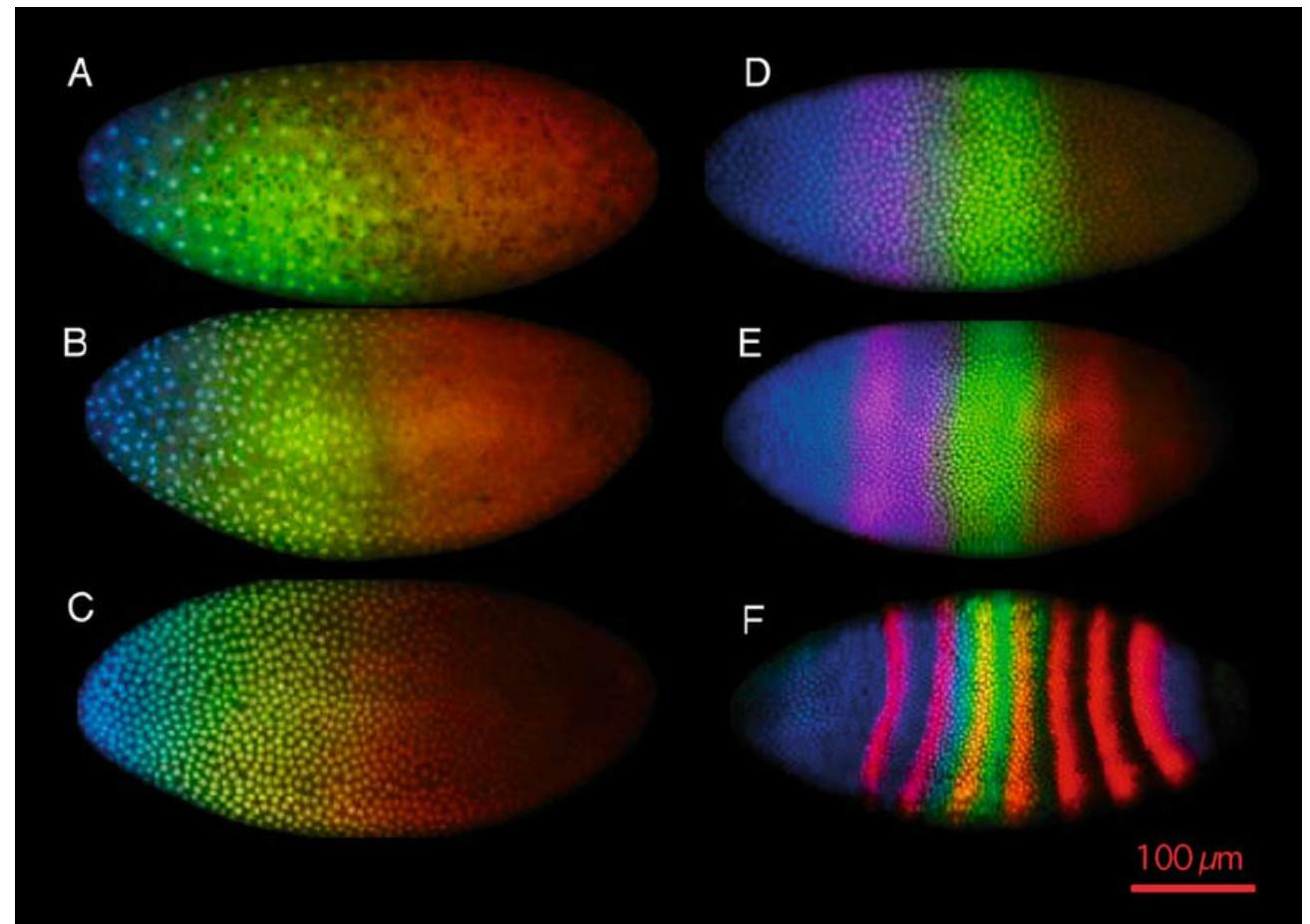
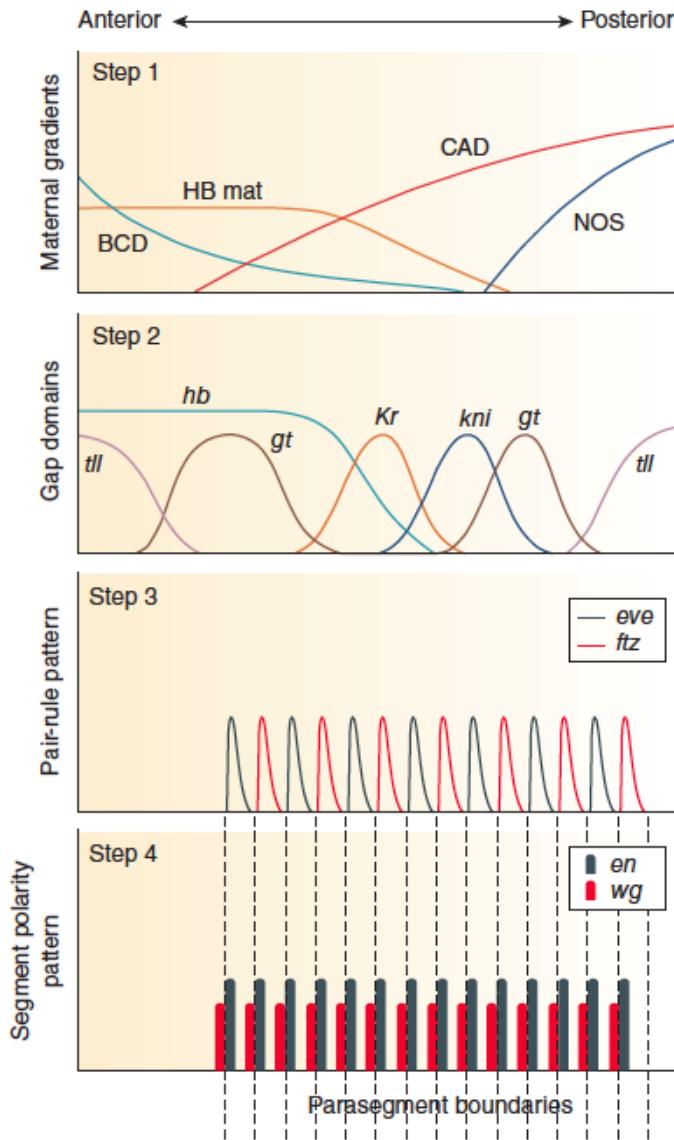
Patterning the embryo

Mutations affecting the antero-posterior axis

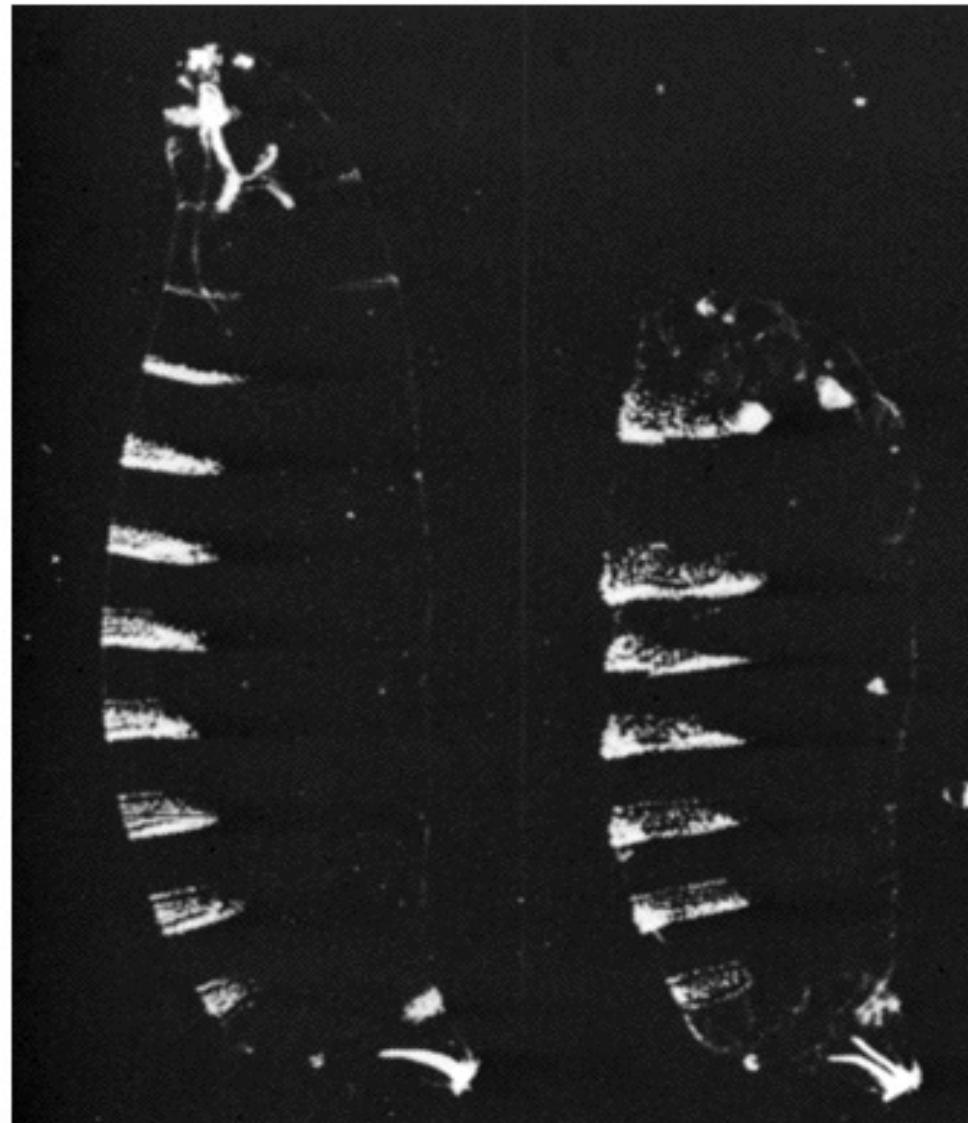
3 independent maternal systems: **anterior**, **posterior**, **terminal**



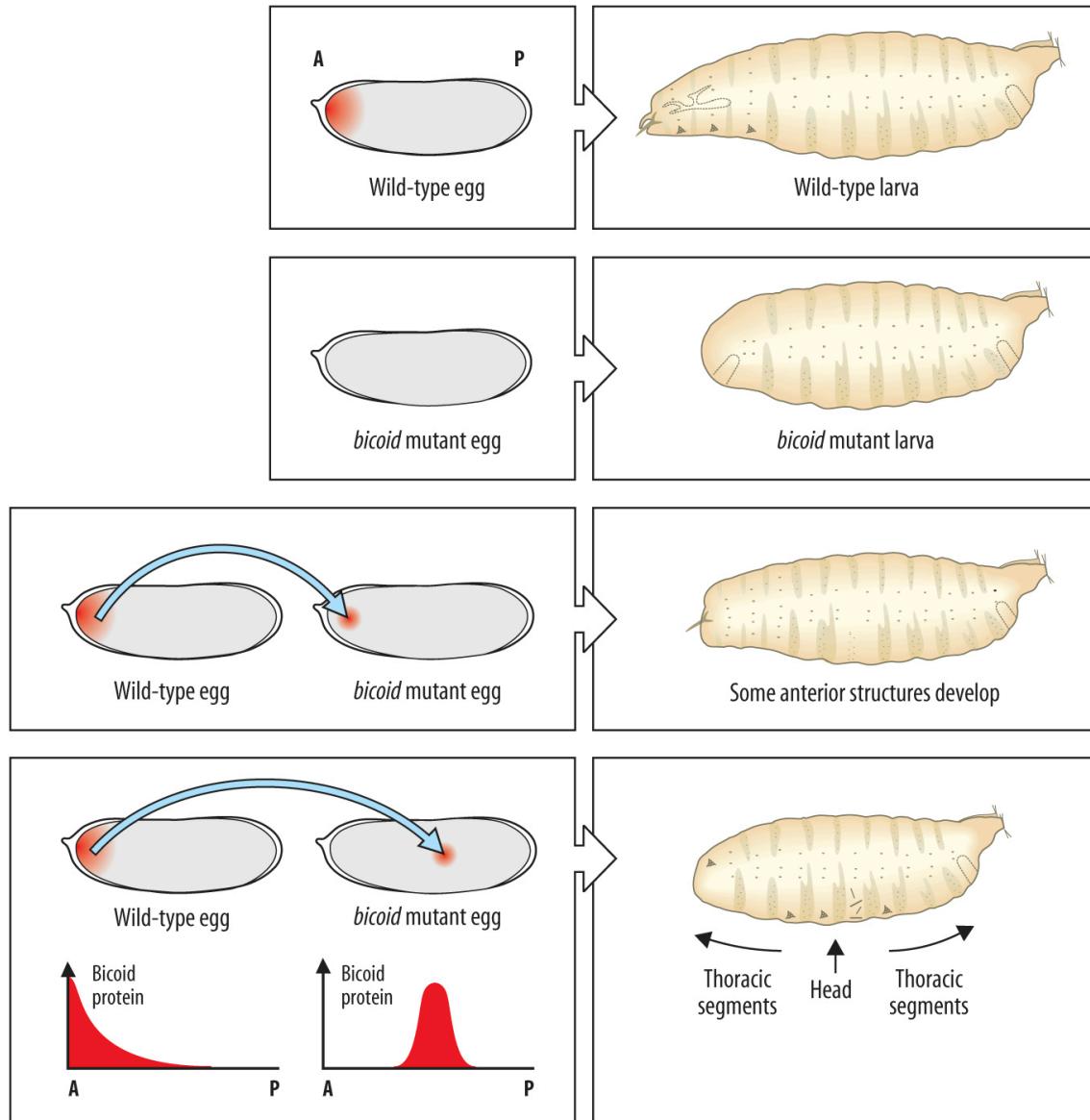
Patterning the embryo



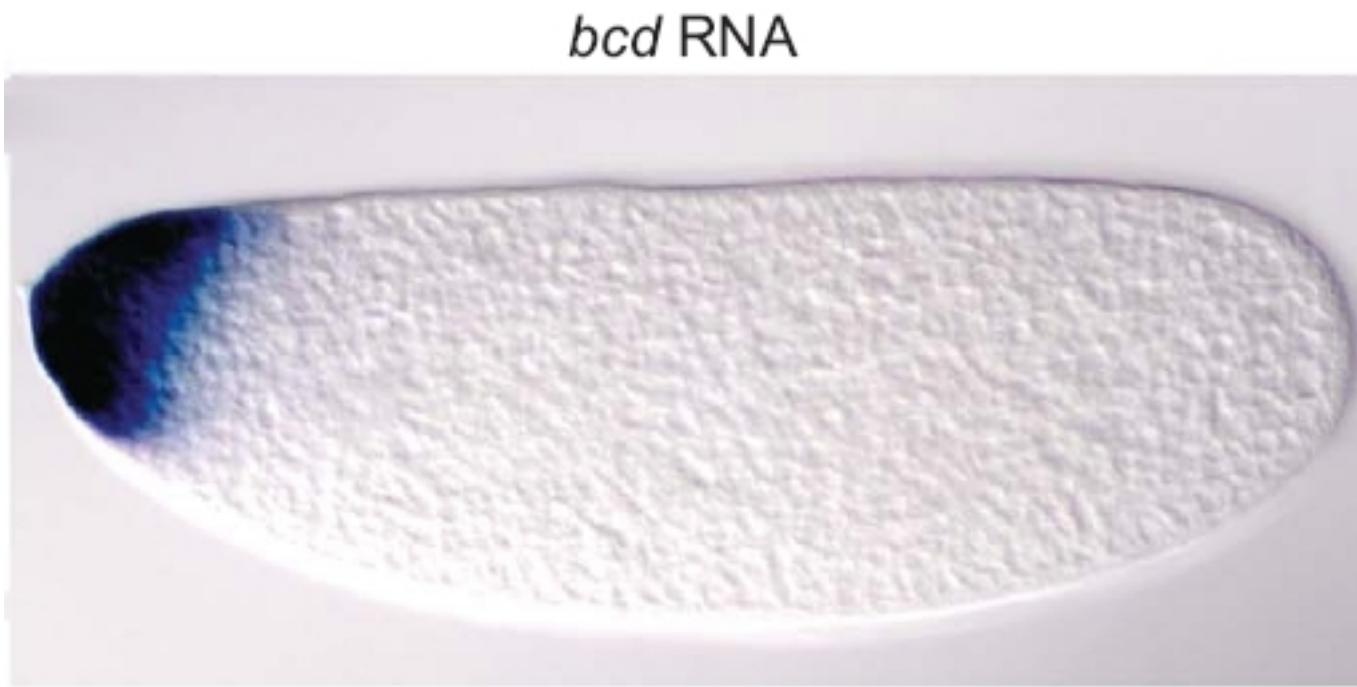
Bicoid: anterior determinant



Bicoid: anterior determinant



Bicoid: anterior determinant



How is *bcd* mRNA localized anteriorly?

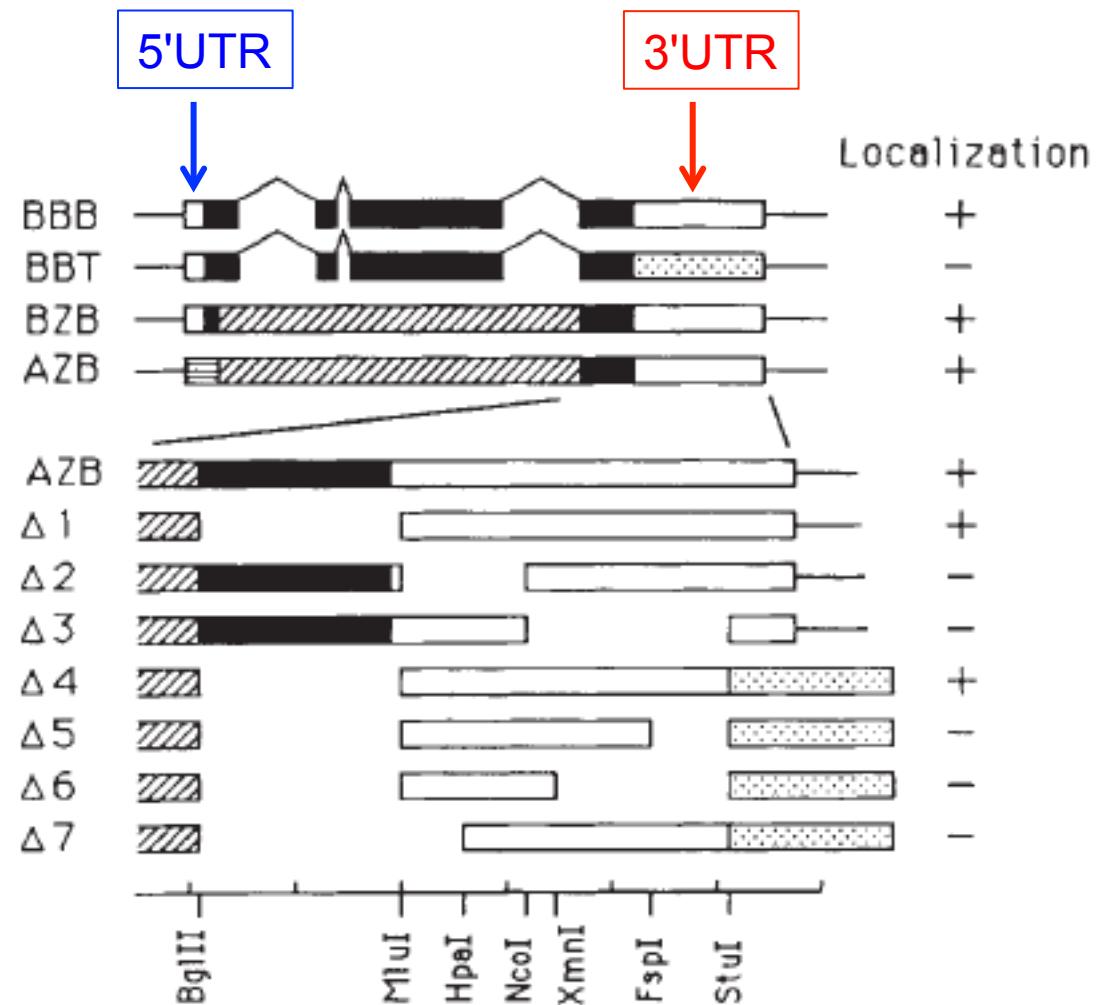
Localization depends on 3'UTR

Cis-acting sequences responsible for anterior localization of *bicoid* mRNA in *Drosophila* embryos

Paul M. Macdonald & Gary Struhl

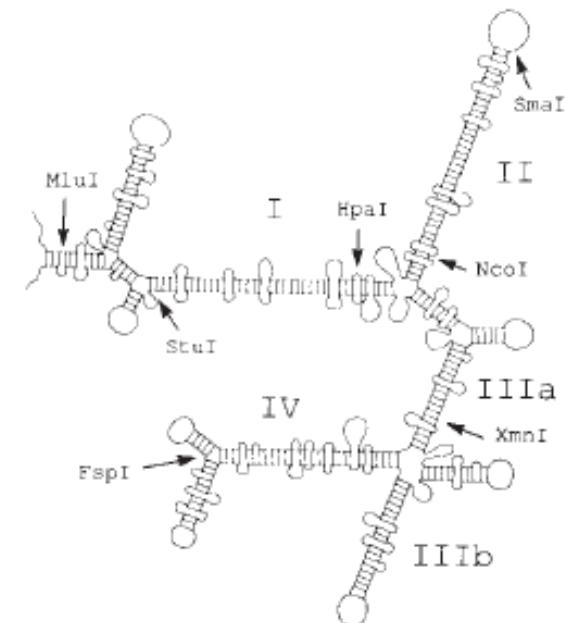
Howard Hughes Medical Institute, Center for Neurobiology and Behavior, Columbia University College of Physicians and Surgeons, 722 W. 168 St, New York, N.Y. 10032, USA

Nature 336, 595-598 (1988)



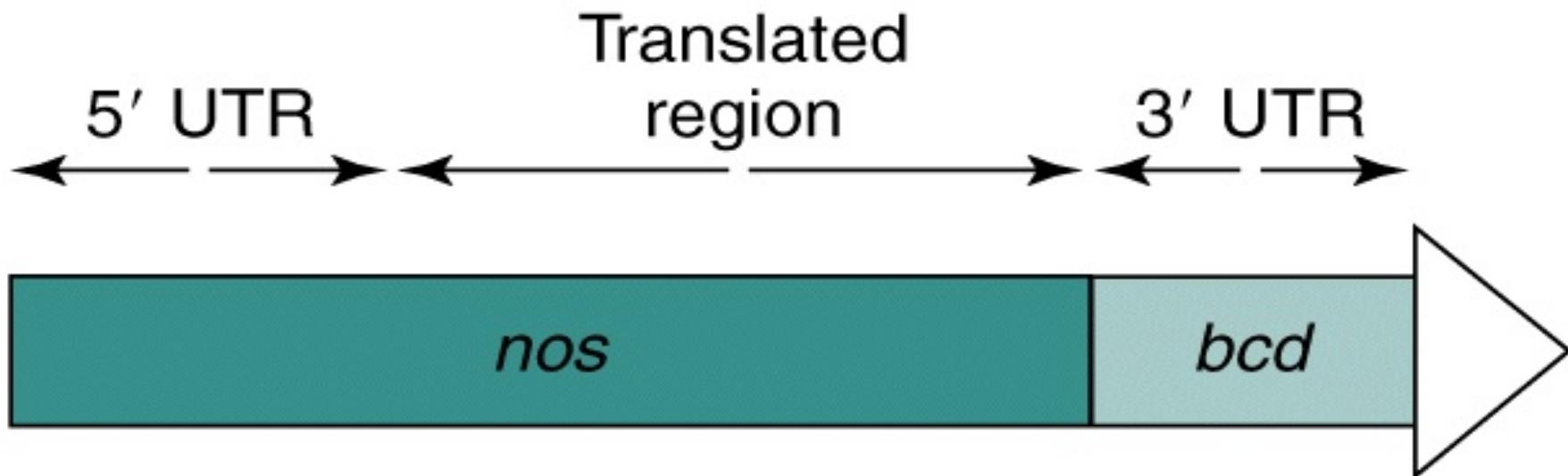
Localization depends on 3'UTR

UACGCGUAGAAAGUUAGGUUCUAGGUCCUAAGAUCCGUGUAAAUGGUUCCCAGGGAGUUUUAGUACUAGCCUAGU 75
 MluI I
 >----oo-----o-----*-----o----->-----ooooo-----*-----o-----> >---o-----o-----
 CAGCAGGCCGACGGAUUCCAGUGCAUAUCUUAGUGAUACUCCAAGGUAAACCUCUAACUUUCCCUGCAAACGCUA 150
 II HpaI II
 **o-----*-----o-----o-----*o--> <--o*-----o-----o-----*-----*-----ooooo
 UUCGCCUUAGAUGUAUCUGGGUGGCUGCUCCACUAAAGCCCGGGAAUGCAACCAGGUACAUUUAGGCCAUU 225
 SmaI IIIa
 o**oooo-----o-----< >-----ooo-----*
 GGGCUUAAGCGUAUUCCAUGGAAAGUUAUCGUCCCACAUUUCGAAAUUUCGGAGCCAGCAAGAAAUCCU 300
 NcoI IIIb XmnI
 o---> >-----*o-----*----->
 CUCUGUUACAAUUGACAUAGCUAAAAACUGUACUAUCAAAAUGAAAAAAUGUUUCUCUUAGGCGUAUUCUA 375
 IIIb IV
 <-----*ooo-----o*-----<>-----ooo-----o-----o-----ooo-----oo*--->
 CAAUGAUUACCCUAAAAGAUCGAACAUUUAAACAAUAAUUGAUAAUUGAUUUUCAAUUUCAUUUCAUUUCAU 450
 FspI IIIa FspI I
 oooooo-----<<----ooo*-----o-----< <---oooooo-----oo*---ooo---
 AGACCAUUUGUUUCAGAUUCGGAGAUUUUUGCUGCCAAACGGAAUACUAUUAGCUUUACUUUACAU 525
 StuI
 -----ooo-----*-----*-----o-----<
 CACUAAGAAGAGCAUUGCAAUCGUUAGGCCUCAAGUUUAAUUAAAAUGCUGCACCUU 660

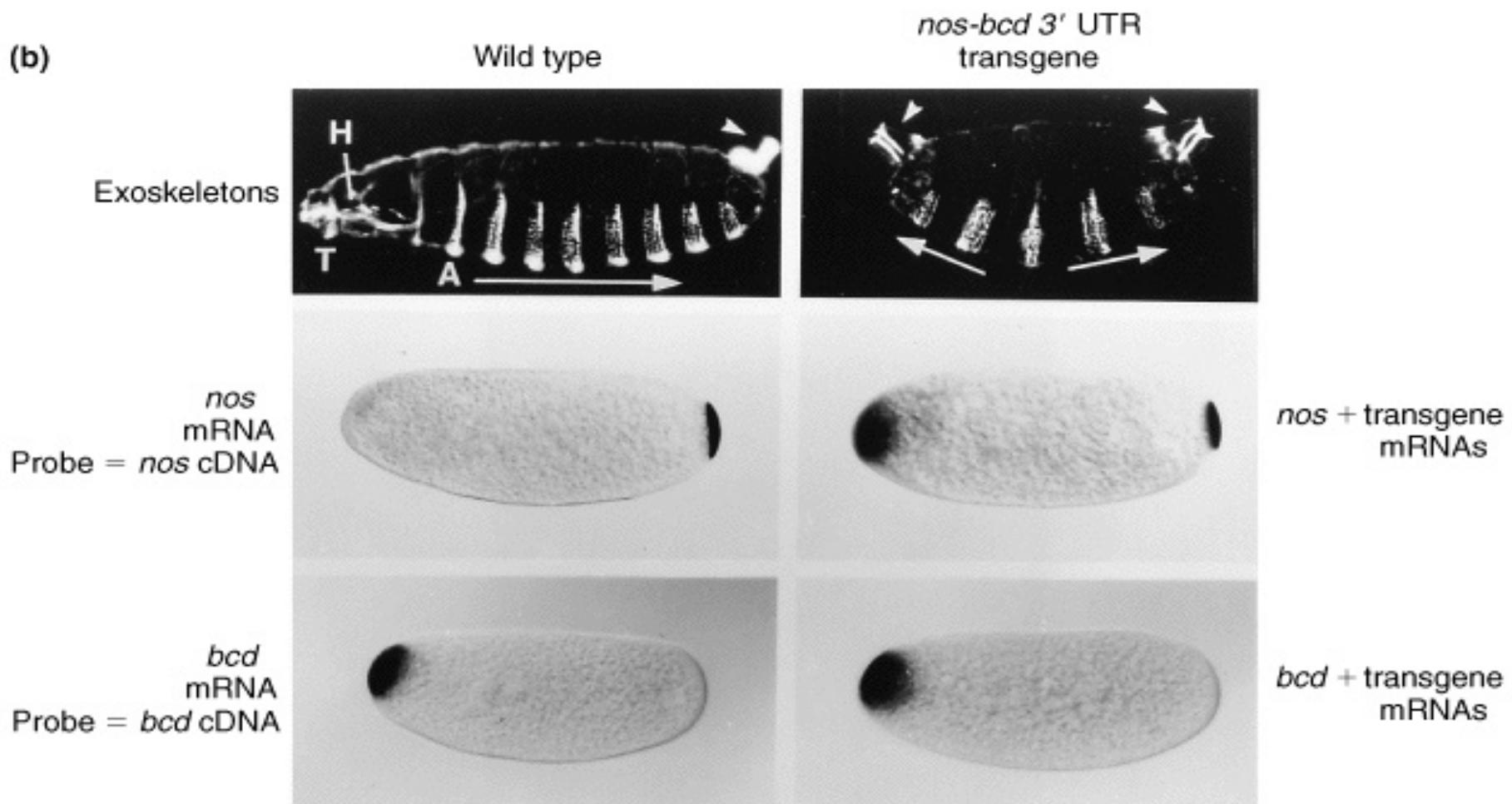


Localization depends on 3'UTR

(a)

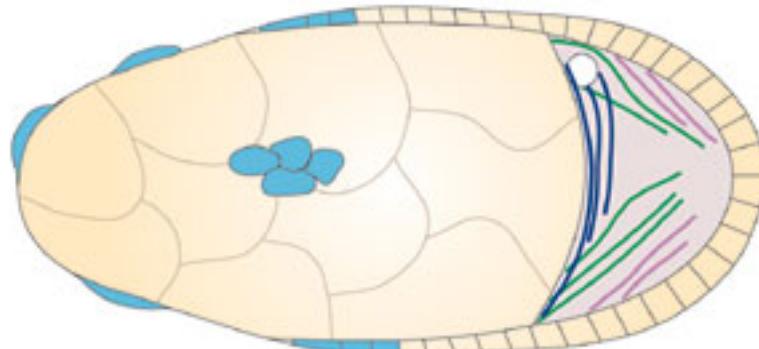


Localization depends on 3'UTR

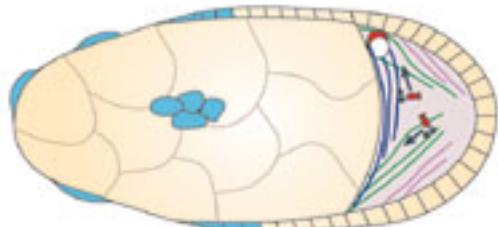


Bicoid: localization of mRNA

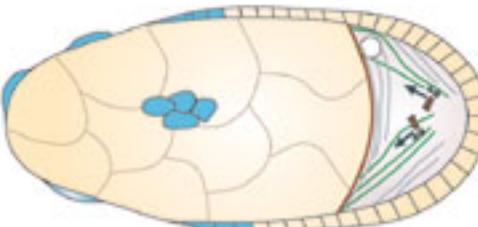
a Microtubule populations in the *Drosophila melanogaster* oocyte



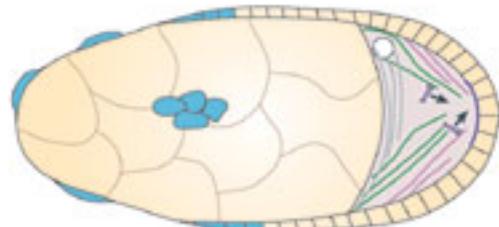
b *gurken* mRNA



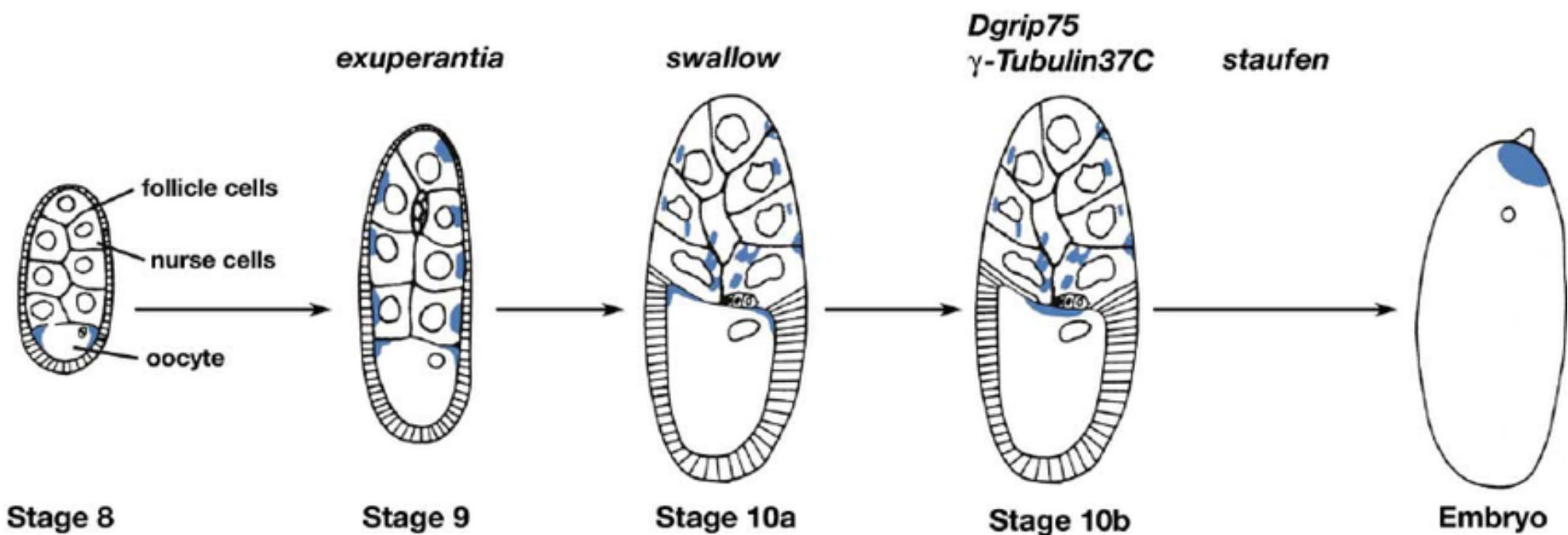
c *bicoid* mRNA



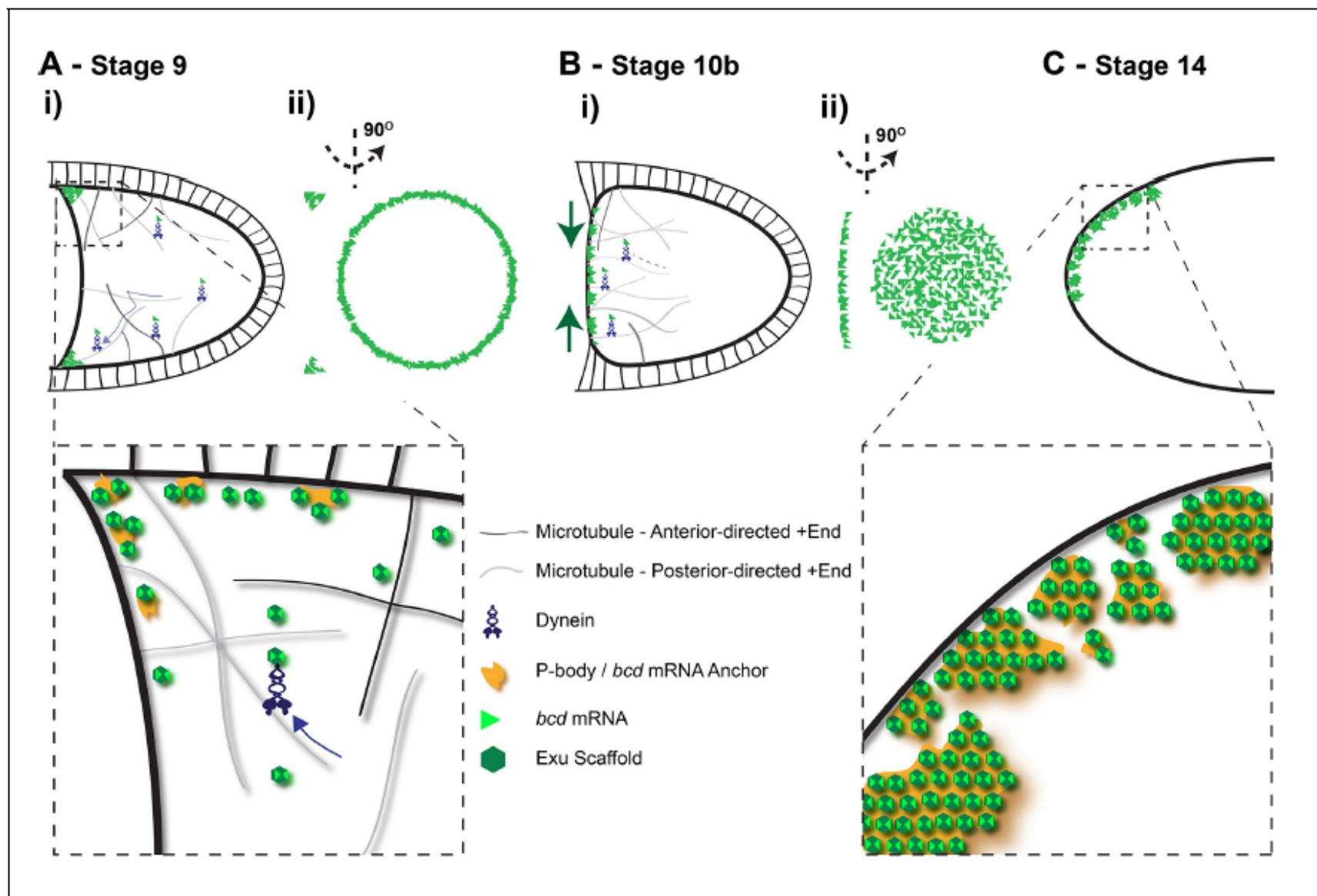
d *oskar* mRNA



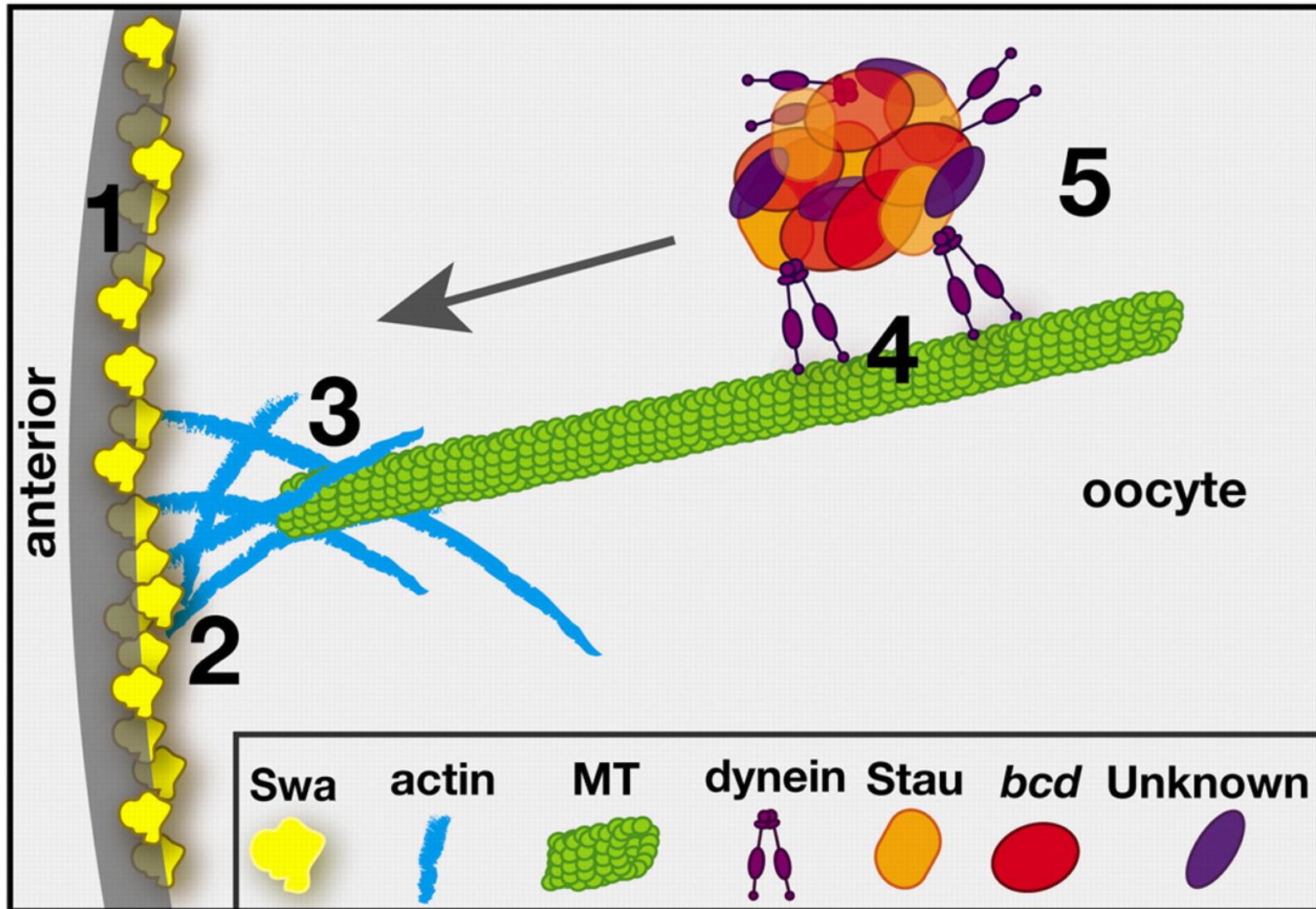
Bicoid: localization of mRNA



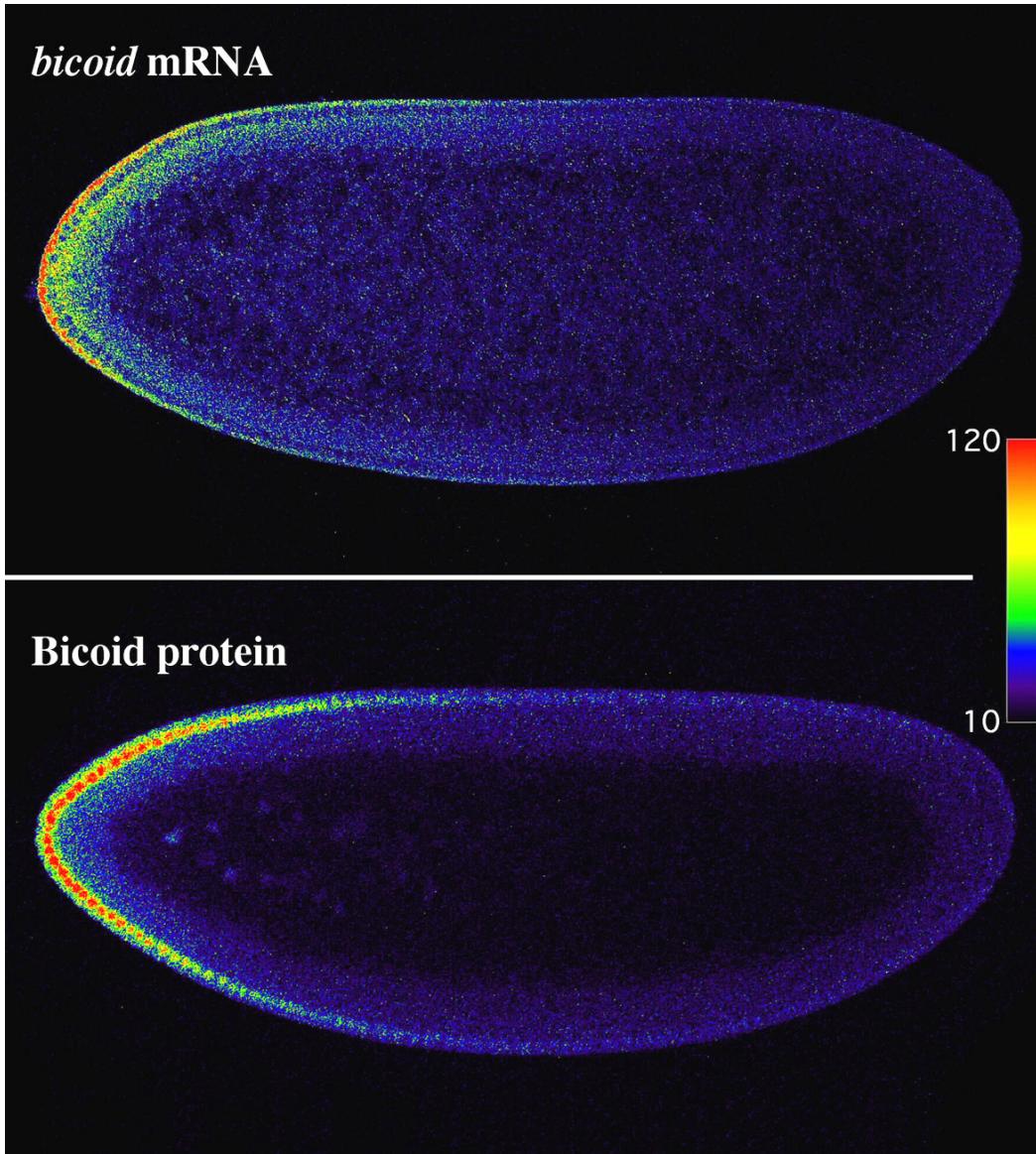
Bicoid: localization of mRNA



Bicoid: localization of mRNA

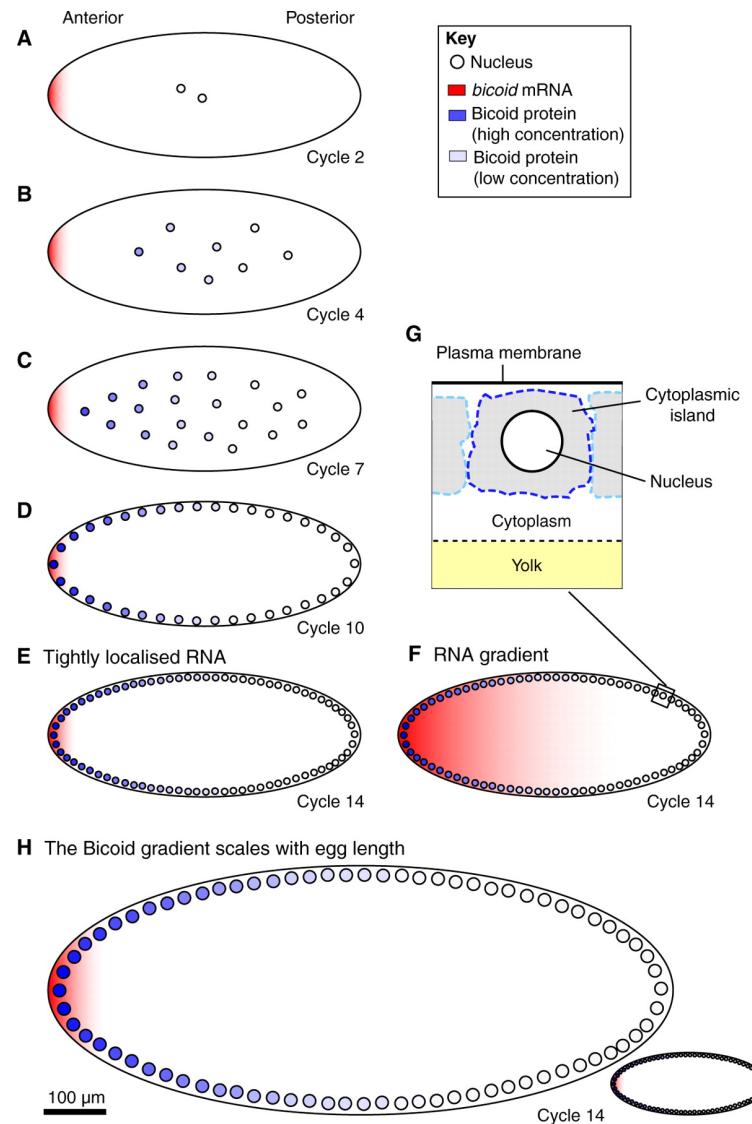


Bicoid: anterior morphogen gradient



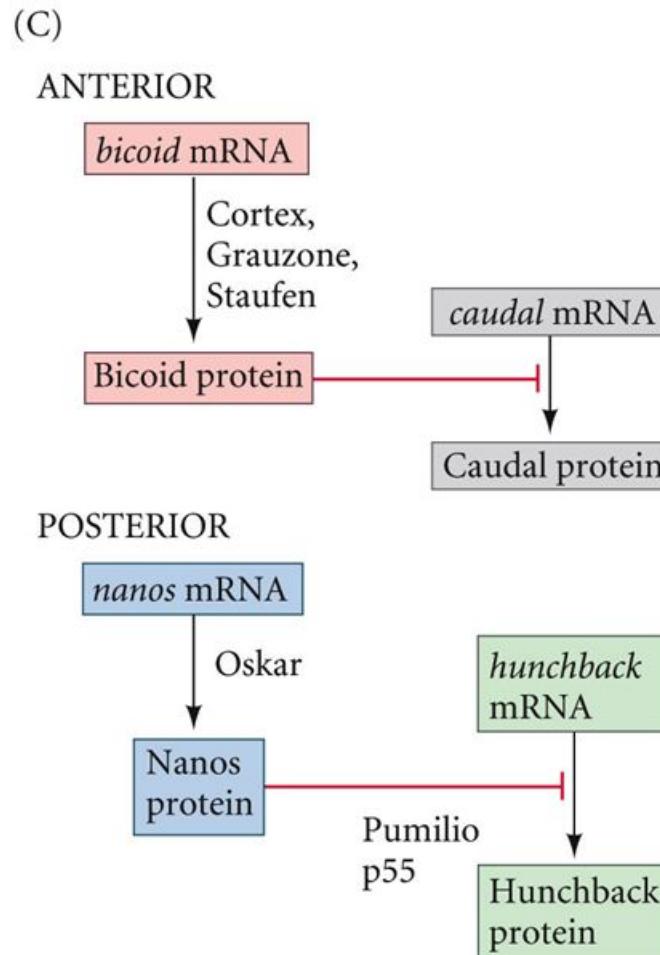
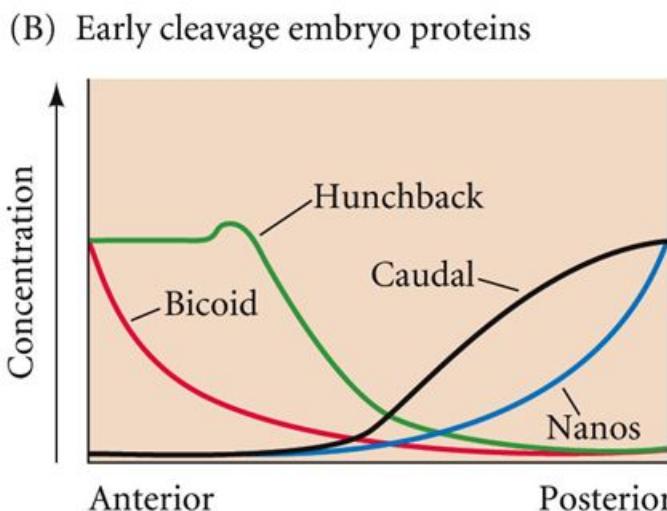
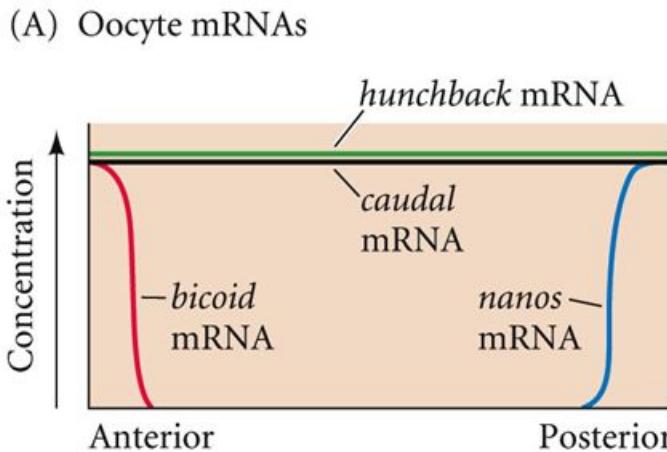
How is the Bcd protein gradient established?

Bicoid: anterior morphogen gradient



Maternal genes: more than one gradient

Model of anterior-posterior pattern generation by *Drosophila* maternal effect genes



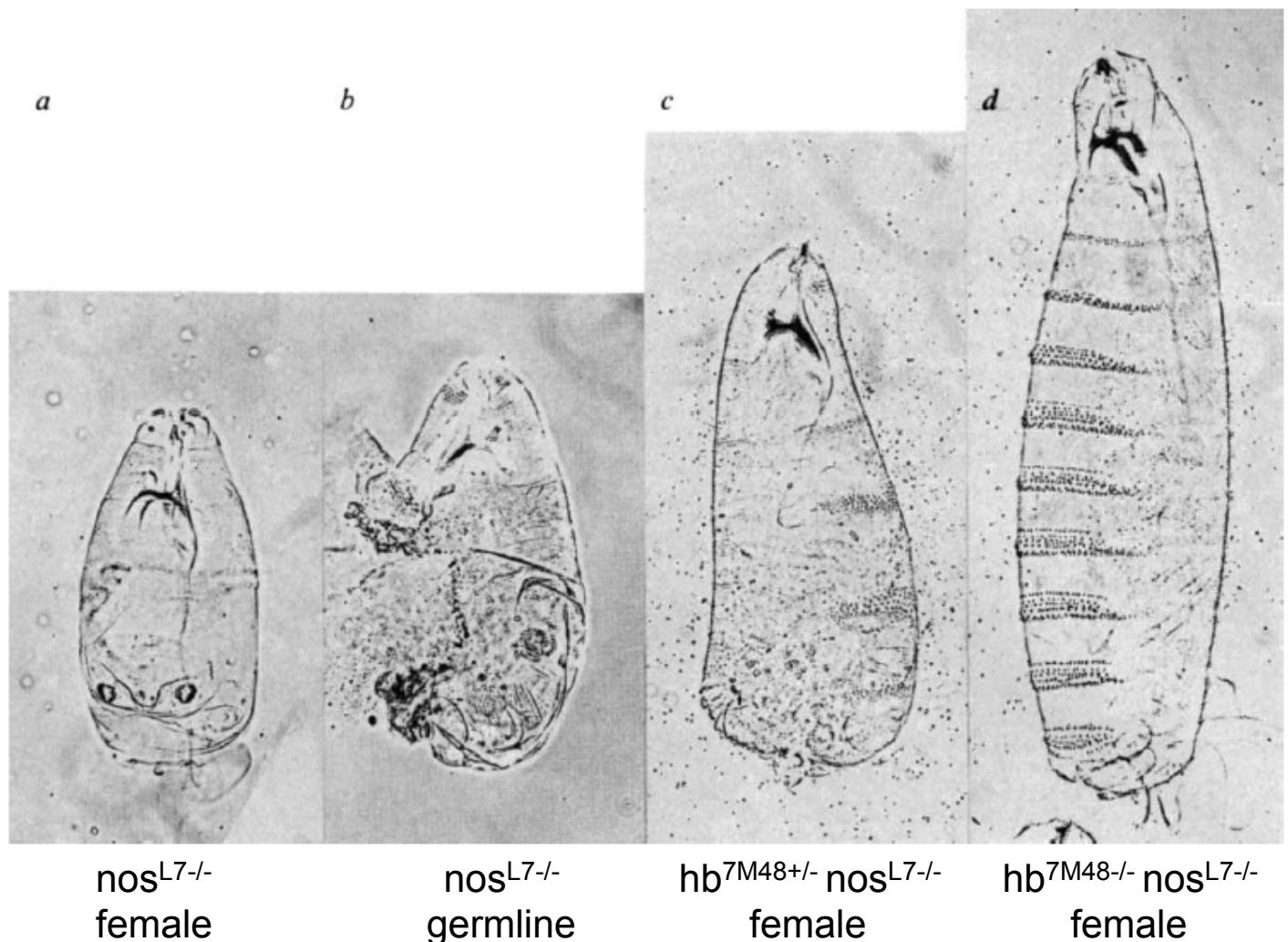
Maternal hunchback and nanos

LETTERS TO NATURE

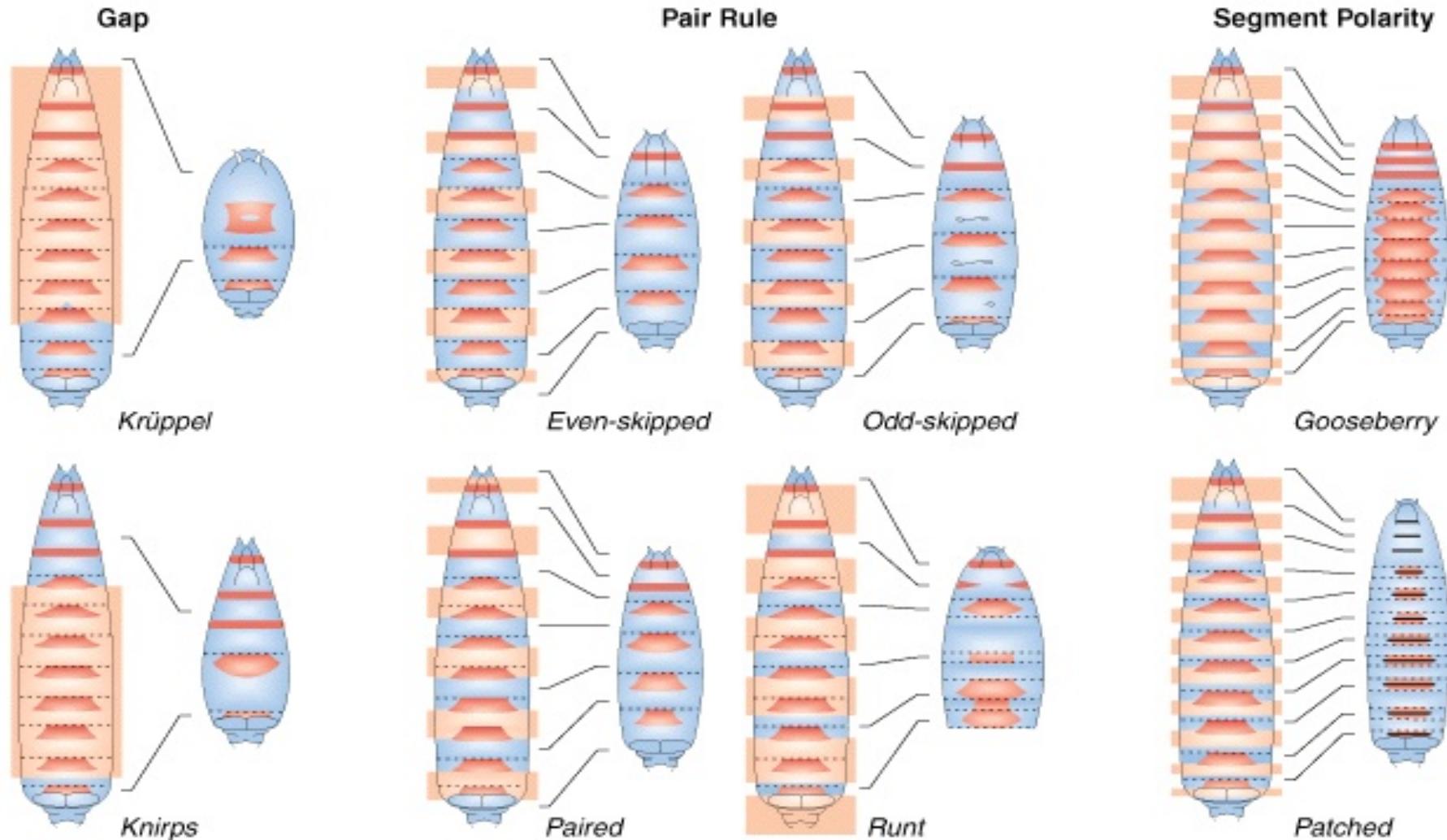
The *Drosophila* posterior-group gene *nanos* functions by repressing *hunchback* activity

Vivian Irish^{*‡}, Ruth Lehmann[†] & Michael Akam^{*}

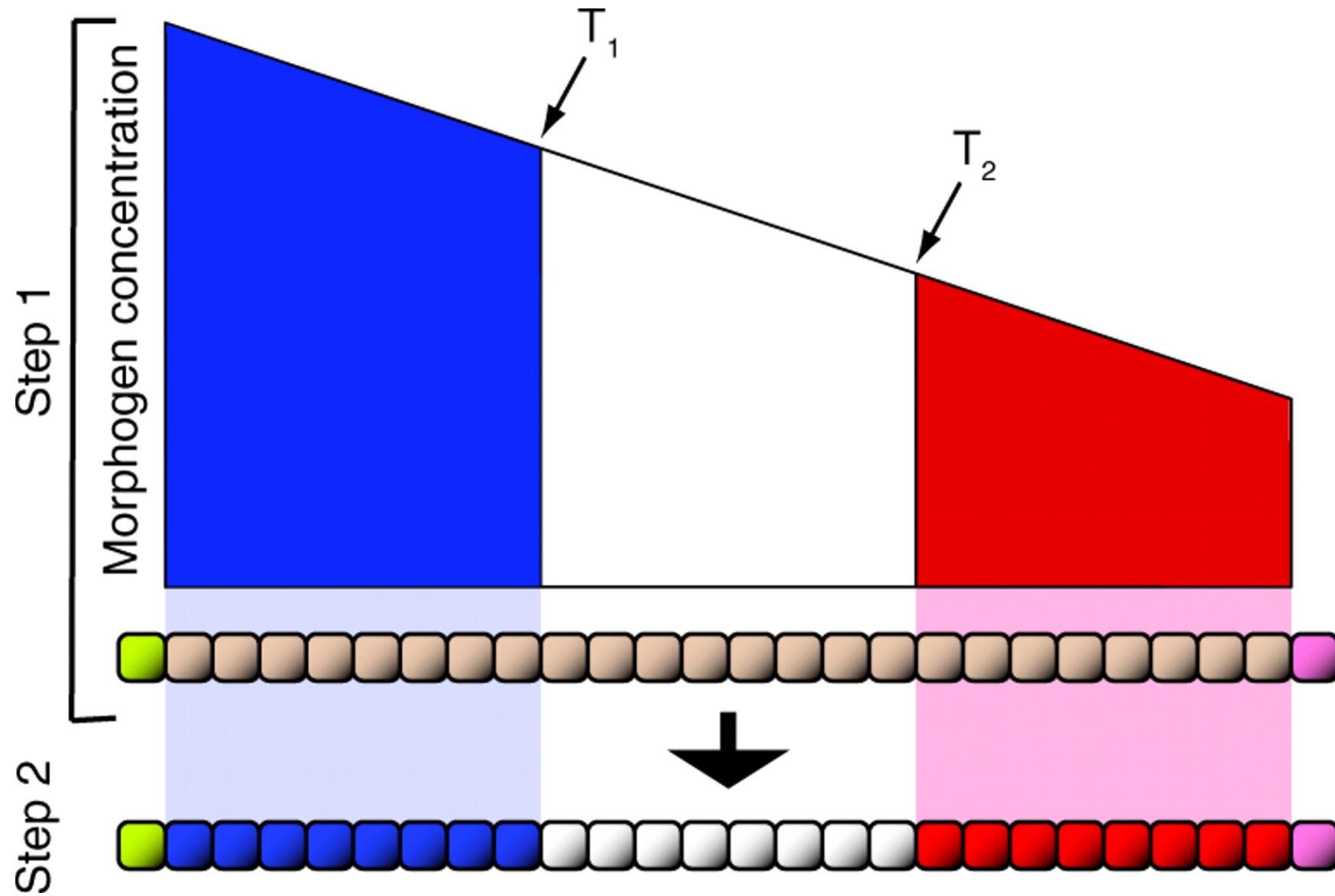
Nature 338, 646-648 (1989)



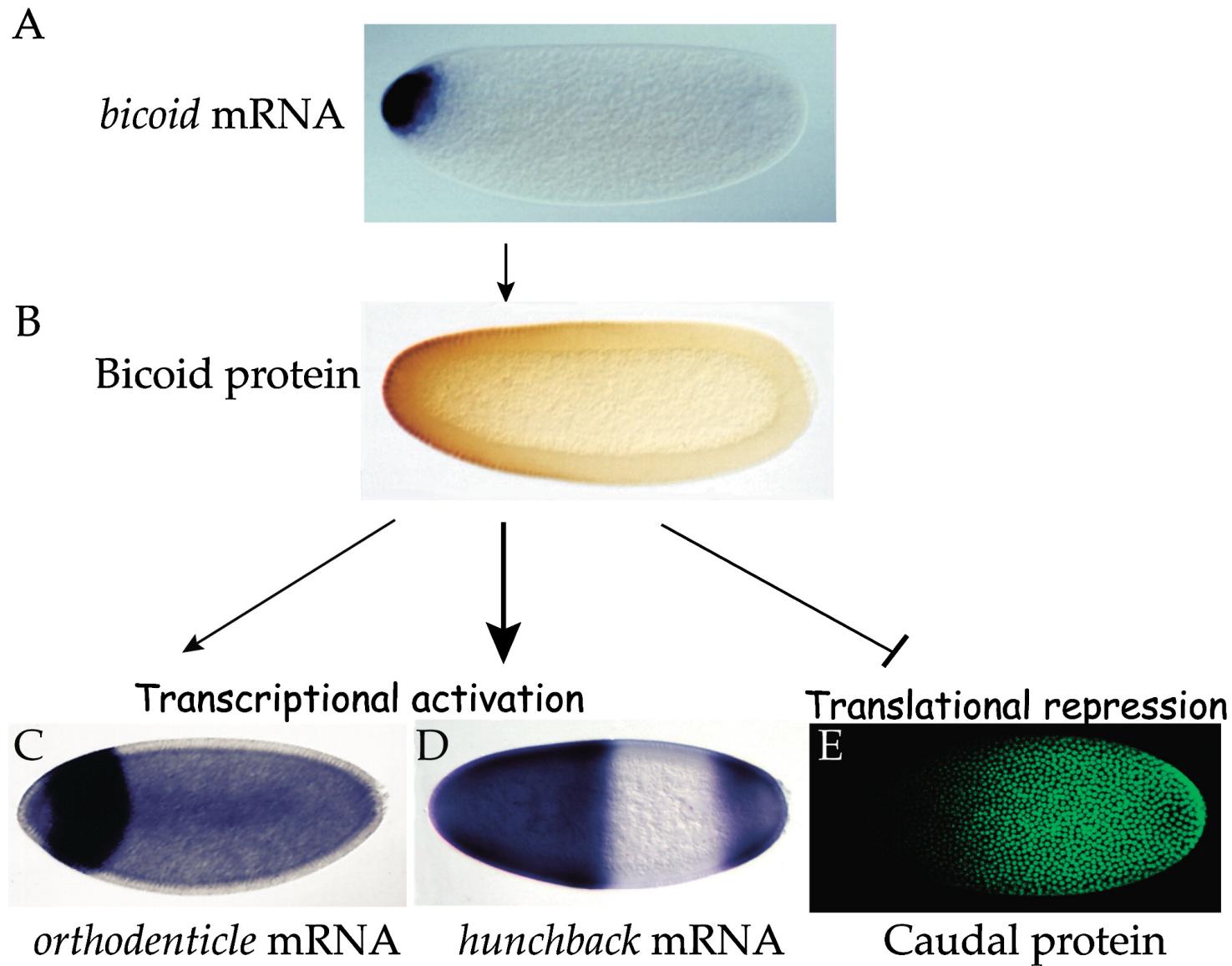
From maternal to segmentation genes



Translating the gradient(s): the French flag model



Bicoid encodes a homeodomain protein



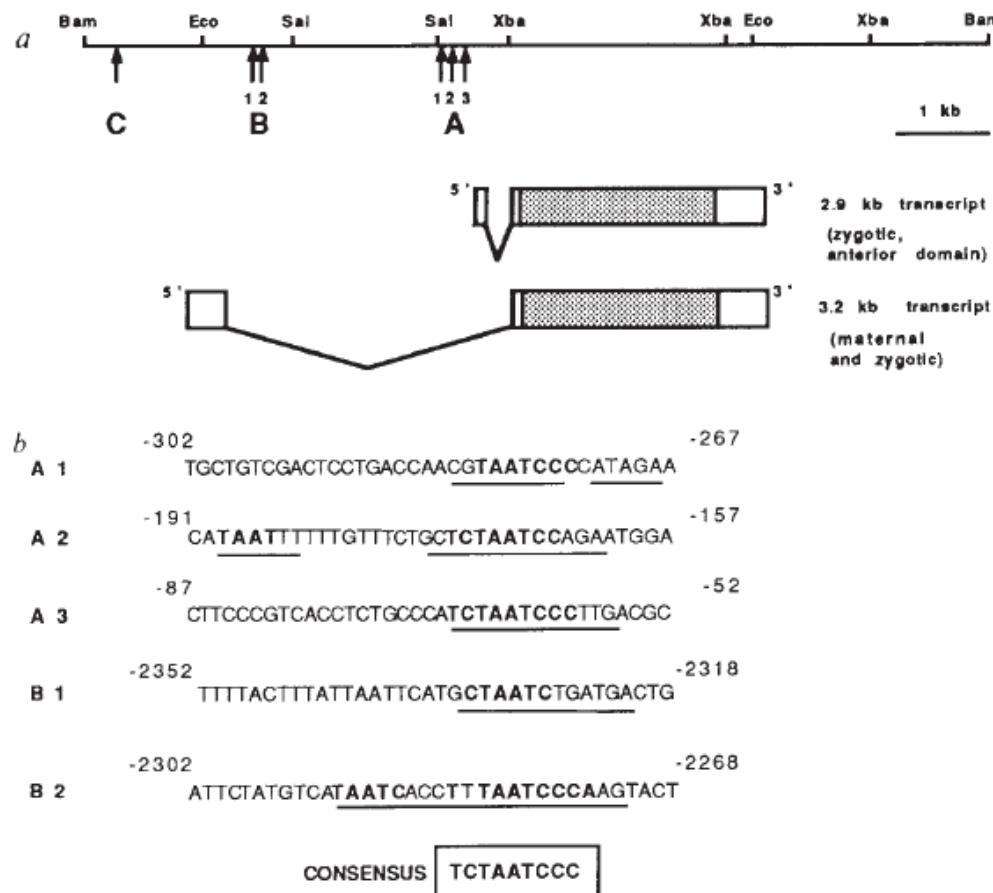
Bicoid binds Hunchback regulatory sequences

The bicoid protein is a positive regulator of *hunchback* transcription in the early *Drosophila* embryo

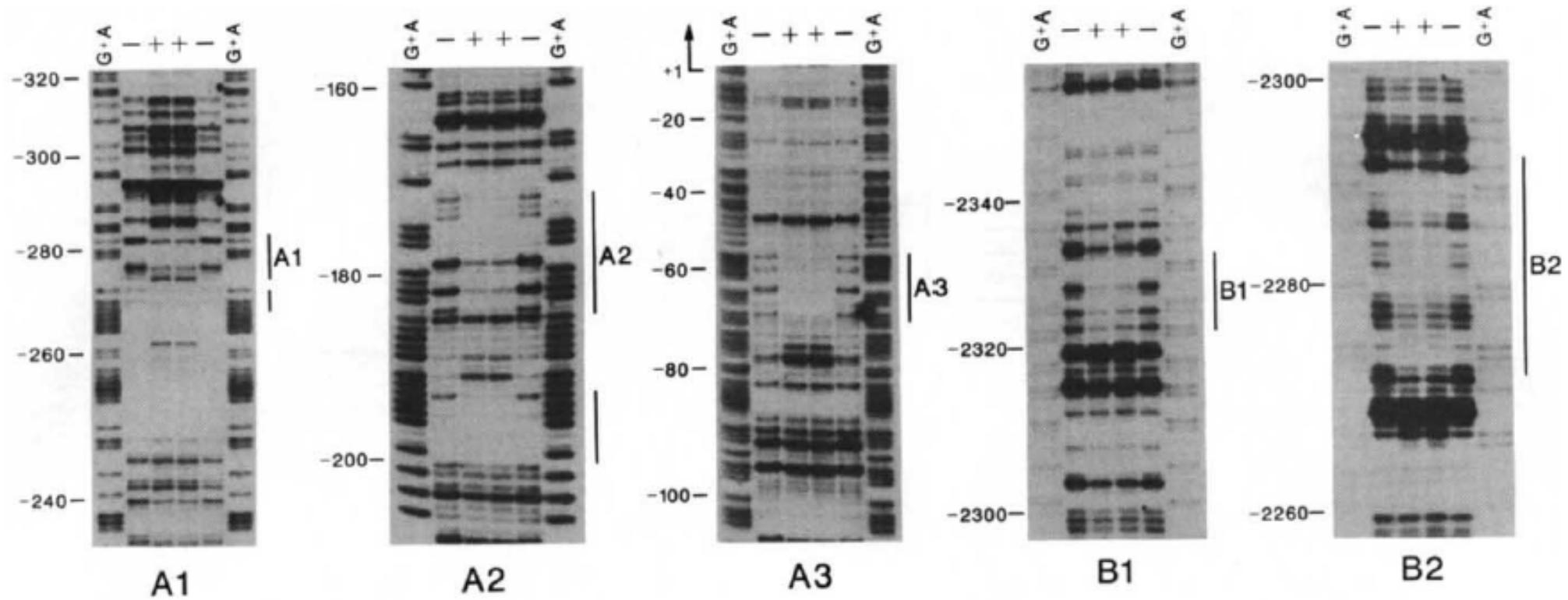
Wolfgang Driever & Christiane Nüsslein-Volhard

Max Planck Institut für Entwicklungsbiologie, Abteilung III Genetik, Spemannstrasse 35, 7400 Tübingen, FRG

Nature 337, 138-143 (1989)



Bicoid binds Hunchback regulatory sequences



Bicoid: DNA binding specificity

DNA Specificity of the Bicoid Activator Protein
Is Determined by Homeodomain Recognition Helix
Residue 9

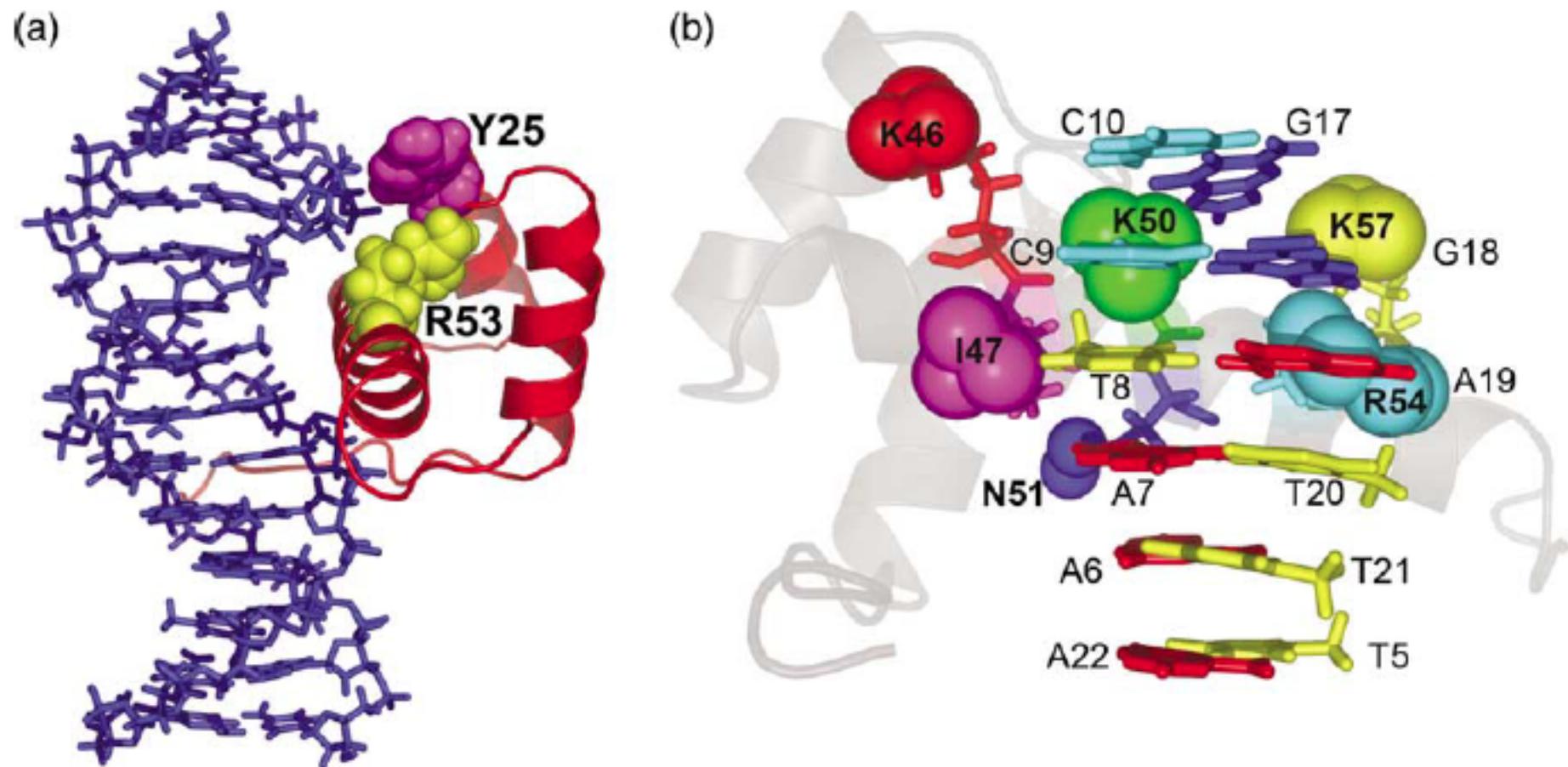
Hanes & Brent (1989) Cell 57, 1275-1283

	Activation of Gene Expression		
	<u>LexA Op</u>	Bicoid site	Antp class site
Wild-type Bicoid	+	+	-
A1	+	+	-
A5	+	+	-
A9	+	-	-
A1A5A6A9	-	-	-
R1 - - - E1 R2 Q9	+	-	+
E1 R2 Q9	+	-	+
E1 R2	+	+	-
Q9	+	-	+
Antp class *	+	-	+

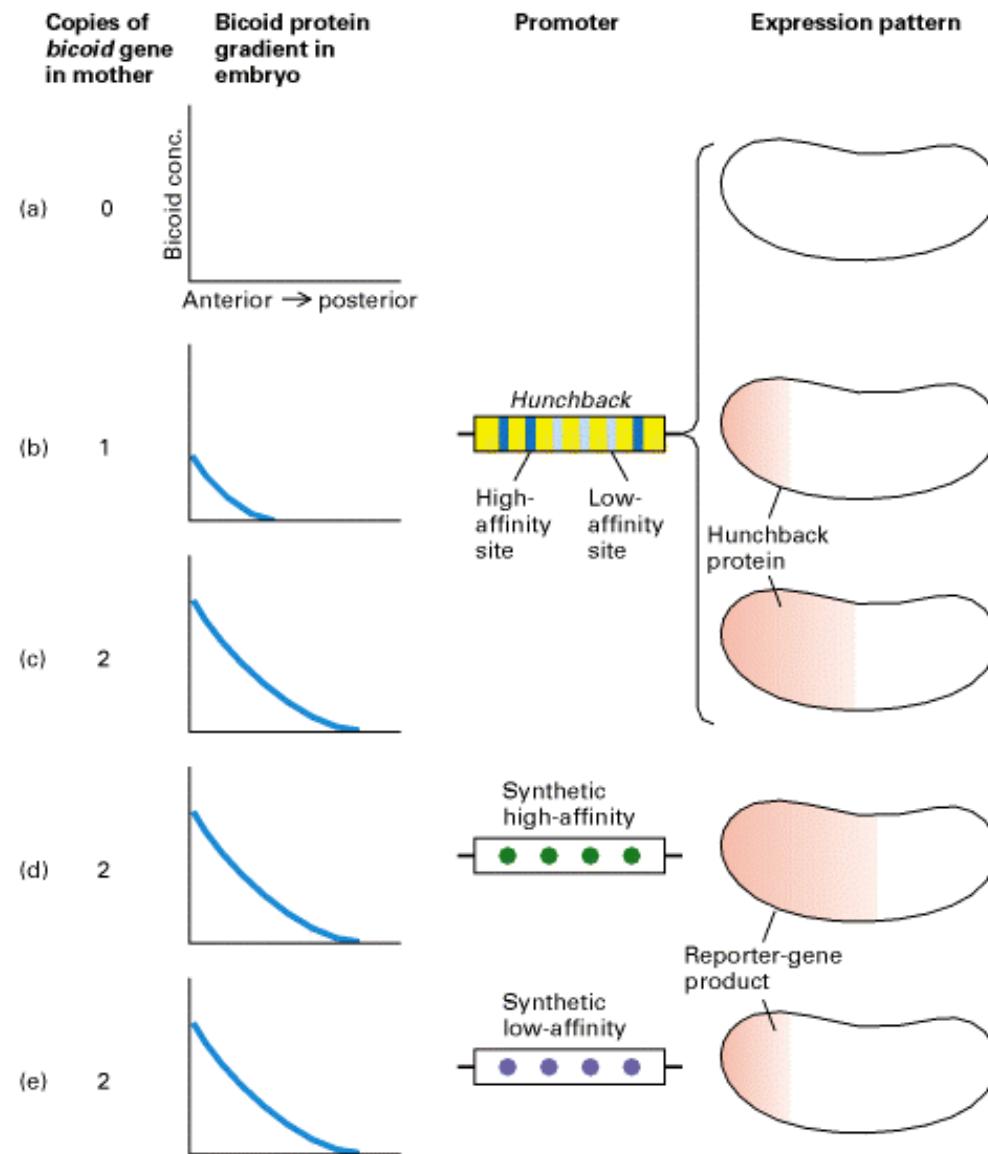
DNA sequences shown above the table:

- Wild-type Bicoid: SAKL[ALG]TAQVKIWFKN (residues 1-10)
- A1: [A]AQVKIWFKN
- A5: [I]AQV[A]WFKN
- A9: [T]AQV[A]WFAN
- A1A5A6A9: [A]AQV[A][A]WFAN
- R1 - - - E1 R2 Q9: [R] - - - [E]BQVKIWFQN
- E1 R2 Q9: [E]BQVKIWFQN
- E1 R2: [E]BQVKIWFKN
- Q9: [T]AQVKIWFQN
- Antp class *: AHAL[CLT][ERQIKIWFQN]

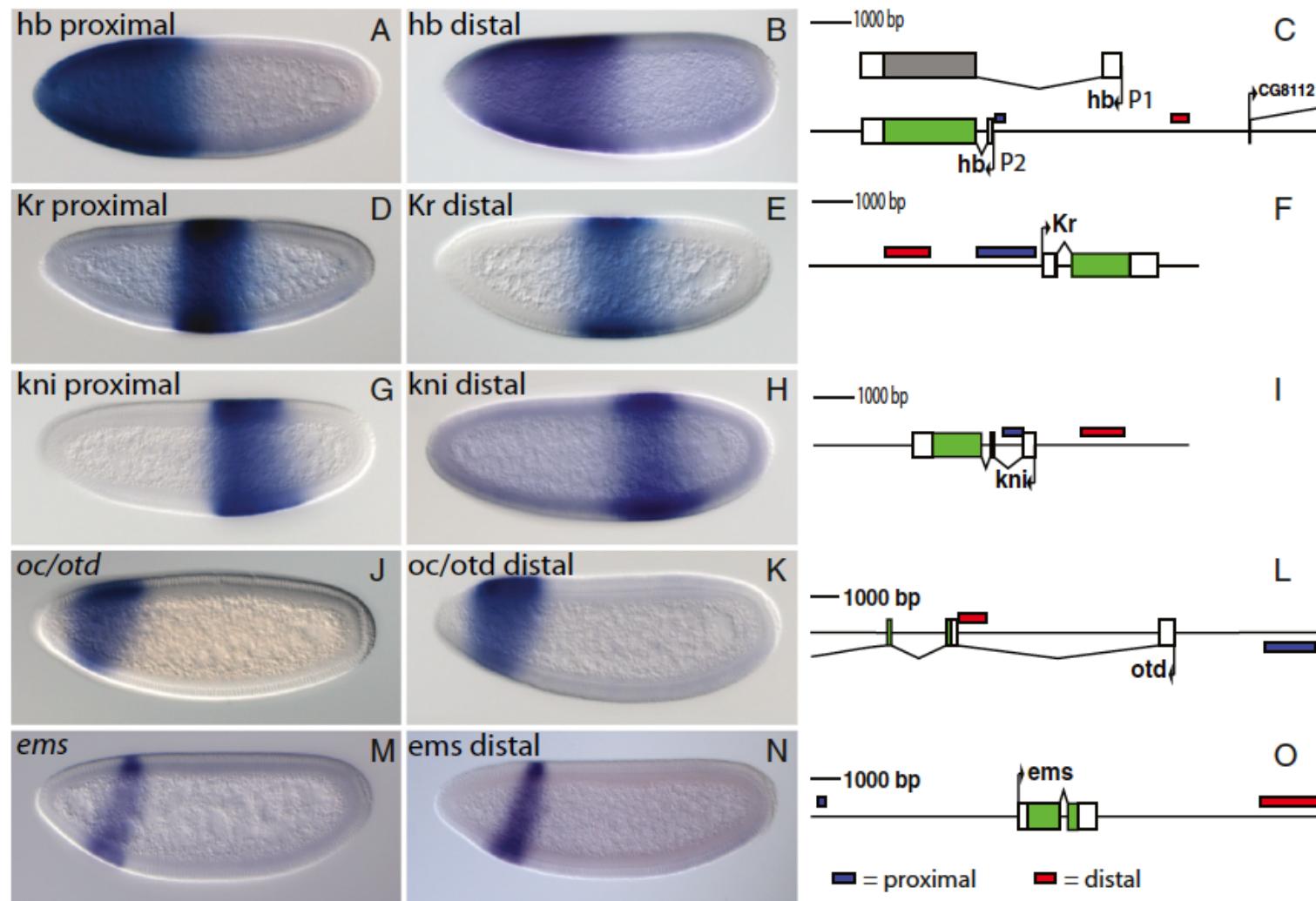
Bicoid-DNA complex



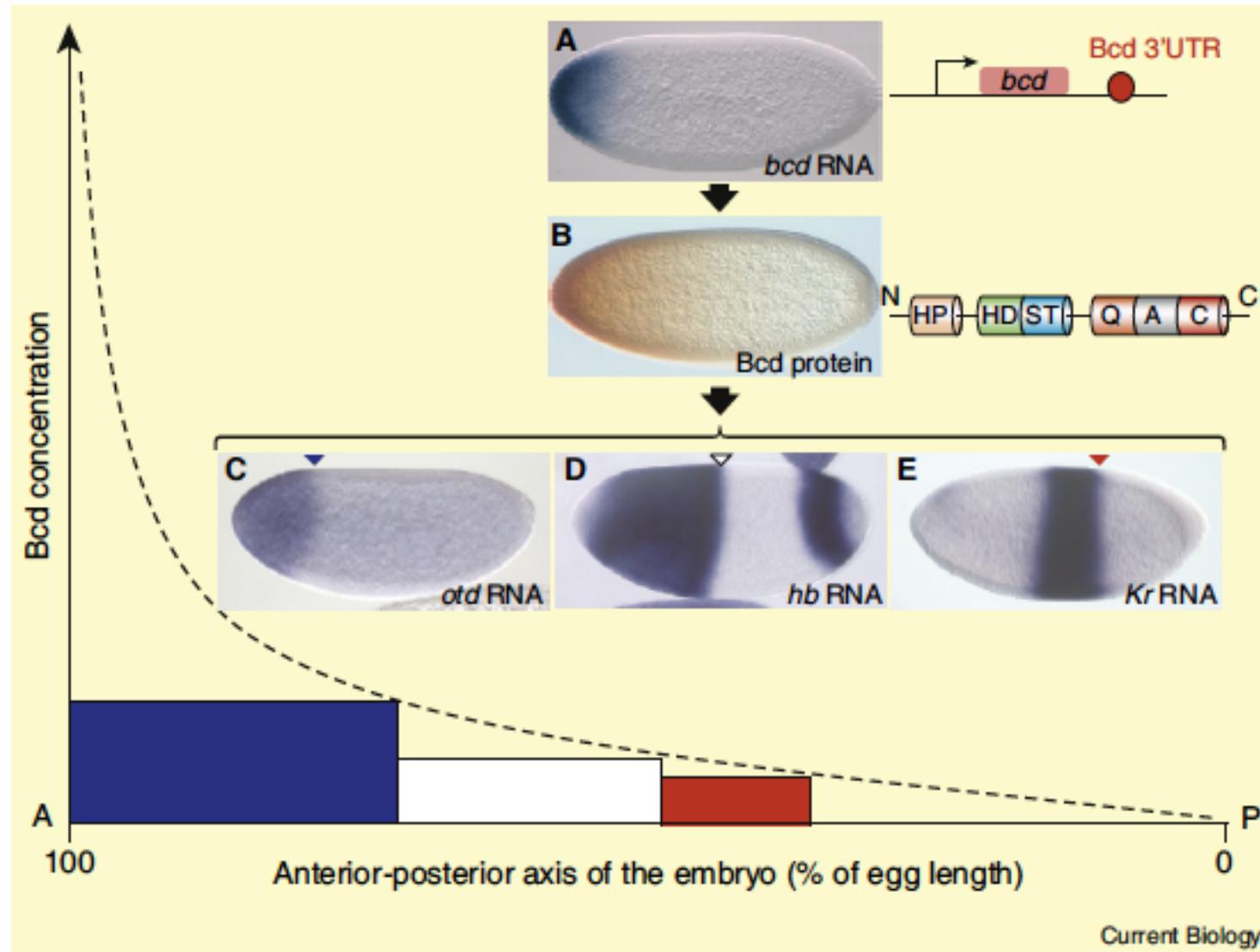
Hunchback: gap gene regulation



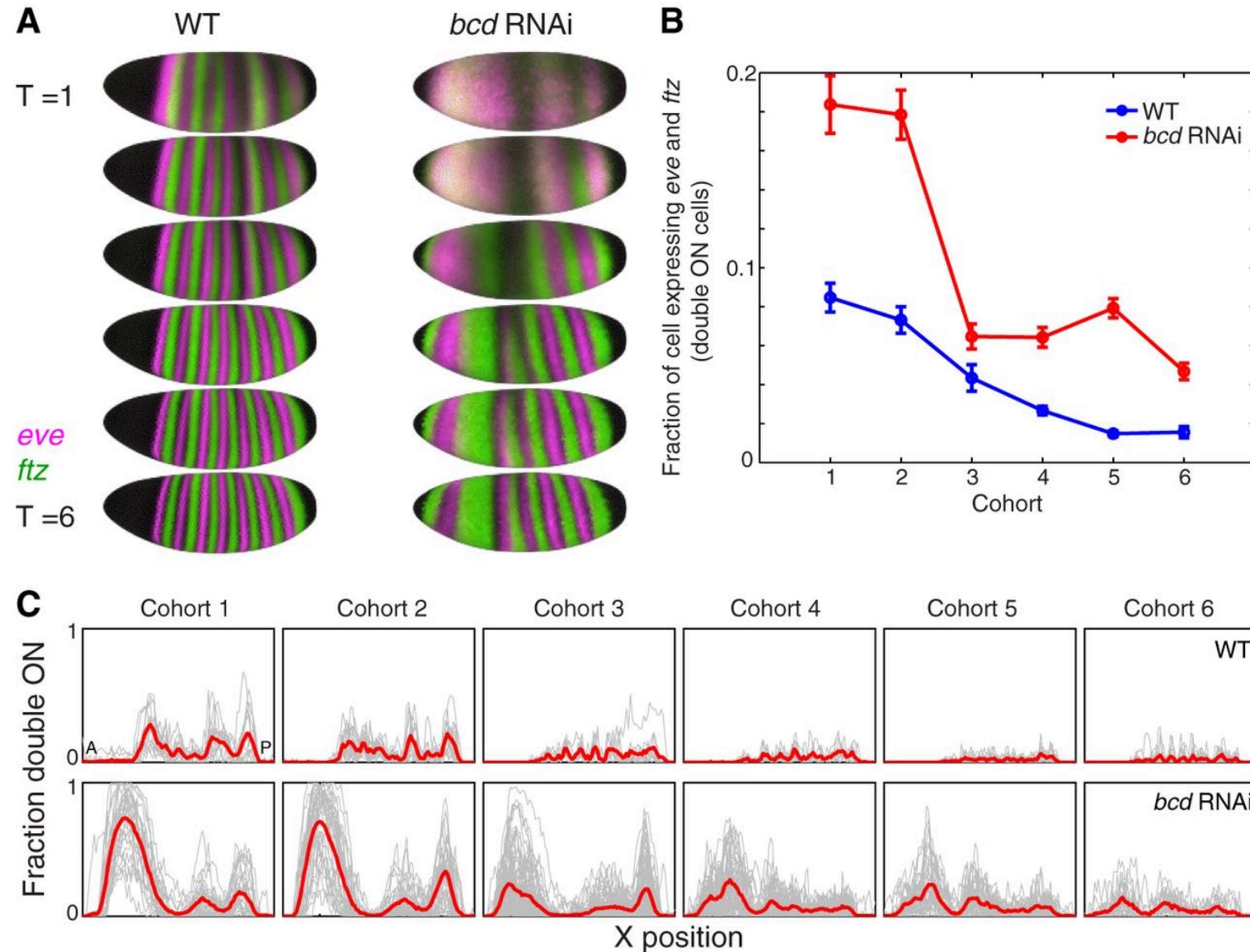
Gap genes



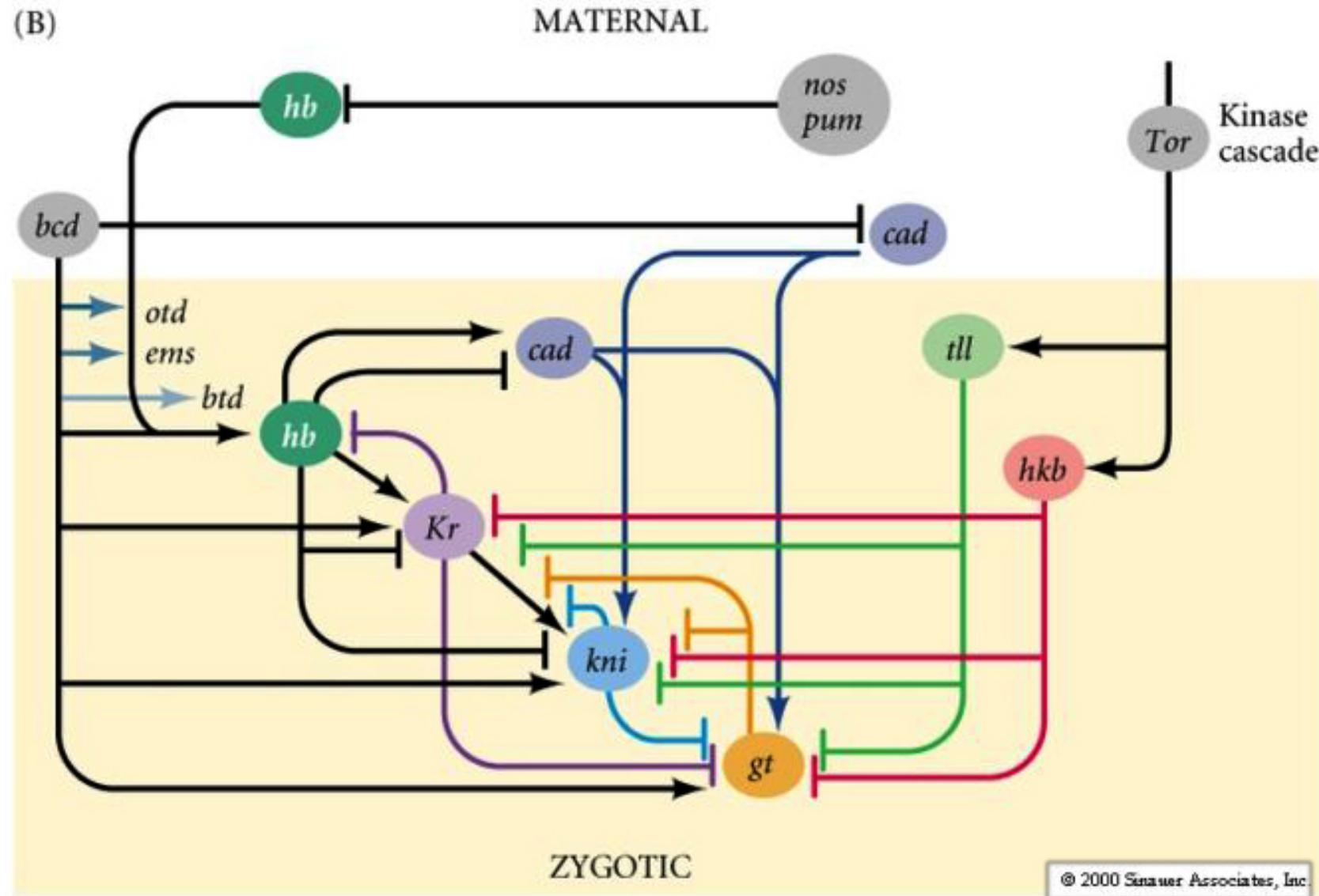
Bicoid: anterior determinant



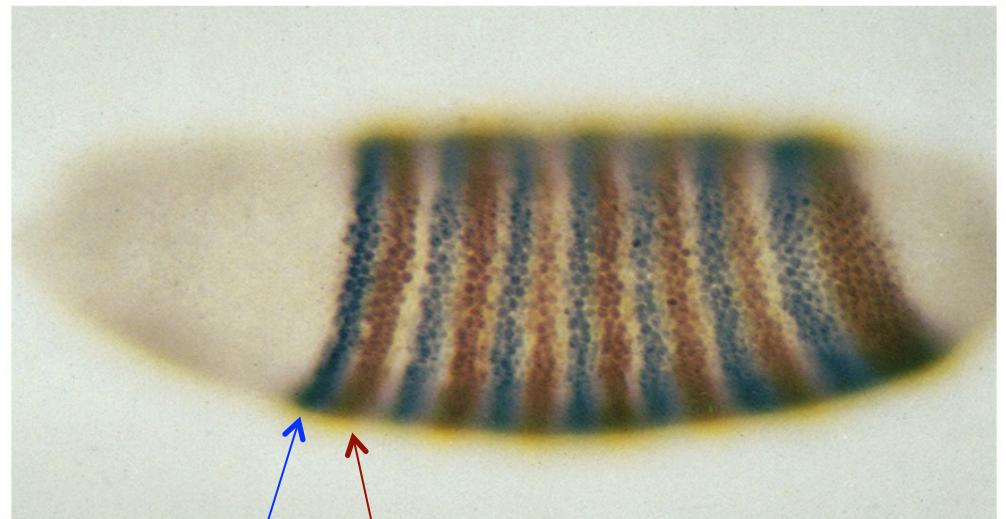
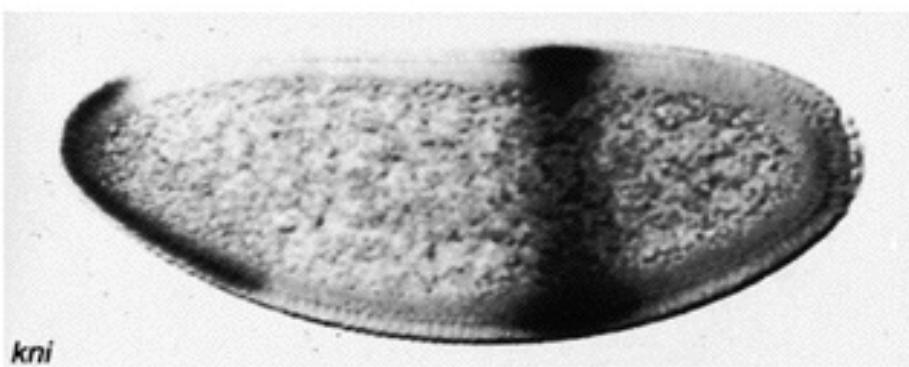
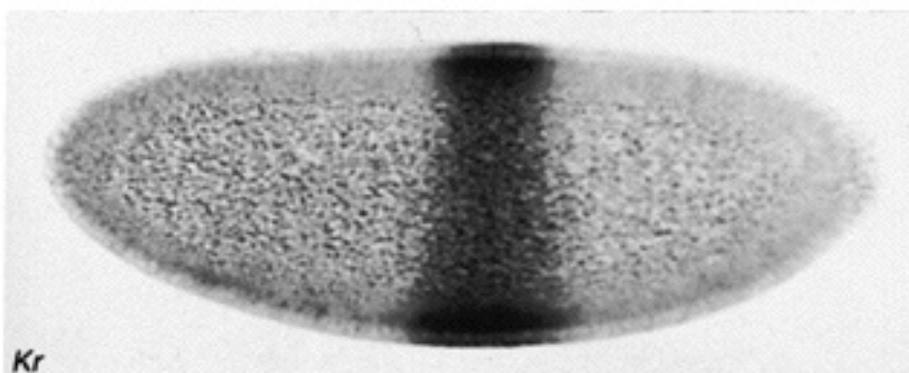
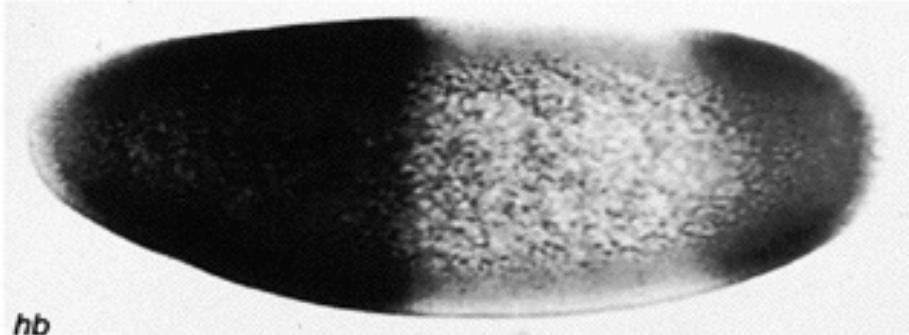
Bicoid: anterior determinant - messing with gradients



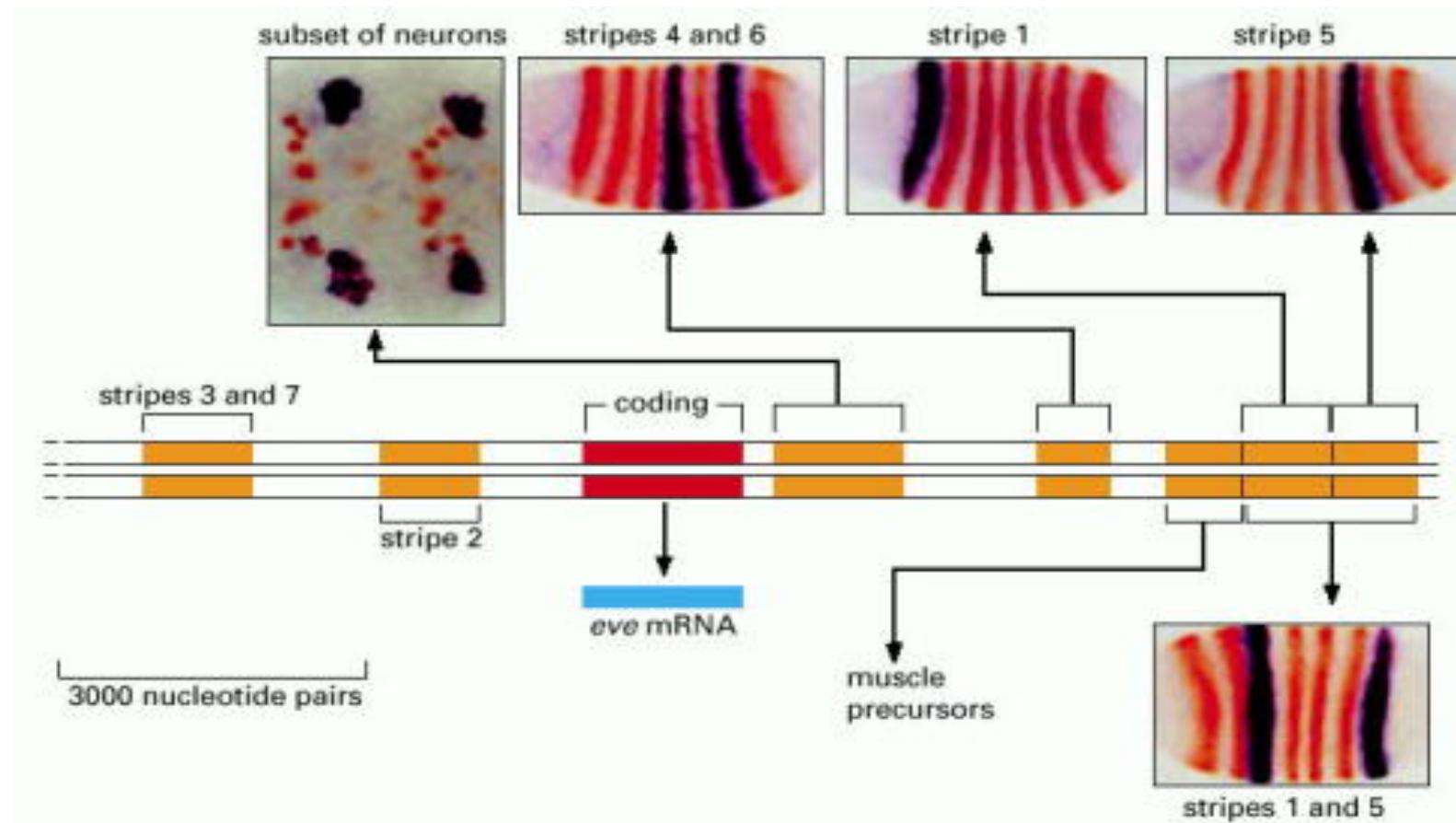
Translating the gradient(s): not quite French flag



Gap and pair-rule genes

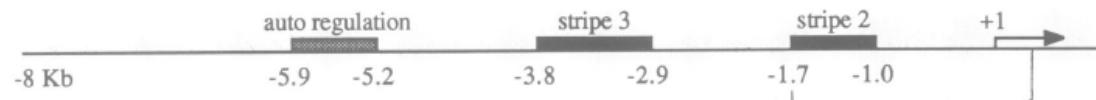


Even-skipped regulation: a tale of many enhancers

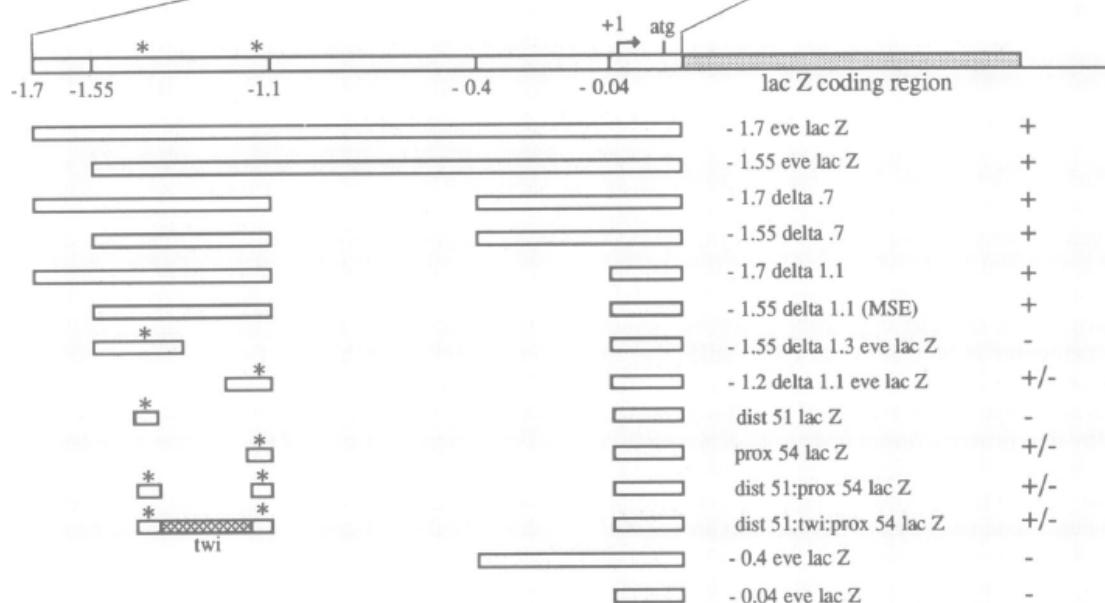


Even-skipped stripe 2: a model enhancer

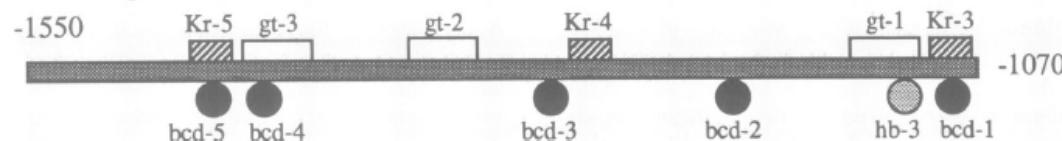
A.



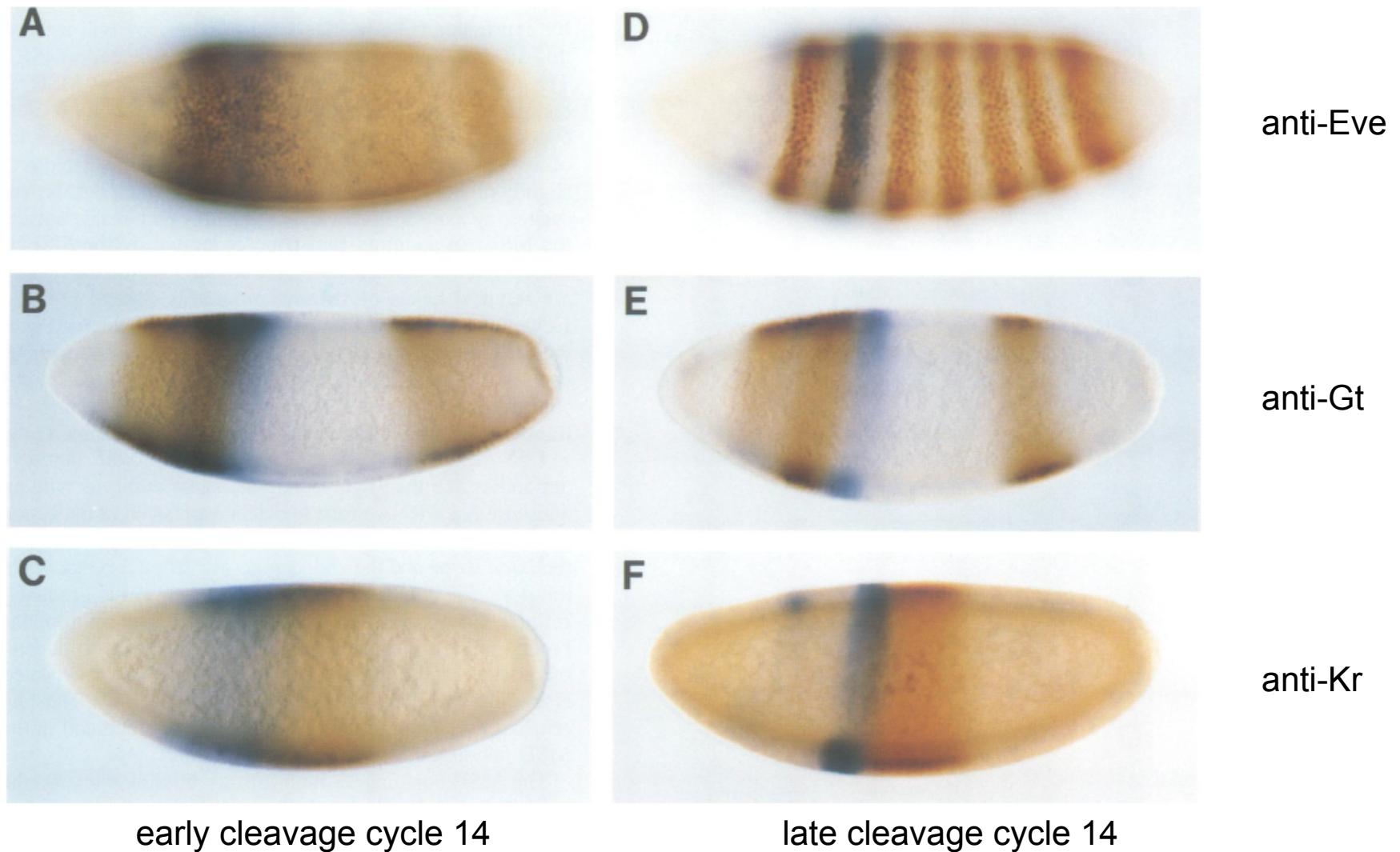
B.



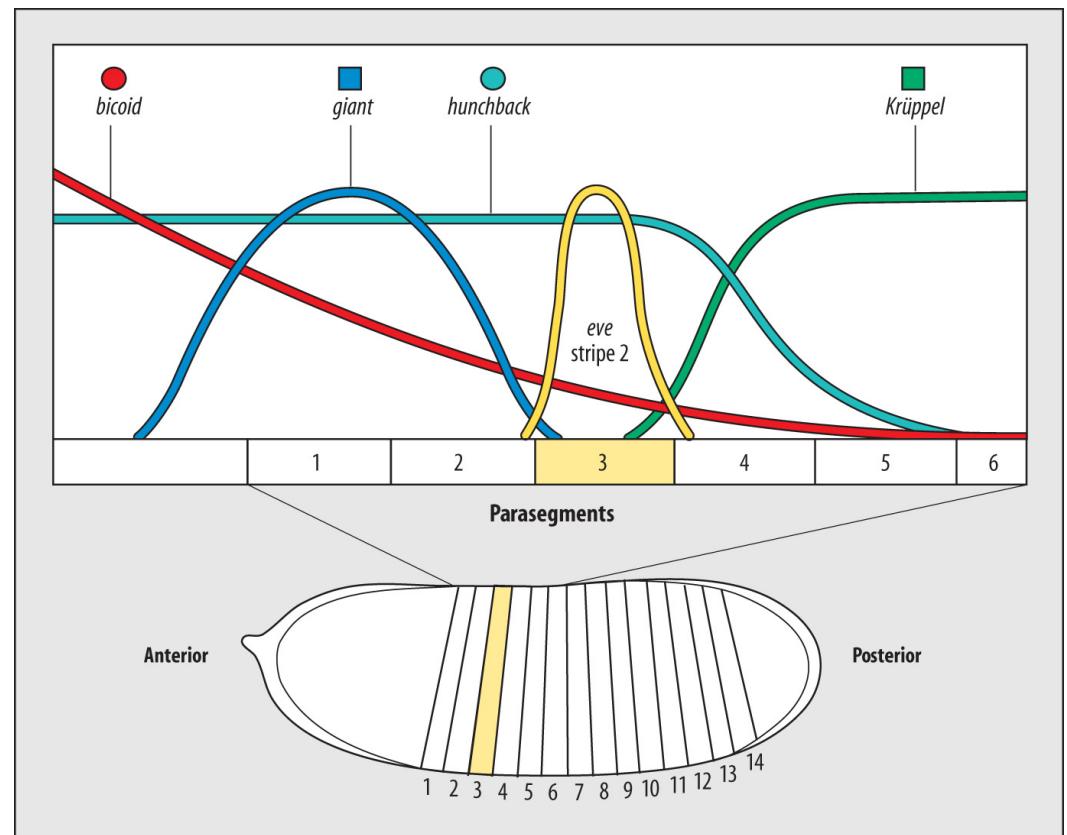
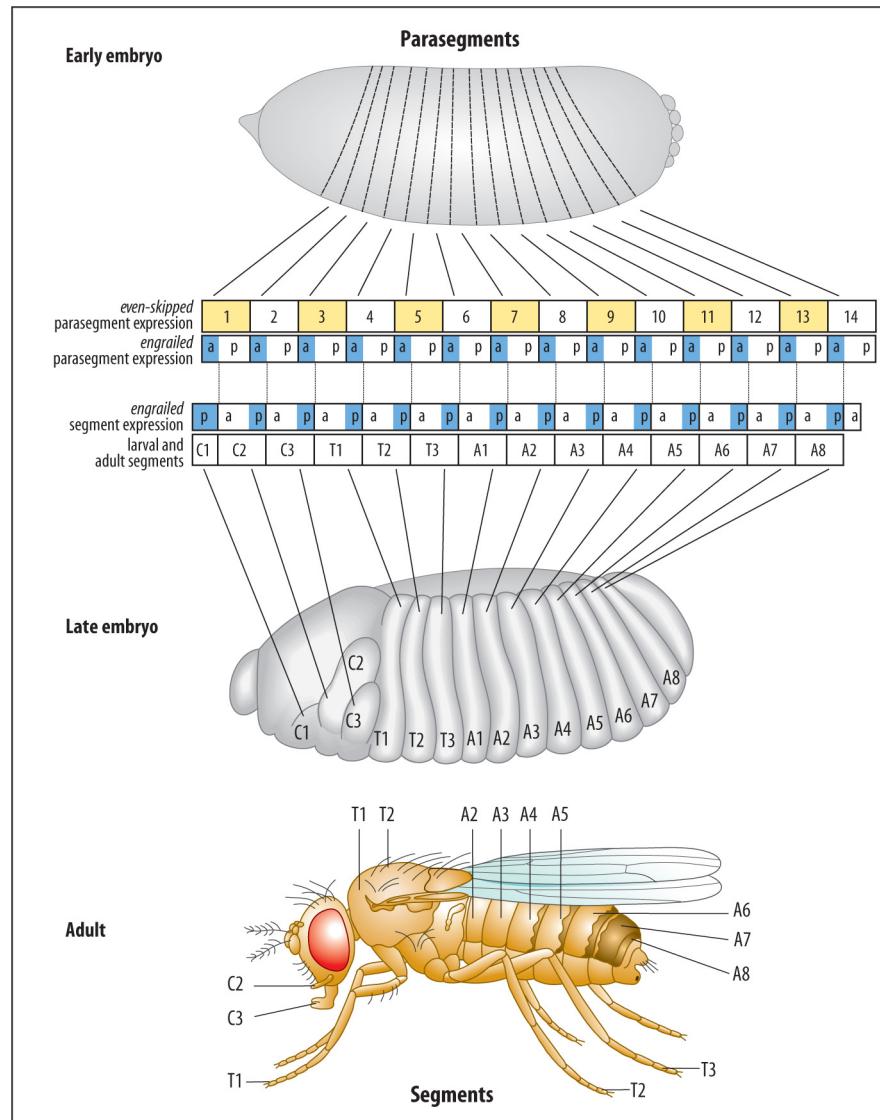
C. Binding sites in the MSE:



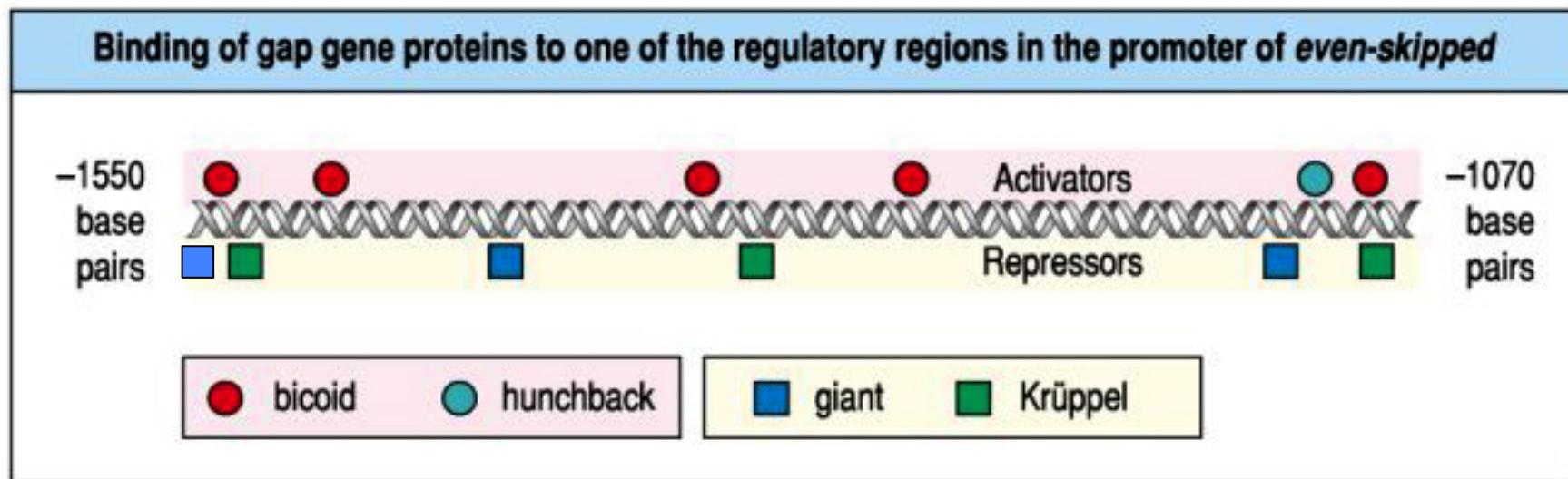
Even-skipped stripe 2: a model enhancer



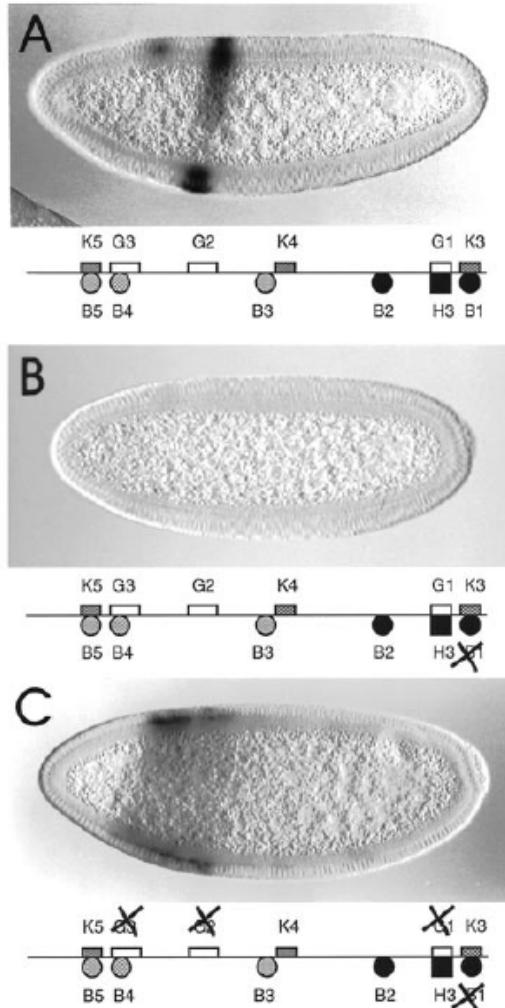
Even-skipped stripe 2: a model enhancer



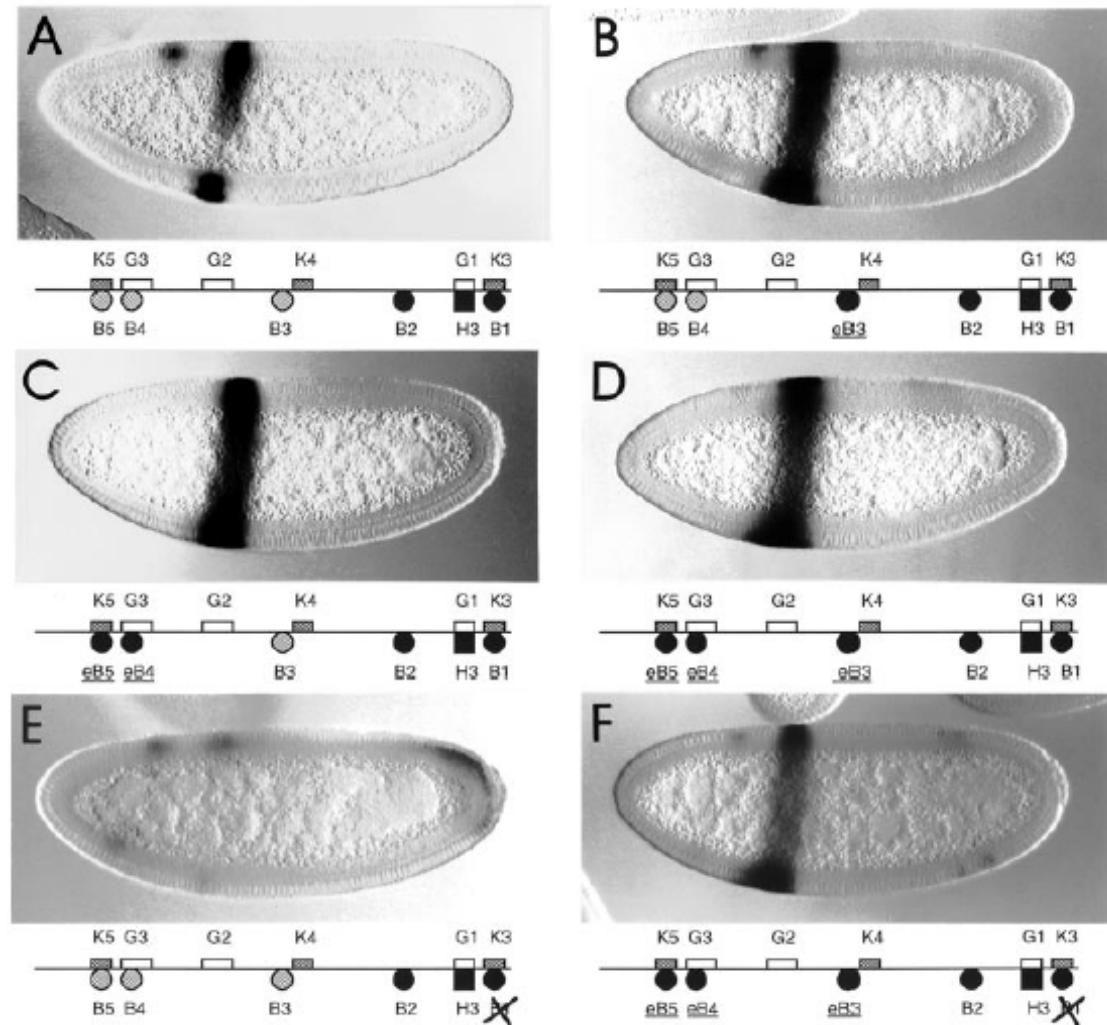
Even-skipped stripe 2: a model enhancer



Even-skipped stripe 2: a model enhancer



Cis-complementation of a defective eve-2 enhancer by removal of gt repressor sites

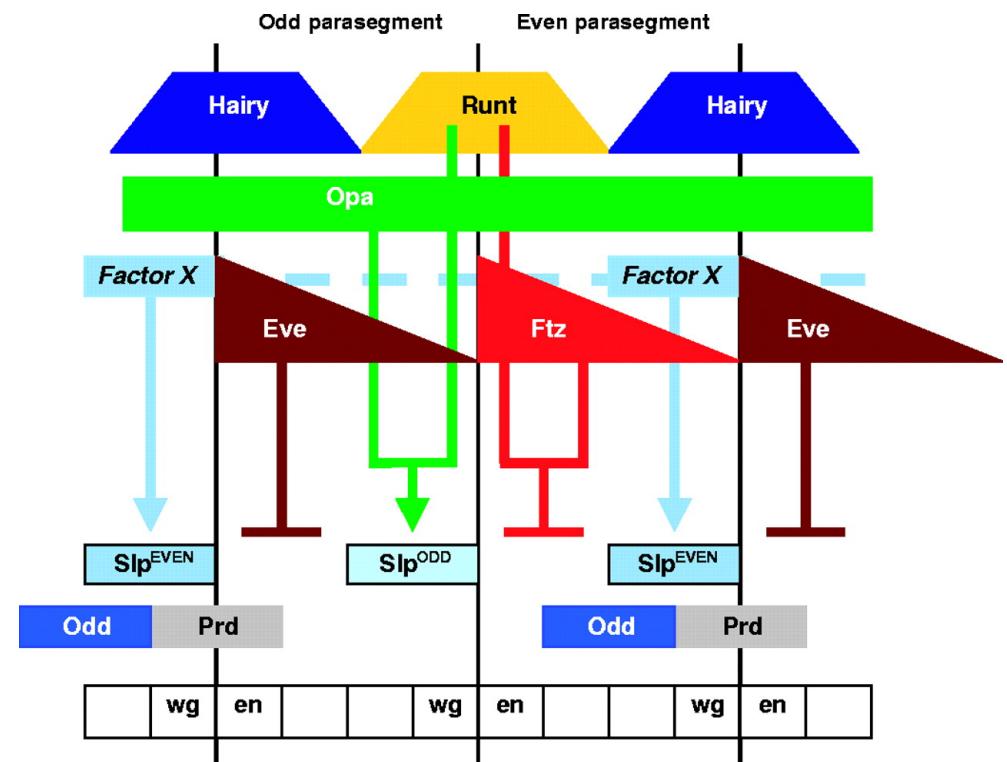
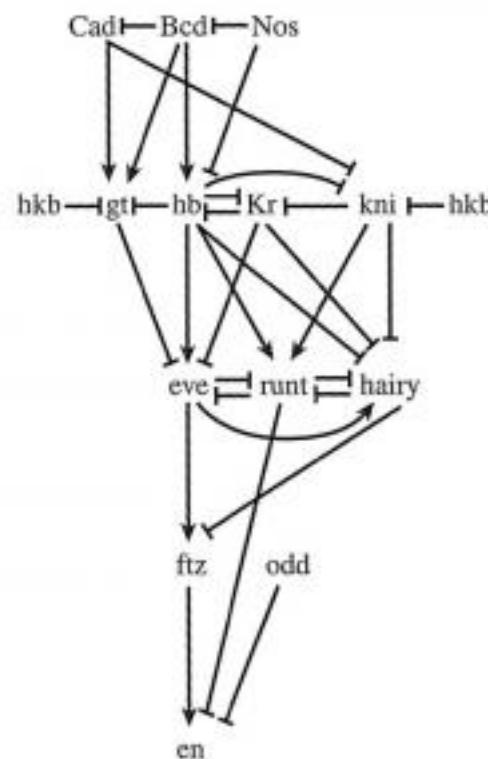
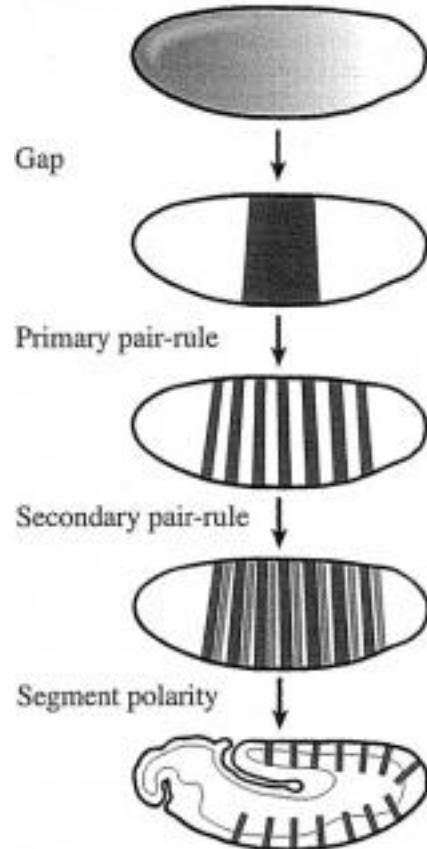


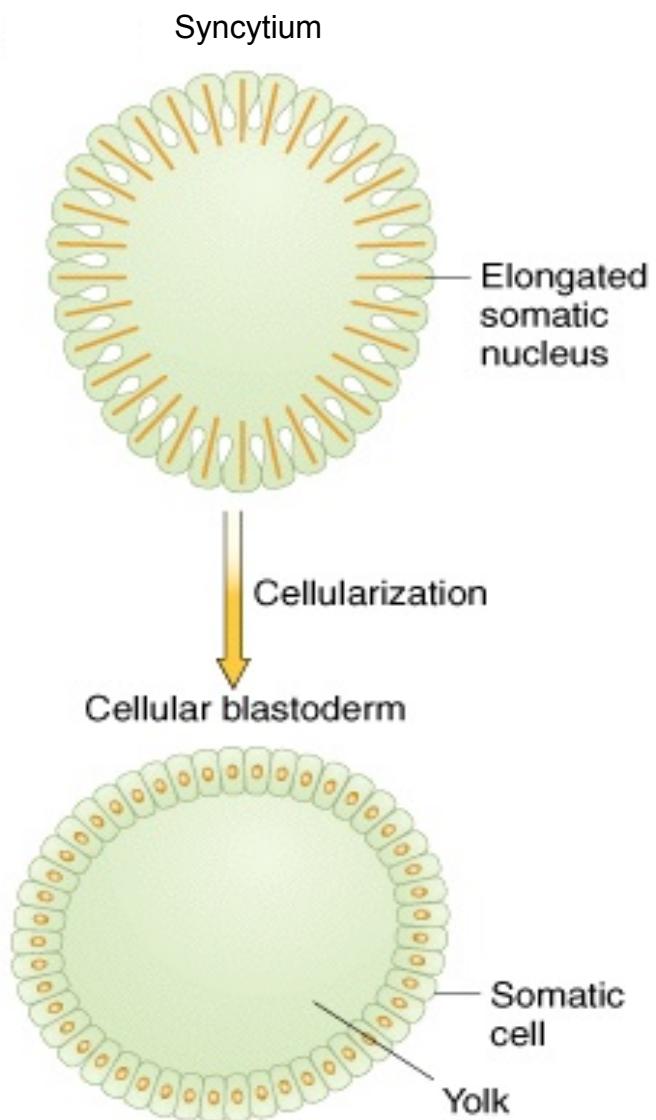
Cis-complementation by augmenting the affinities of weak bcd-binding sites

Arnosti et al. (1996) Development 122, 205-214.

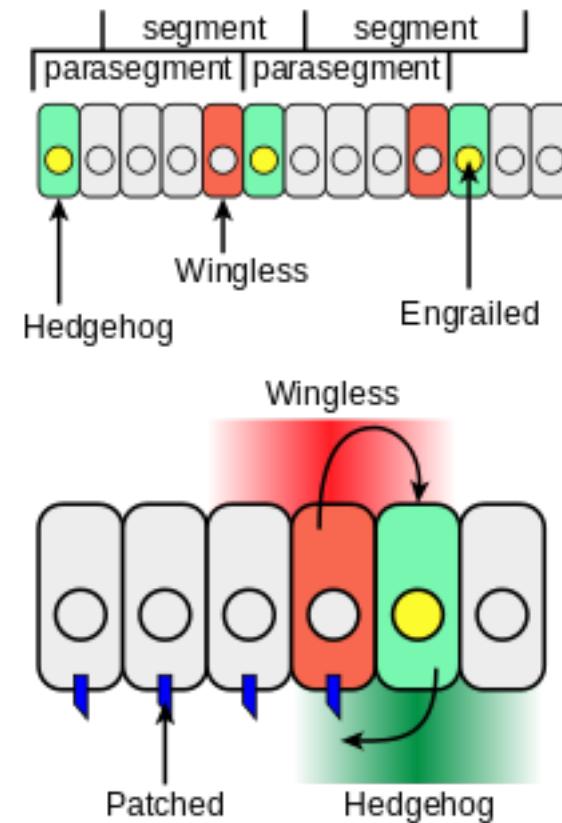
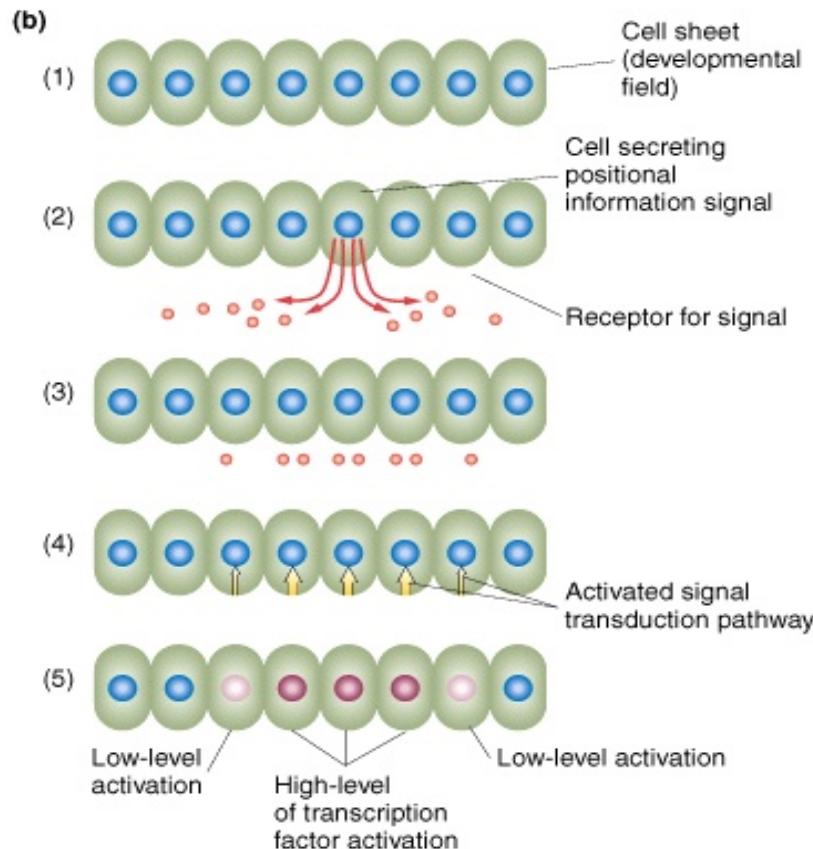
Pair-rule and segment polarity genes

Maternal

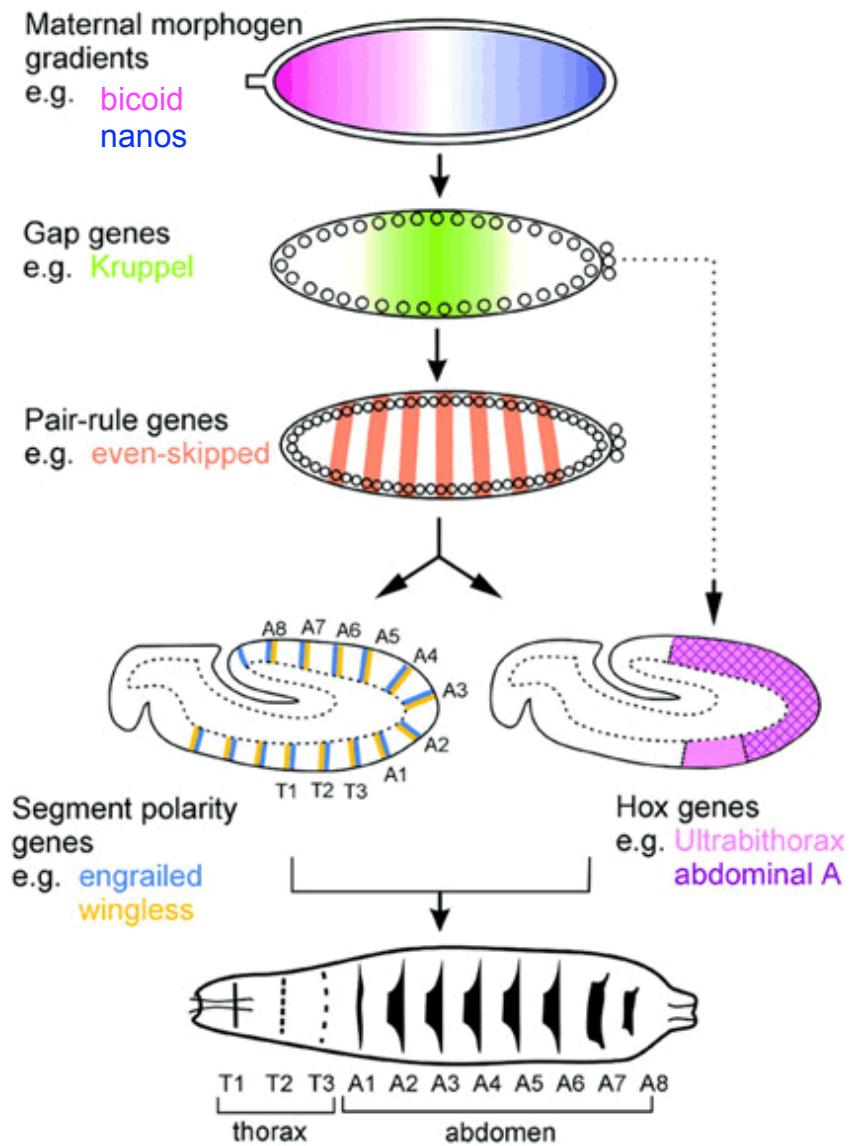
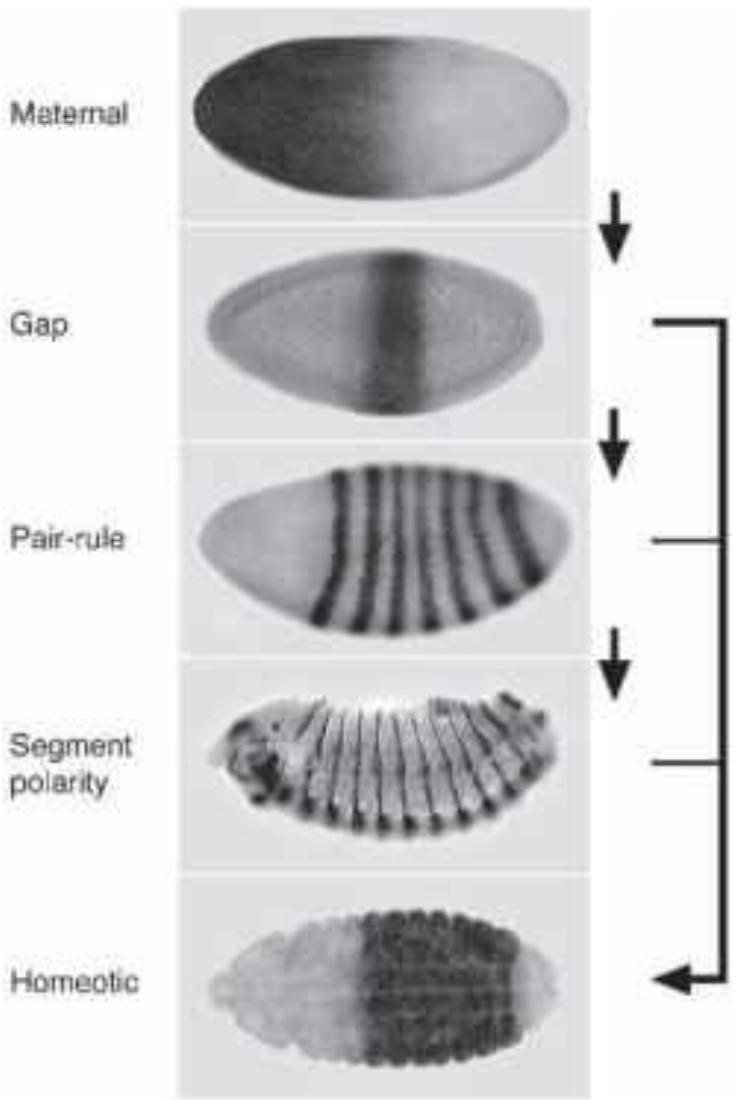




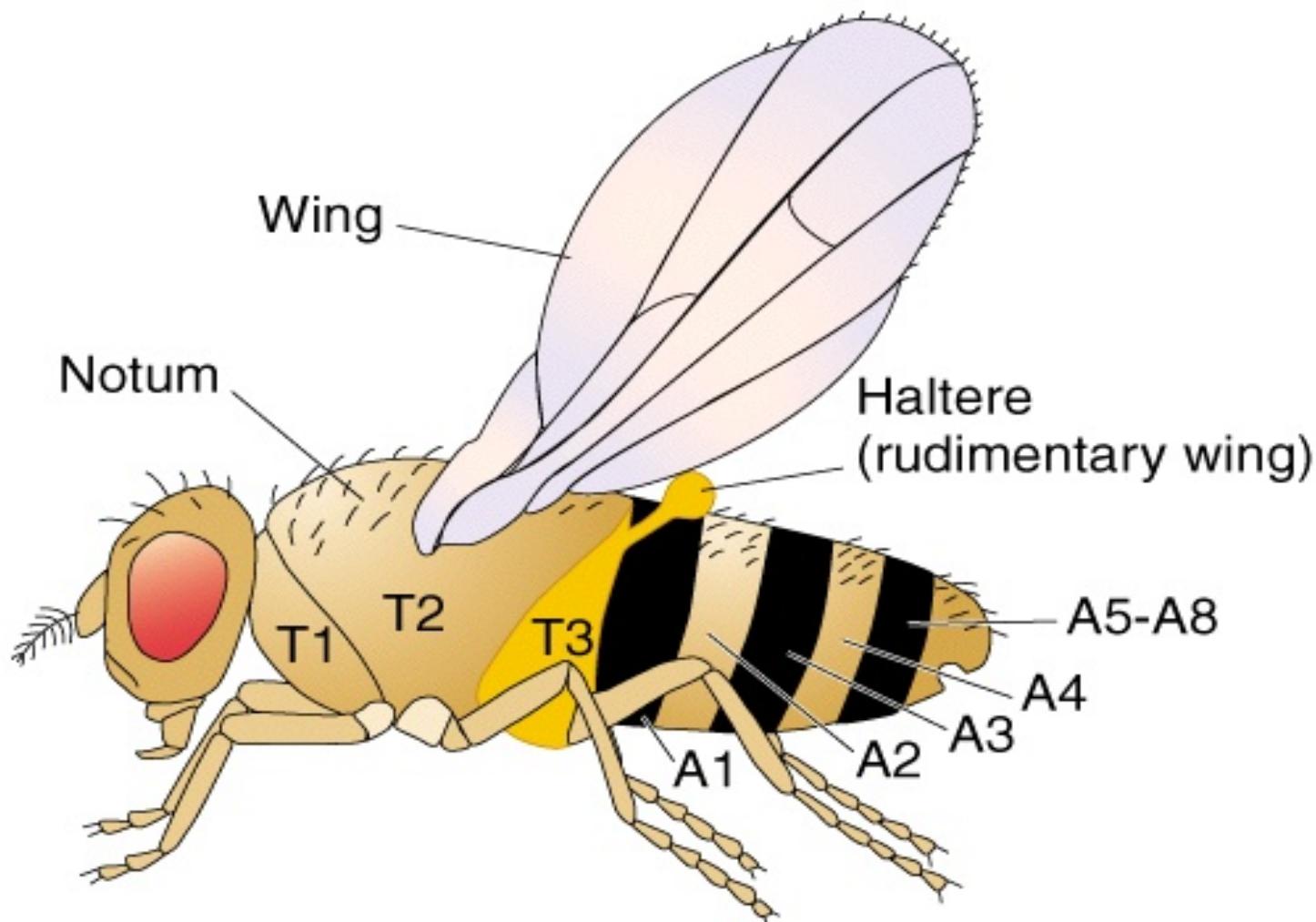
Pair-rule and segment polarity genes



Putting it all together

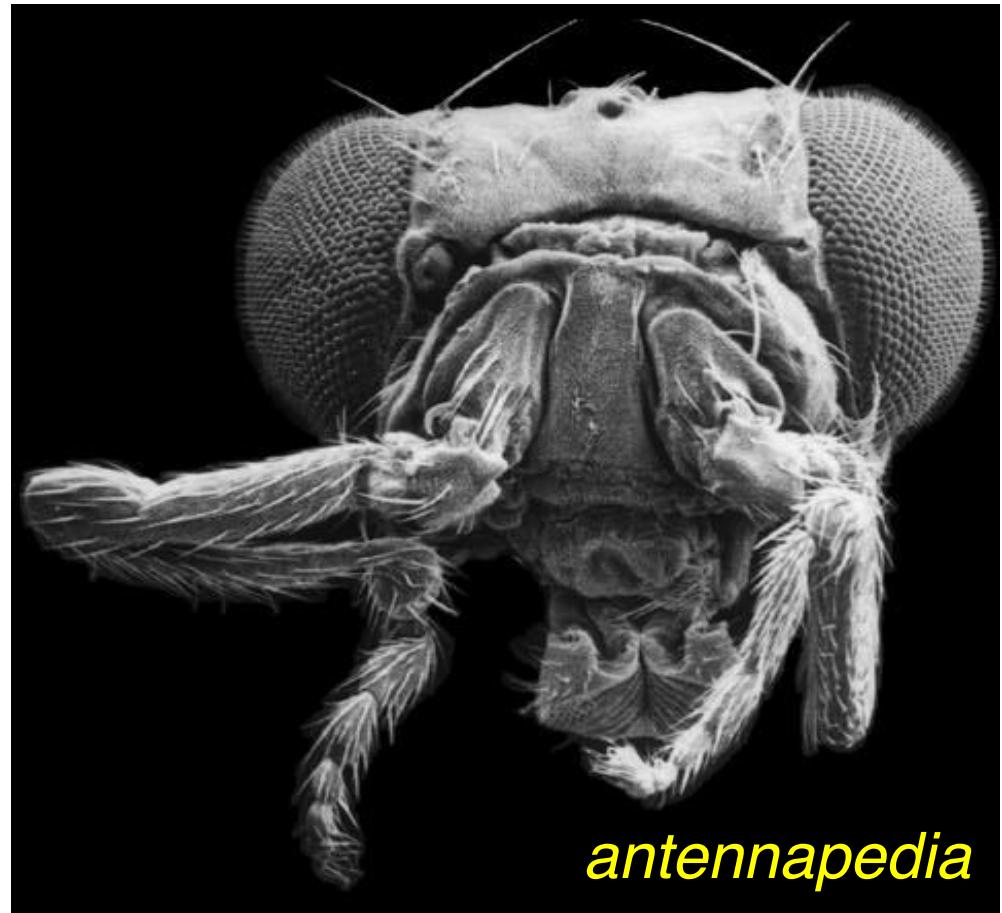
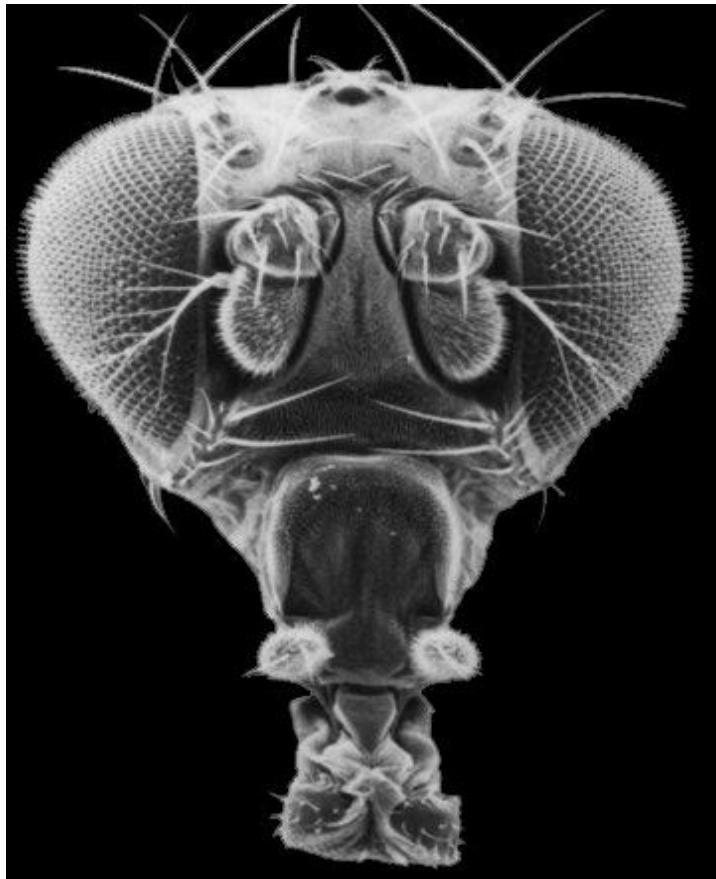


Homeotic genes and body plan

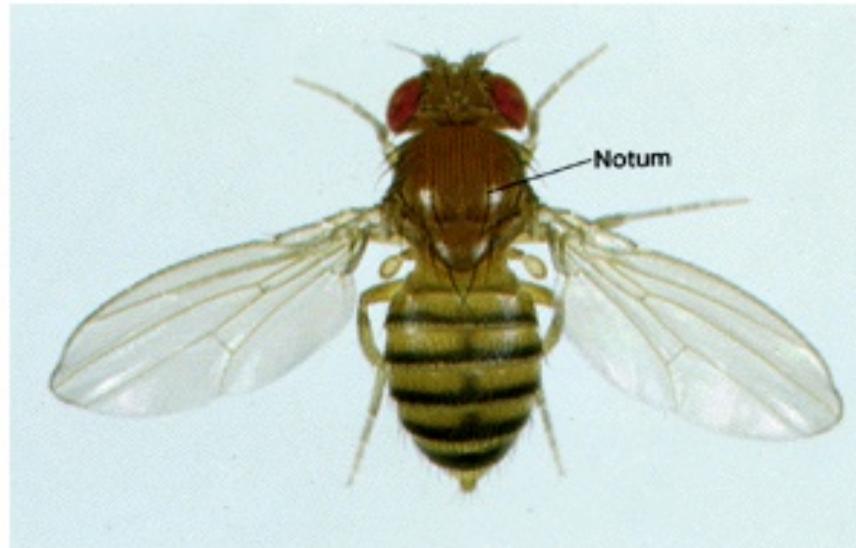


(a)

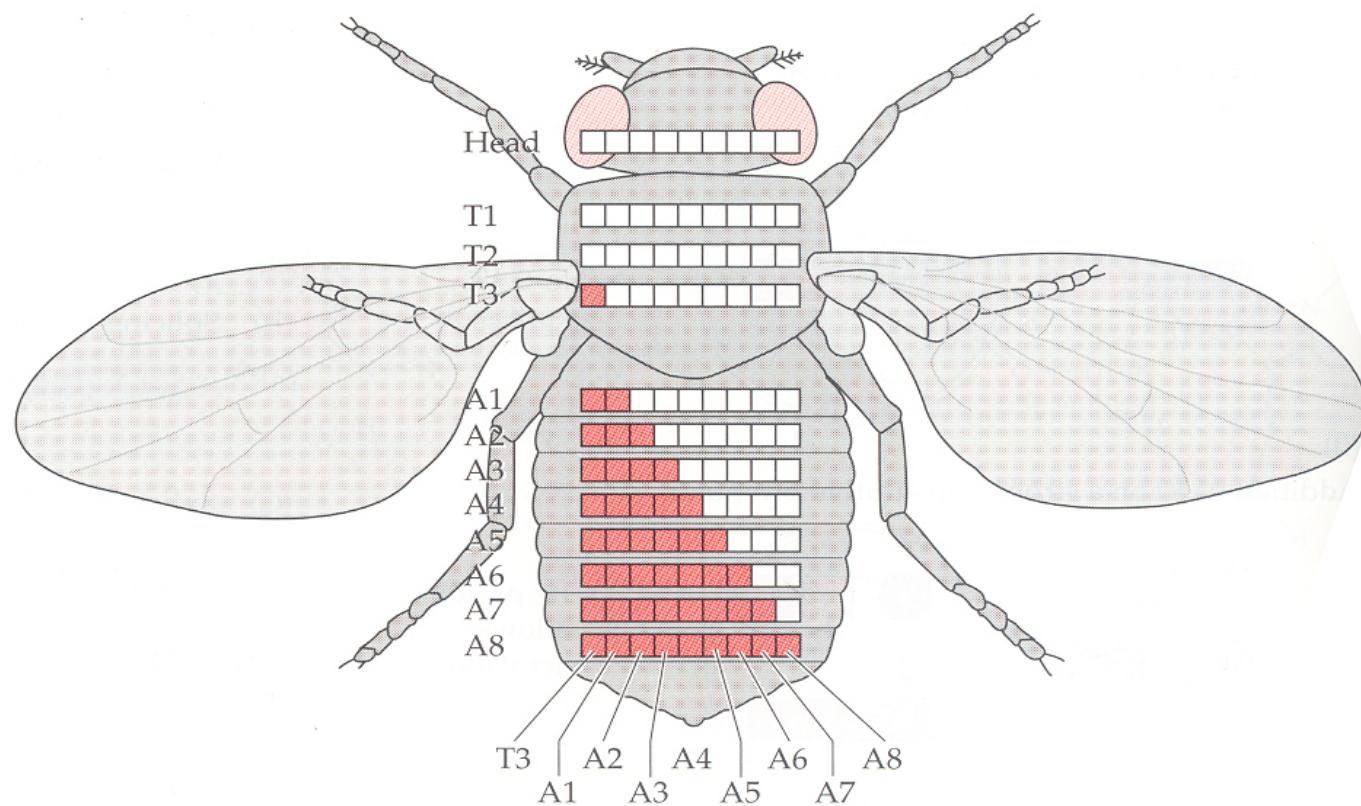
Homeotic transformations: change of developmental fate



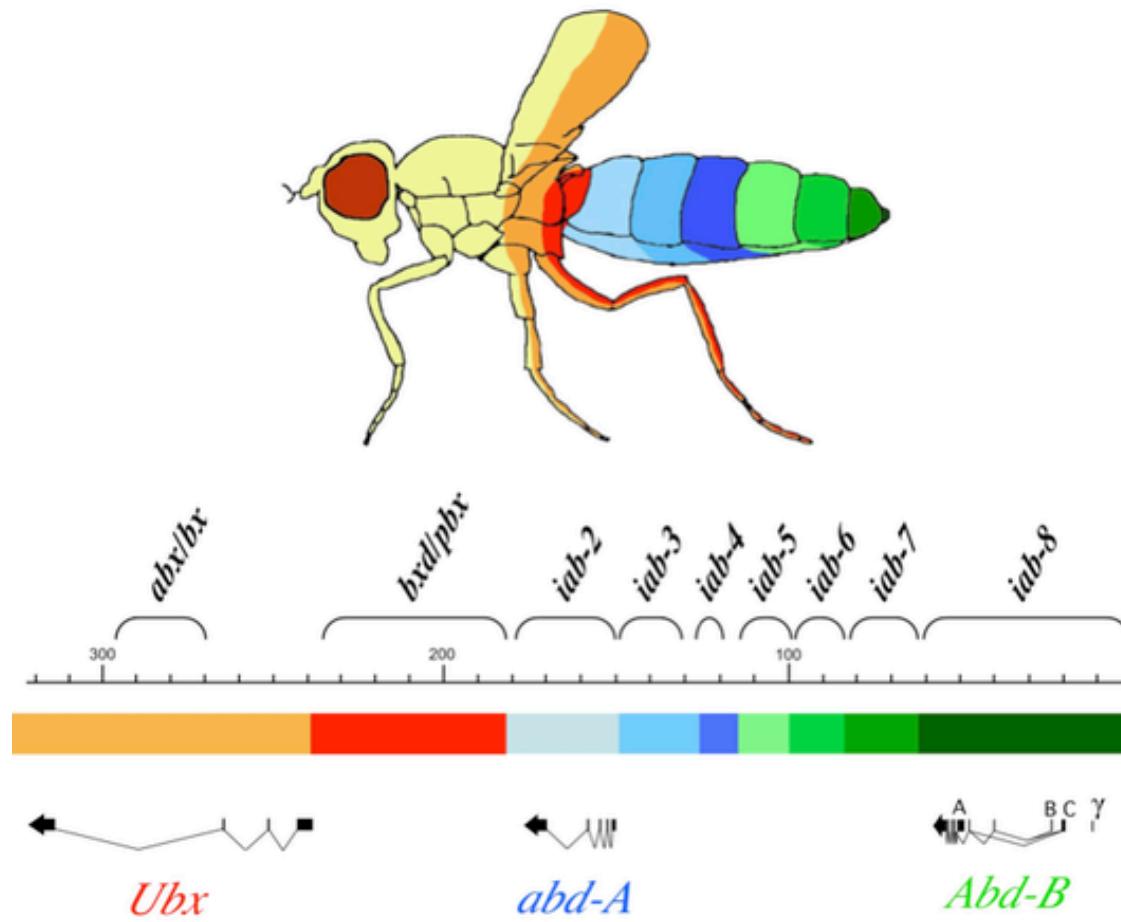
Homeotic transformations: change of developmental fate



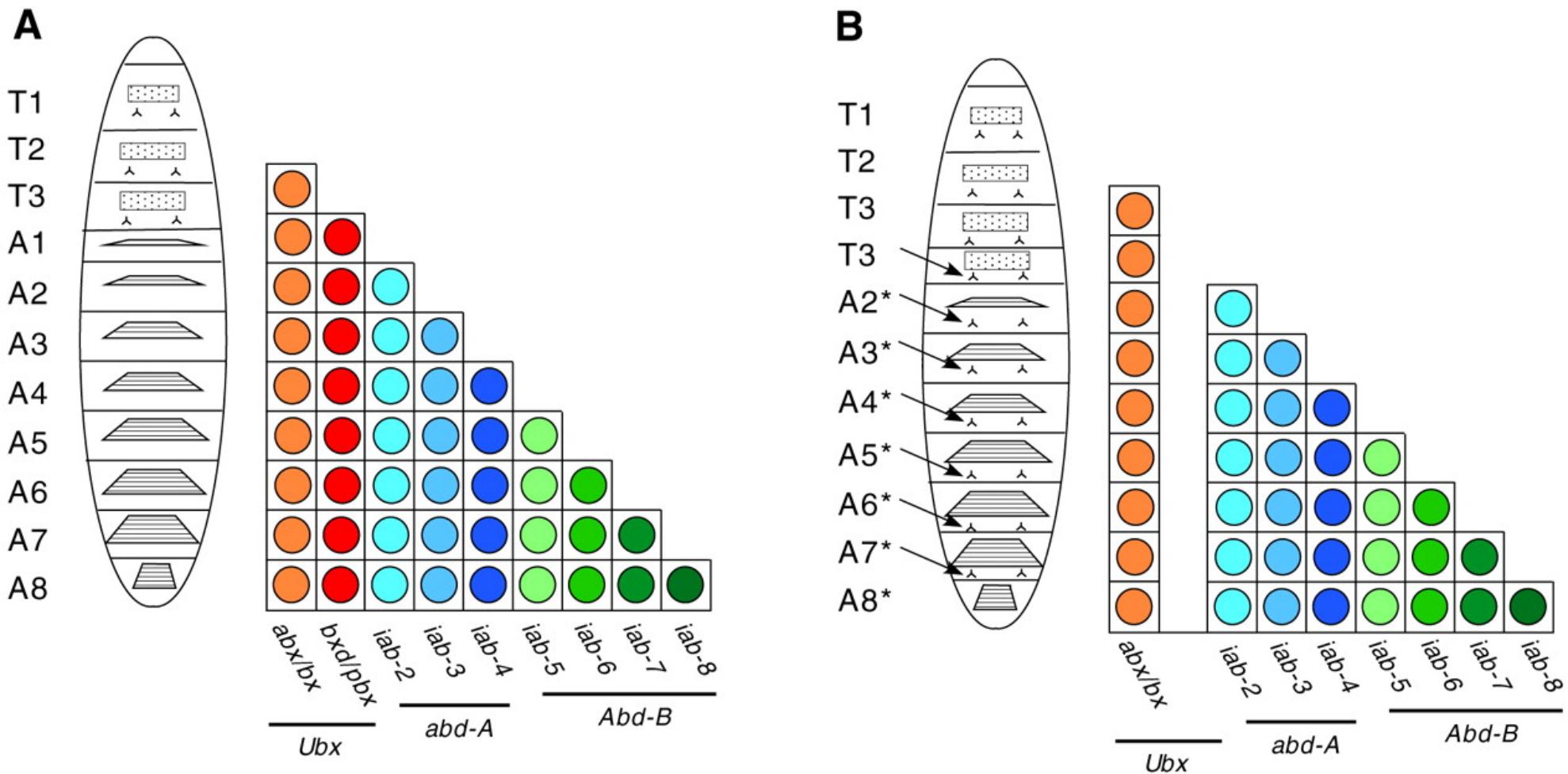
Lewis model



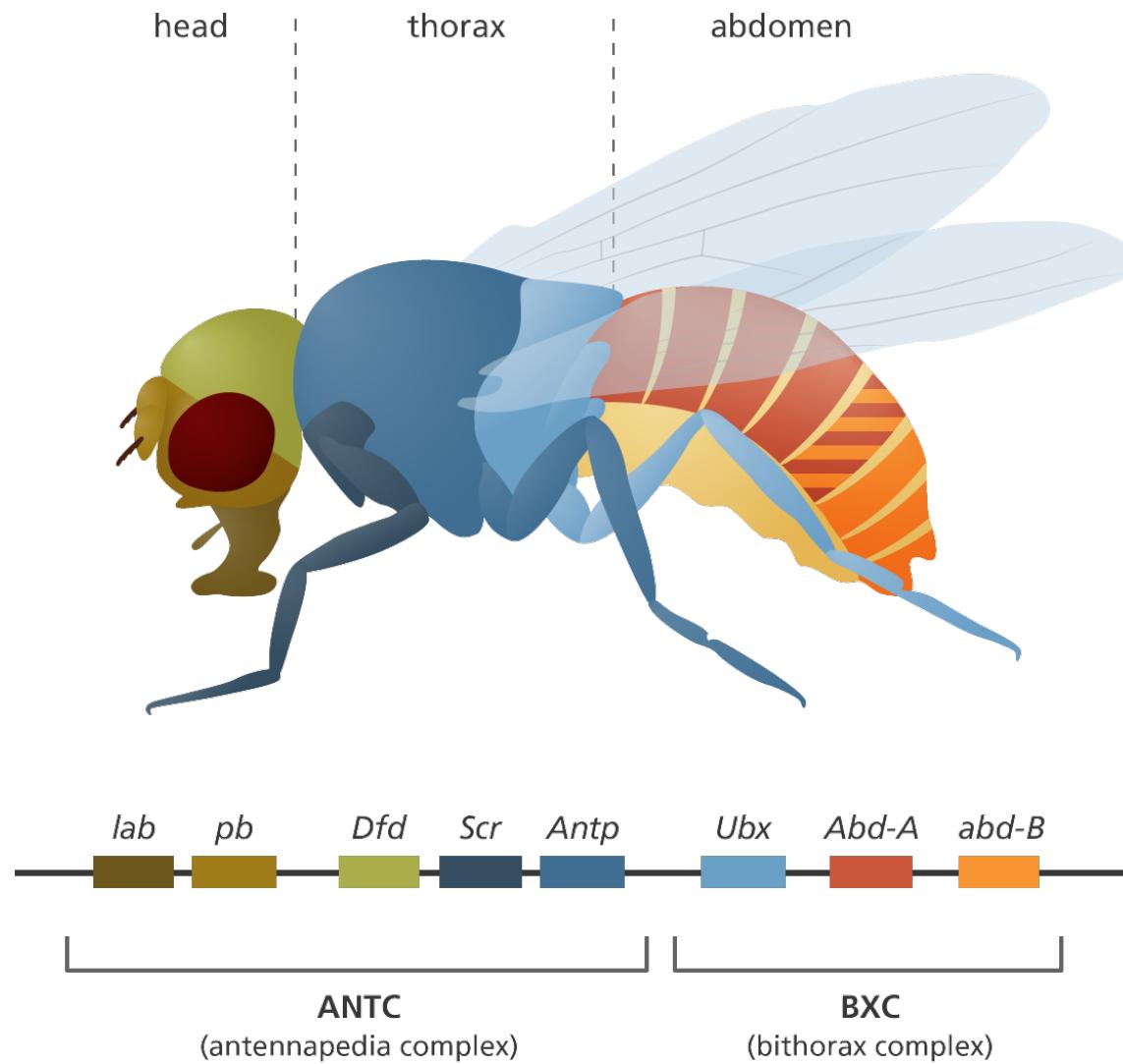
Lewis model



Lewis model



Homeotic genes are organized in two complexes



Cloning Antennapedia

The EMBO Journal Vol.2 No.11 pp.2027 – 2036, 1983

Genomic and cDNA clones of the homeotic locus *Antennapedia* in *Drosophila*

Richard L.Garber¹, Atsushi Kuroiwa and
Walter J.Gehring*

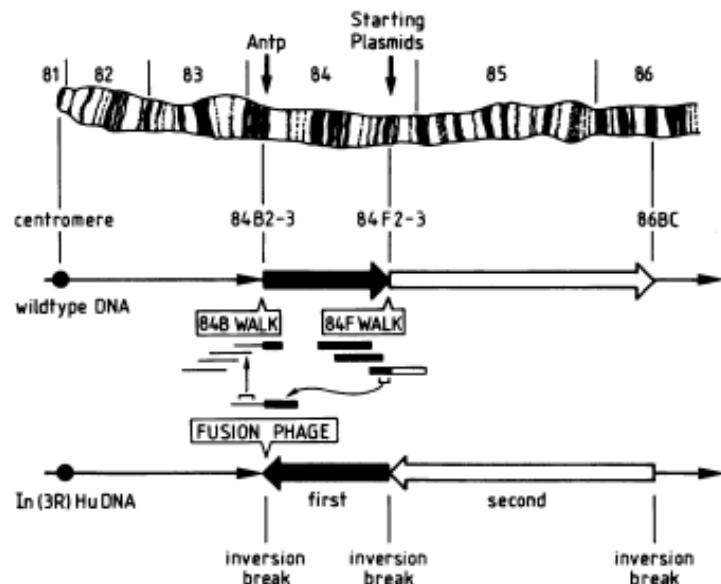


Fig. 1. Strategy for cloning *Antennapedia*. Starting with plasmids mapping to 84F1-2, a chromosome walk yielded overlapping DNA segments until the 84F2-3 breakpoint for the inversion *In(3R)Hu* was reached. A probe from the 84F clone just to the left of the breakpoint was used to select hybrid phage from a library constructed from homozygous *In(3R)Hu* DNA. A new probe derived from the 84B portion of a fusion phage was then used to select 84B clones. From these a chromosome walk in 84B was initiated until *Antp* was reached. The upper drawing represents the banding pattern of the proximal portion of chromosome 3R. The orientation of two stretches of DNA are shown in the wild-type configuration (upper) and in the doubly-inverted *In(3R)Hu* configuration. The three *In(3R)Hu* breakpoints are marked at the bottom.

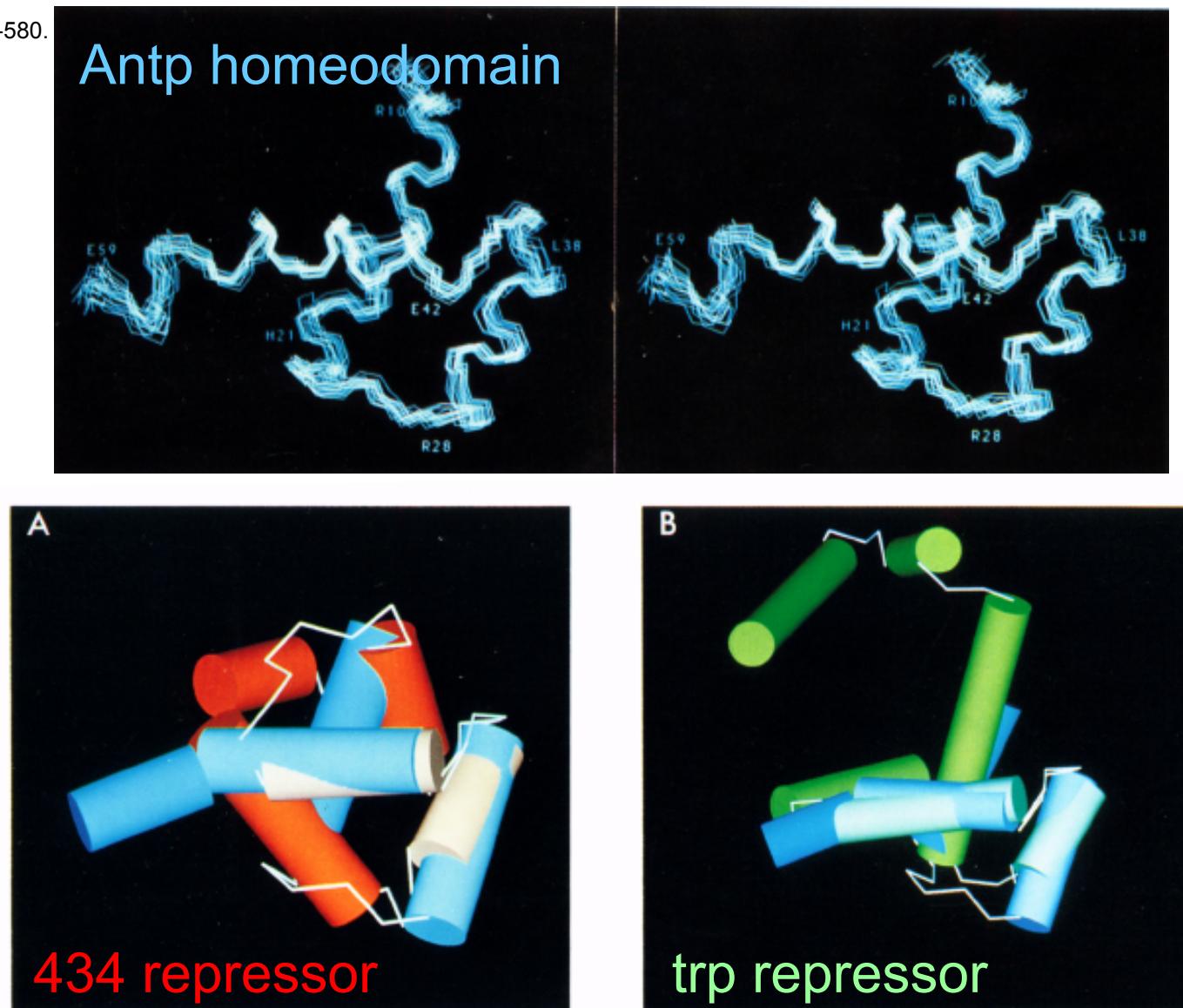
Cell, Vol. 35, 763–776, December 1983 (Part 2). Copyright © 1983 by MIT

The Molecular Organization of the *Antennapedia* Locus of *Drosophila*

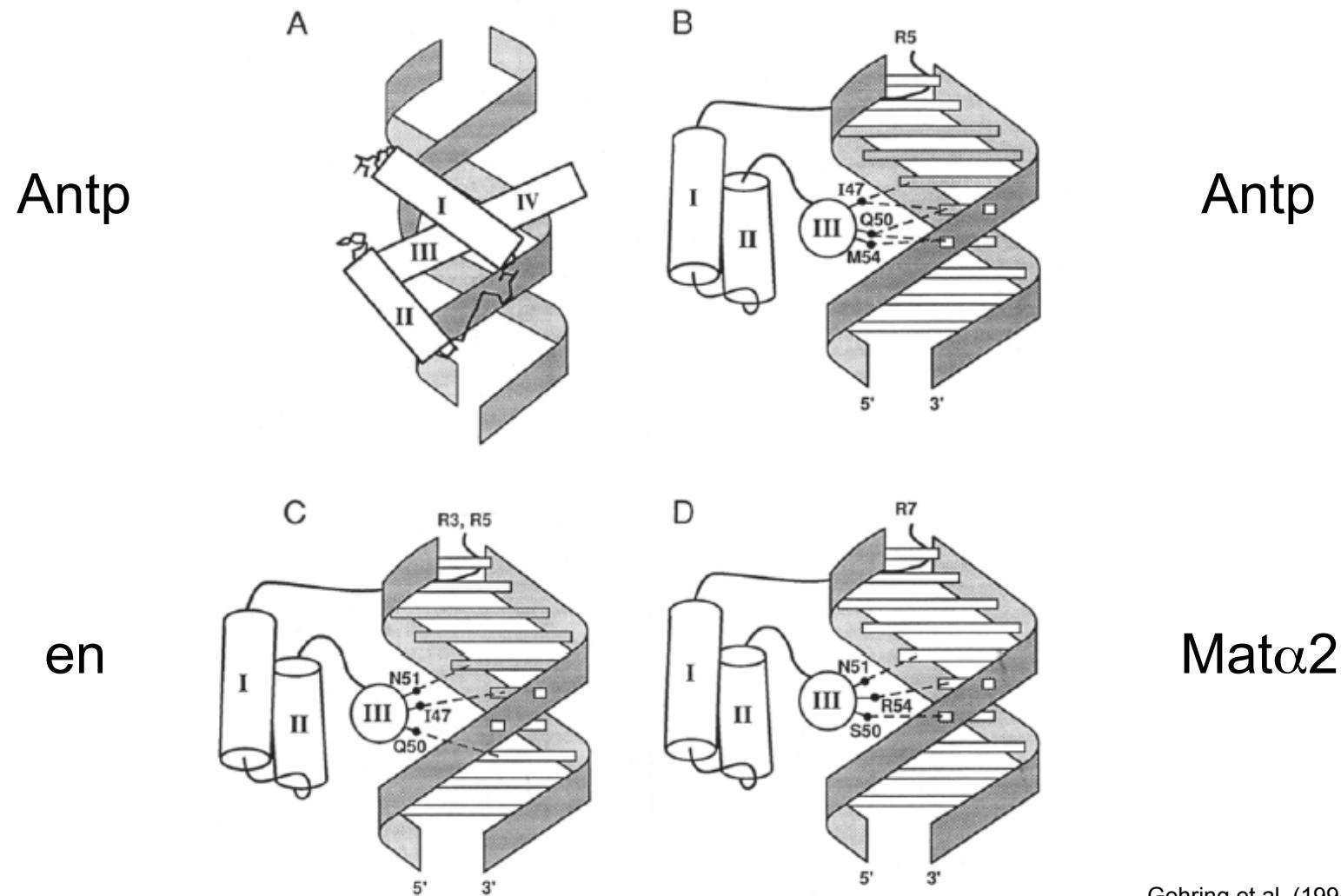
Matthew P. Scott,^{**} Amy J. Weiner,^{*}
Tulle I. Hazelrigg,^{**} Barry A. Polisky,^{*}
Vincenzo Pirrotta,[†] Franco Scalenghe,[†] and
Thomas C. Kaufman^{*}

The Antennapedia homeodomain

Qian et al. (1989) Cell 59, 573-580.



Homeodomain binding specificity

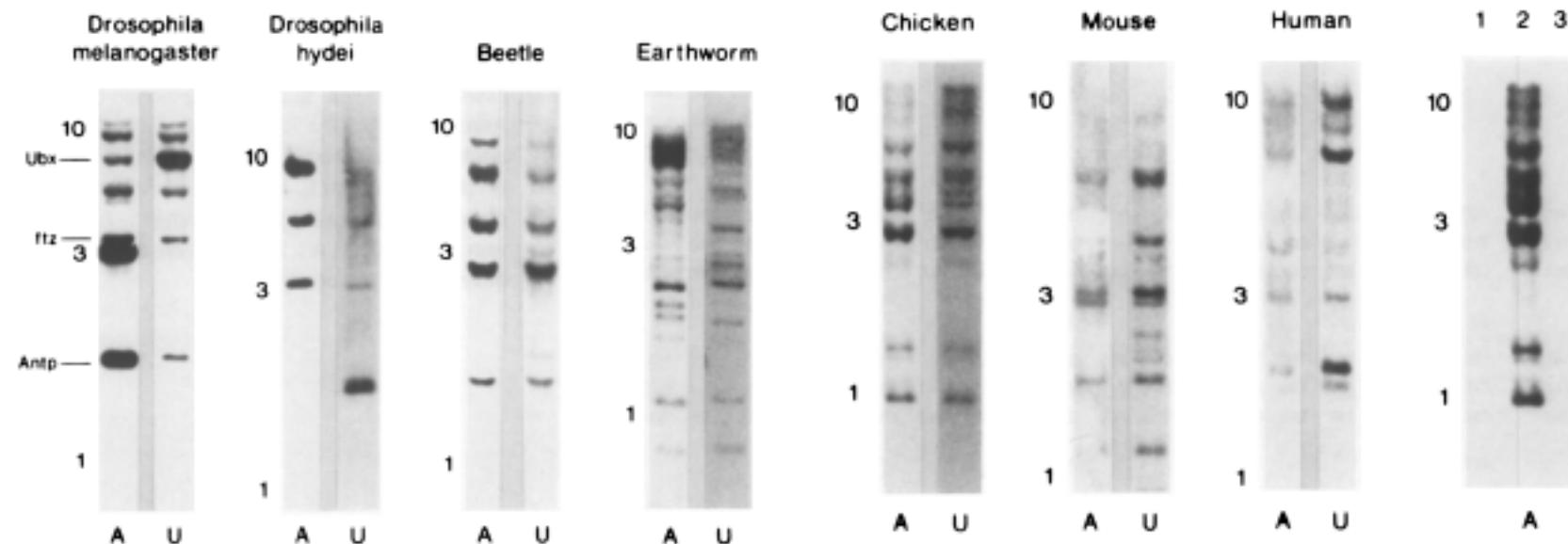


The homeobox: a highly conserved sequence

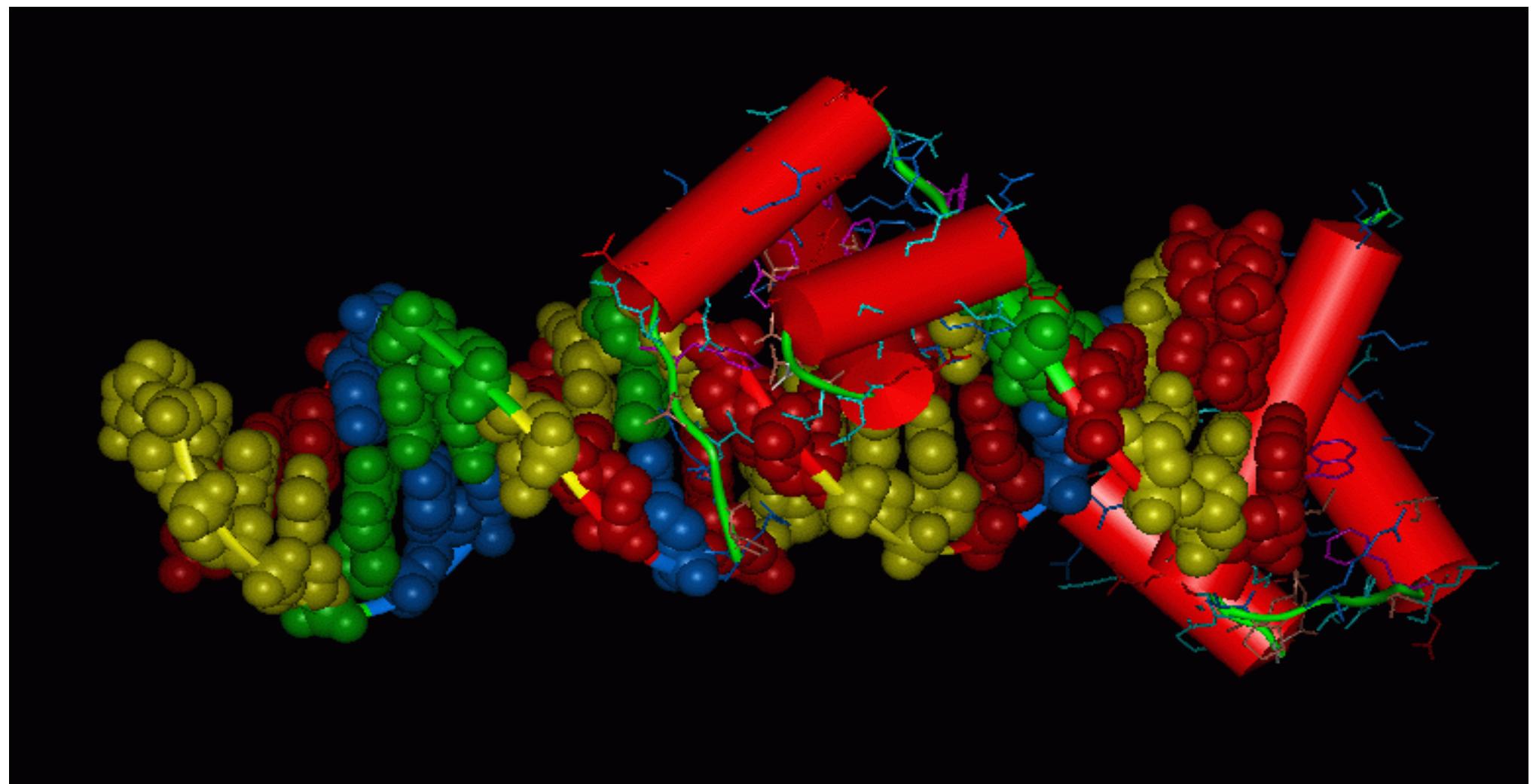
Cell, Vol. 37, 403-408, June 1984, Copyright © 1984 by MIT

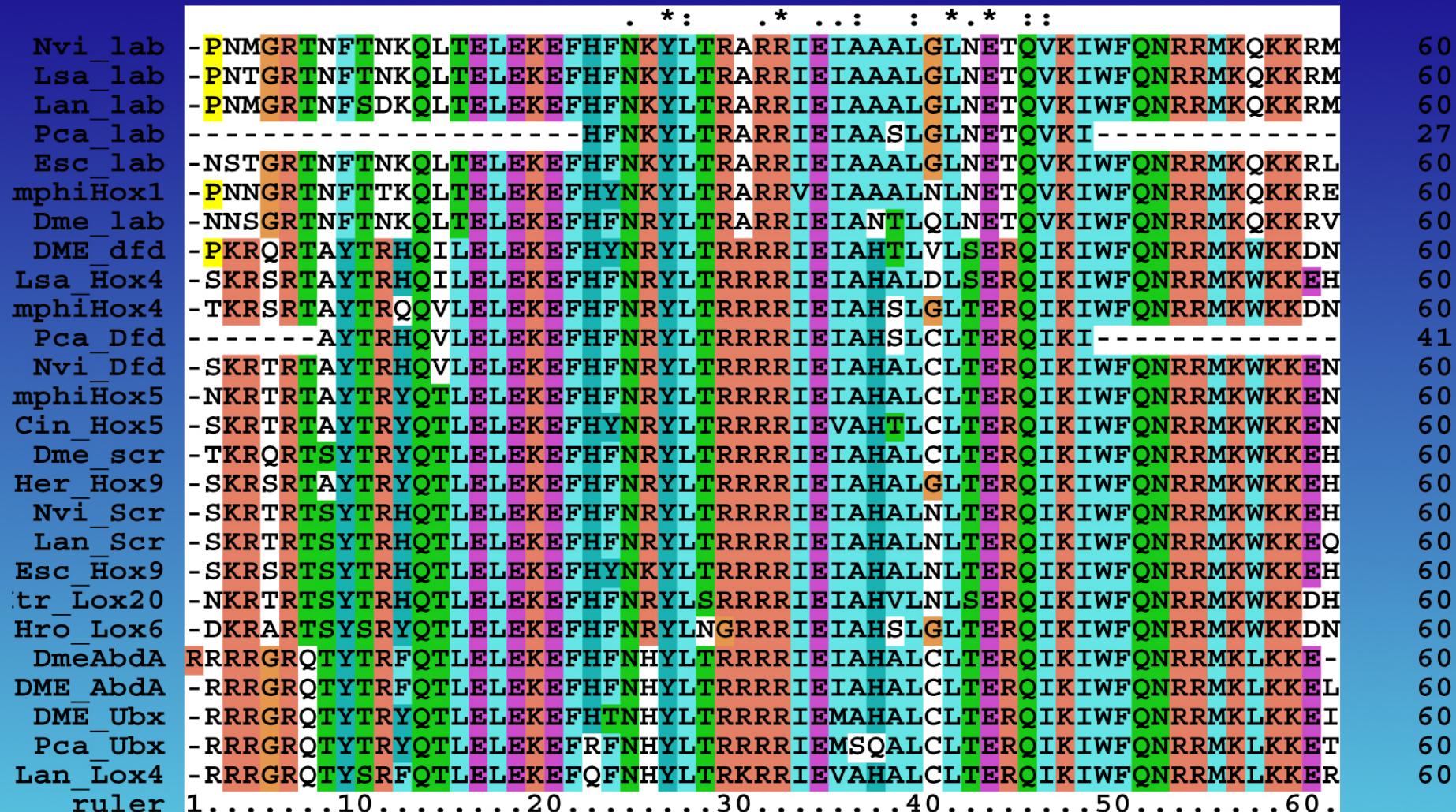
A Homologous Protein-Coding Sequence in *Drosophila* Homeotic Genes and Its Conservation in Other Metazoans

William McGinnis, Richard L. Garber,*
Johannes Wirz, Atsushi Kuroiwa, and
Walter J. Gehring

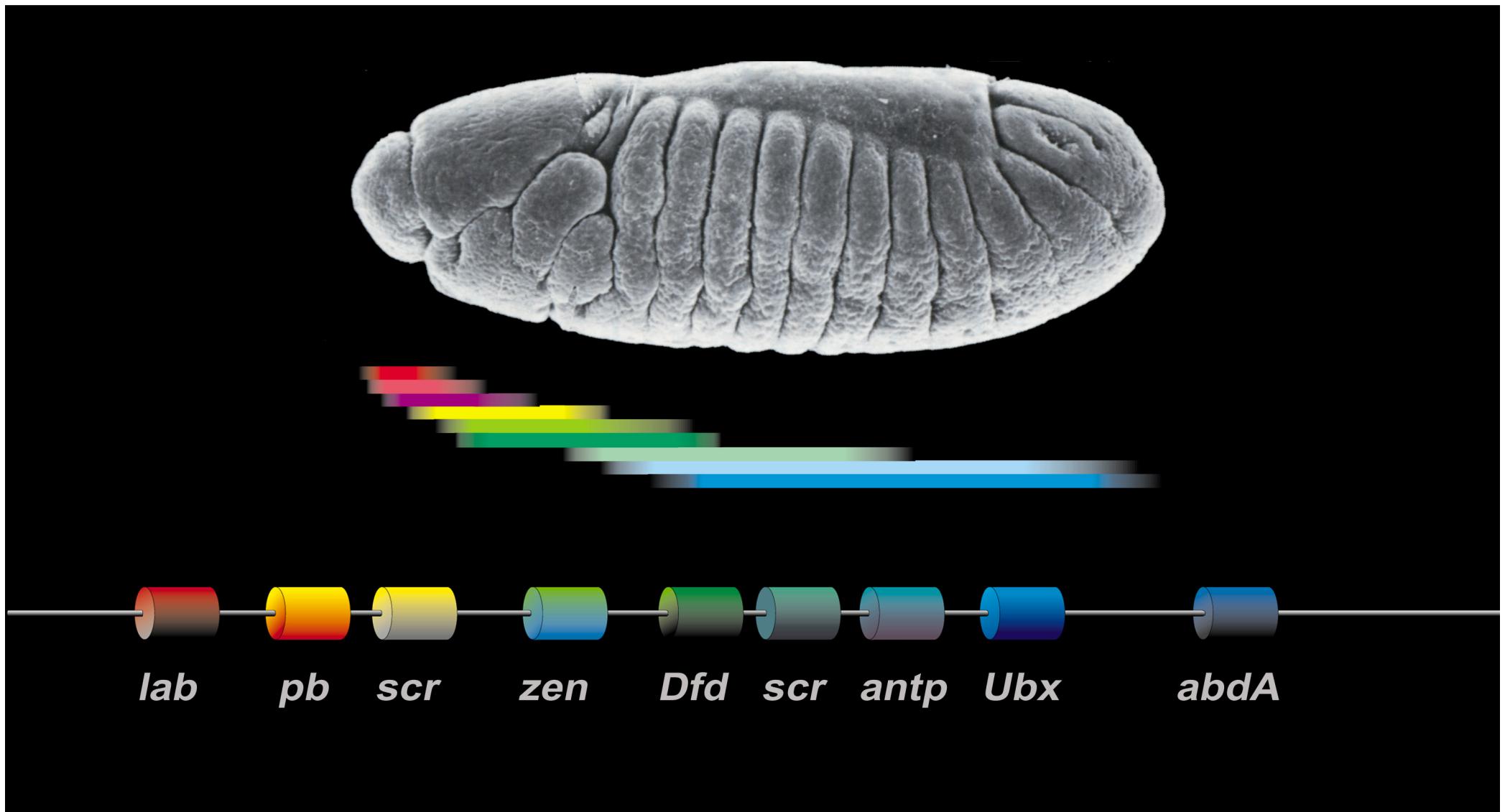


The homeodomain: a conserved DNA binding motif shared among a conserved family of transcription factors

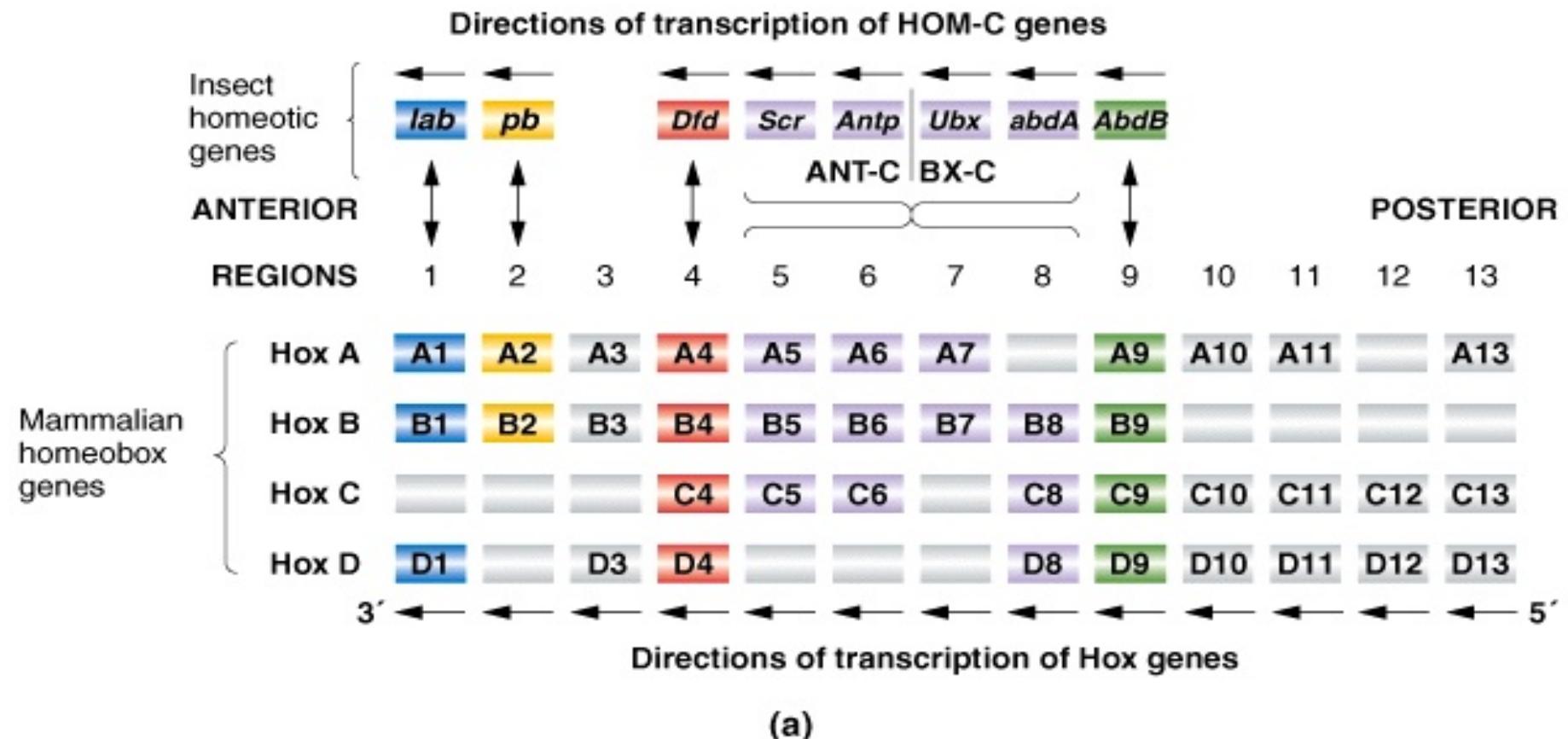




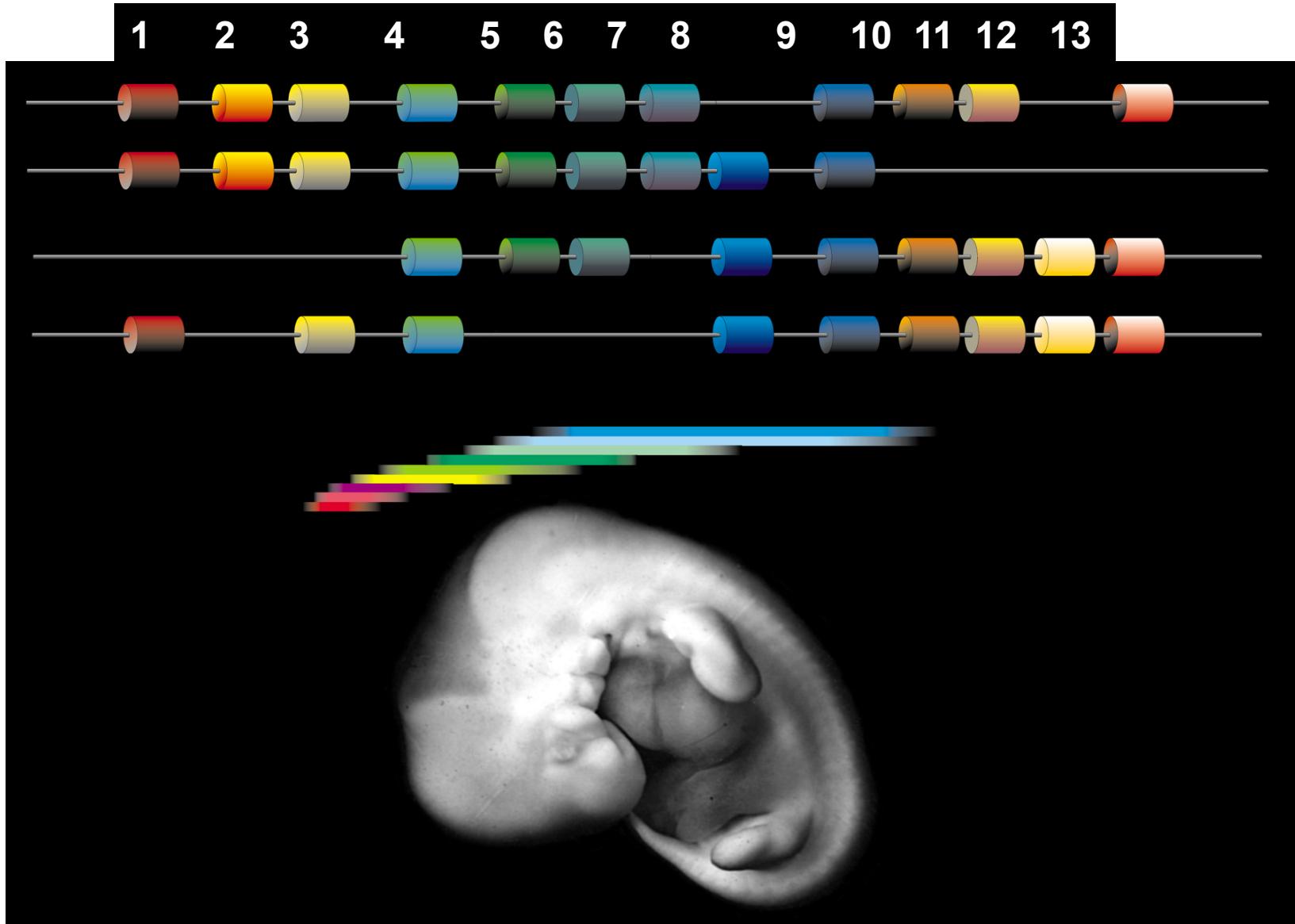
Colinear HOX gene expression in the Drosophila embryo



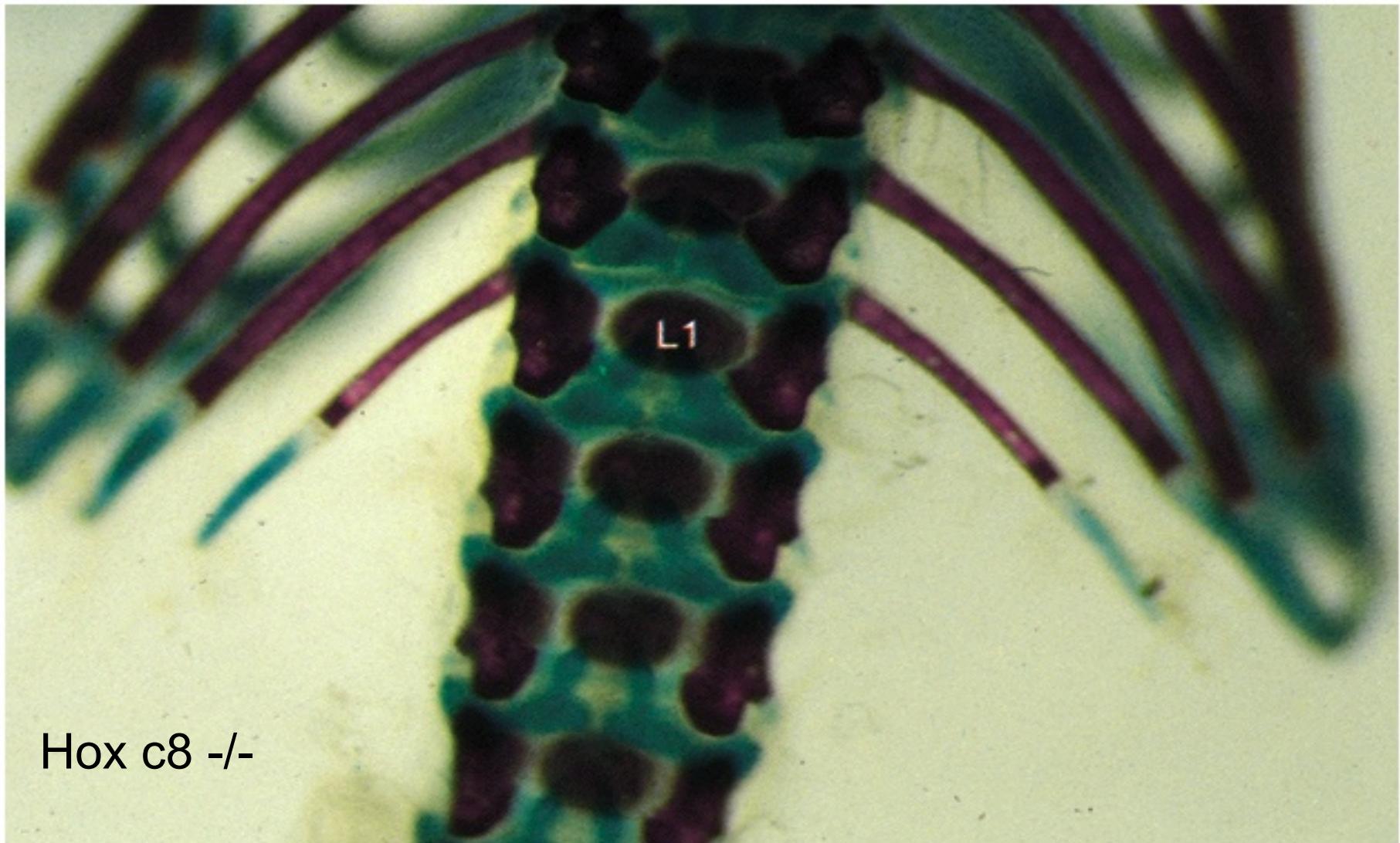
Mammalian Hox clusters



Colinear HOX gene expression in the mammalian embryo

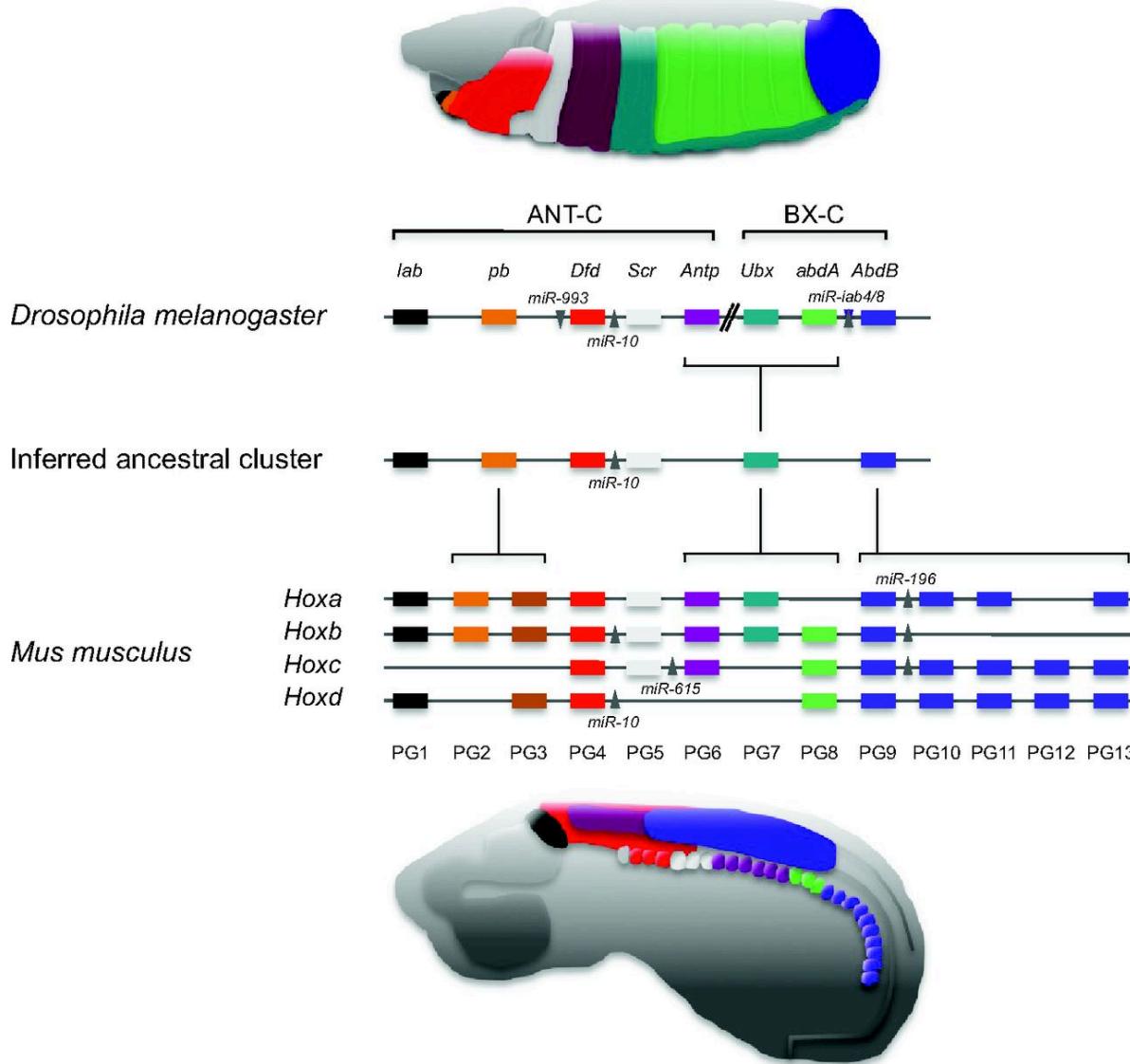


Homeotic transformation in the mammalian embryo



Hox c8 -/-

Hox genes: an ancient system for body patterning



REPORT

Mutations in *HOXD13* Underlie Syndactyly Type V and a Novel Brachydactyly-Syndactyly Syndrome

Xiuli Zhao,* Miao Sun,* Jin Zhao, J. Alfonso Leyva, Hongwen Zhu, Wei Yang, Xuan Zeng, Yang Ao, Qing Liu, Guoyang Liu, Wilson H. Y. Lo, Ethylin Wang Jabs, L. Mario Amzel, Xiangnian Shan, and Xue Zhang

The American Journal of Human Genetics Volume 80: 361-371

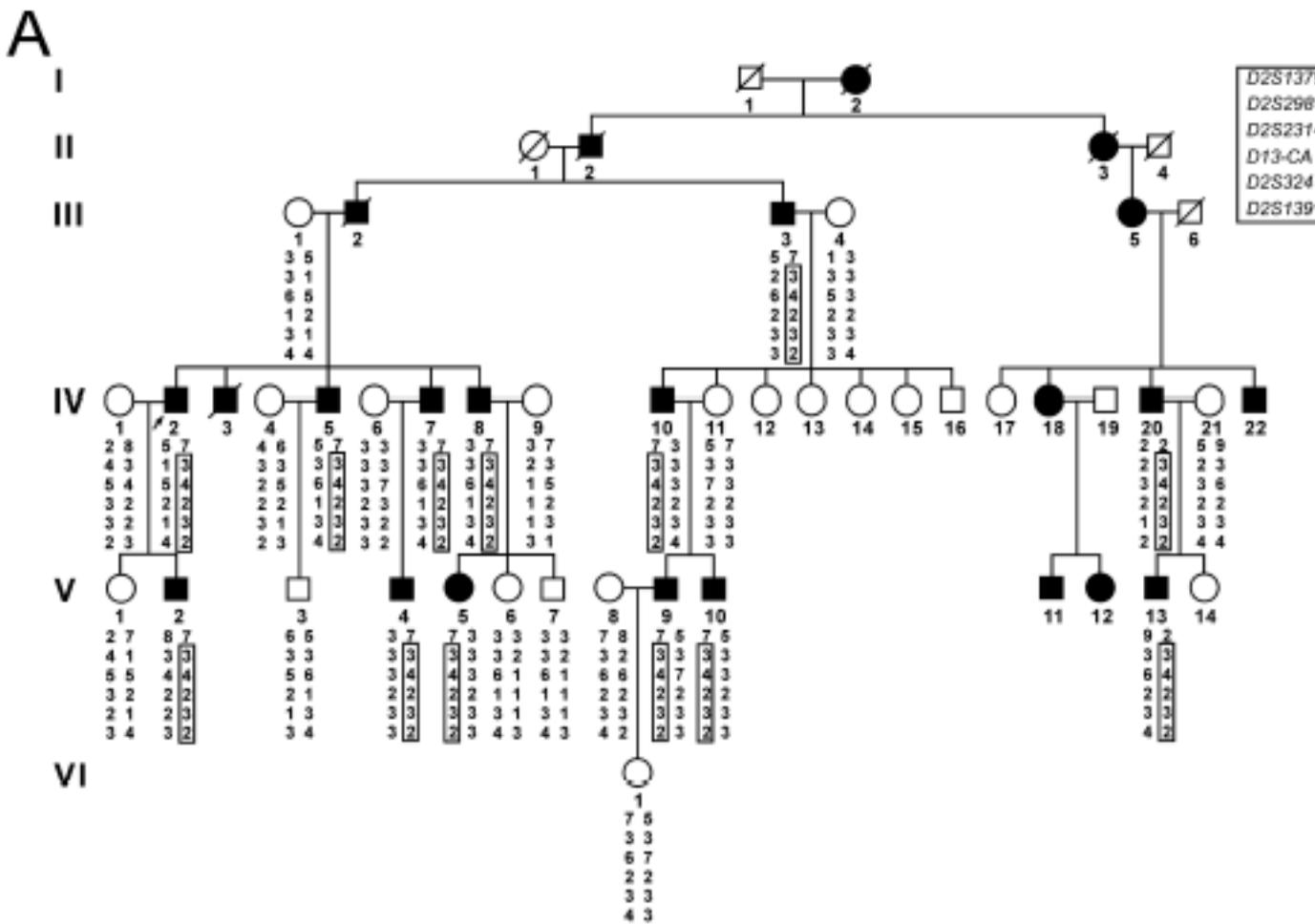


Figure 1. Pedigrees and disease-haplotype segregation of family 1 (A) and family 2 (B). Blackened symbols represent affected individuals with abnormal limb phenotype, and unblackened symbols represent individuals with a normal limb phenotype. Circles and squares indicate females and males, respectively. The arrows identify the probands. The disease haplotype is boxed.

B

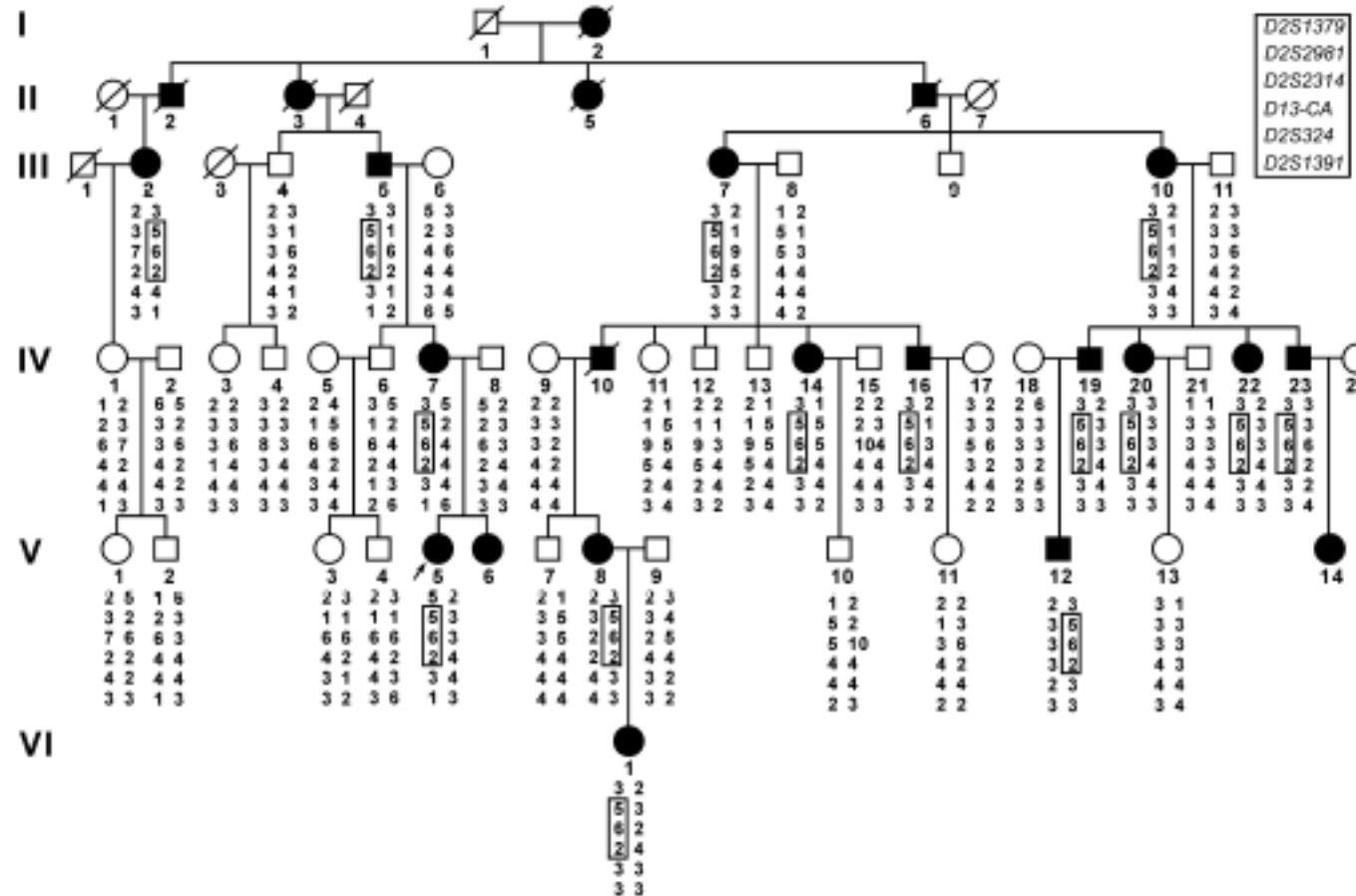


Figure 1. Pedigrees and disease-haplotype segregation of family 1 (A) and family 2 (B). Blackened symbols represent affected individuals with abnormal limb phenotype, and unblackened symbols represent individuals with a normal limb phenotype. Circles and squares indicate females and males, respectively. The arrows identify the probands. The disease haplotype is boxed.

BONES OF THE HAND AND WRIST

ANTERIOR VIEW



POSTERIOR VIEW



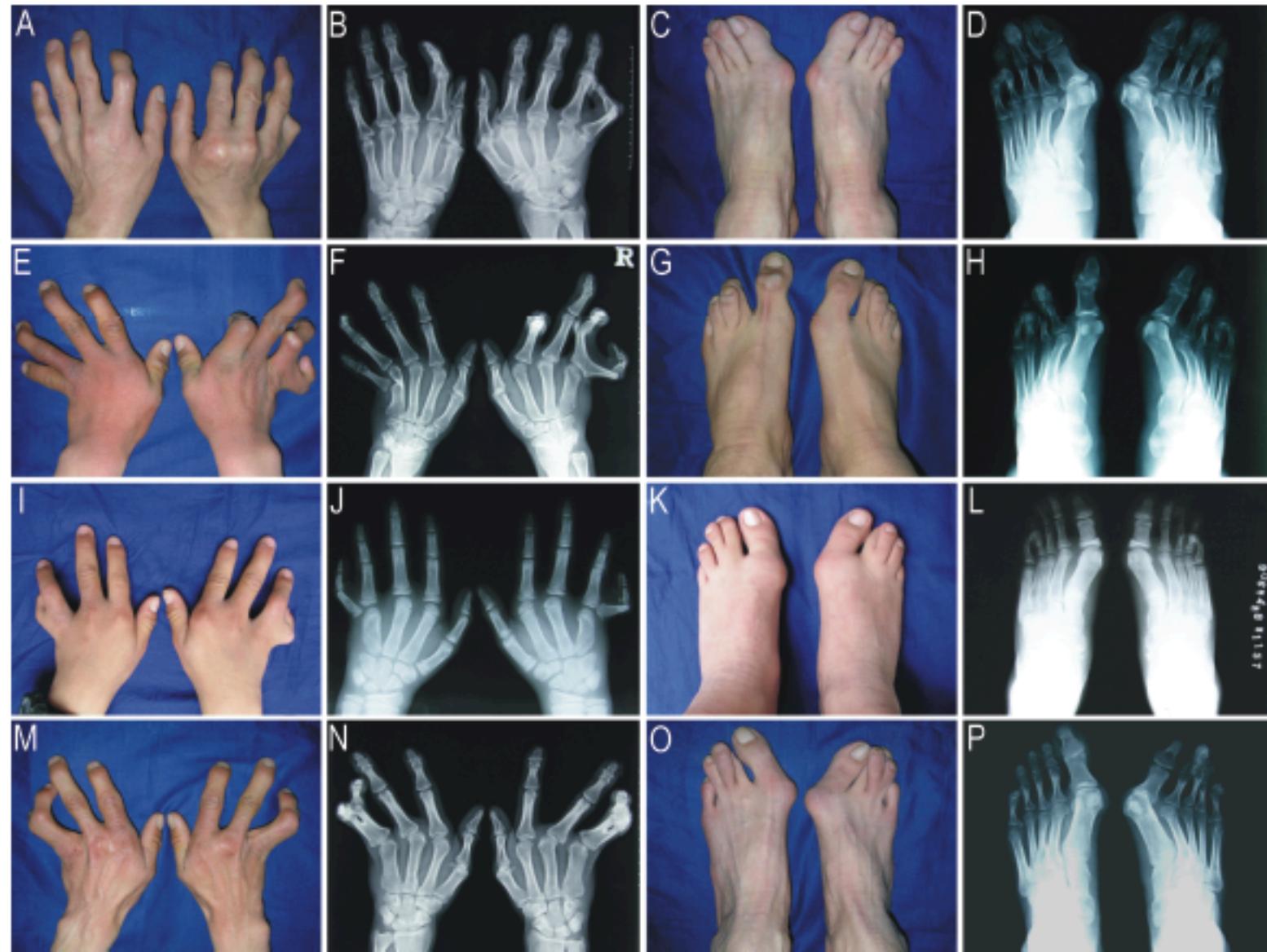


Figure 2. Photographs and radiographs of the proband (A-D), individual V-10 (E-H), individual V-13 (I-L), and individual IV-10 (M-P) of family 1.



Figure 3. Photographs and radiographs of the proband (A-D), individual III-10 (E-H), individual IV-19 (I-L), and individual IV-23 (M-P) of family 2.

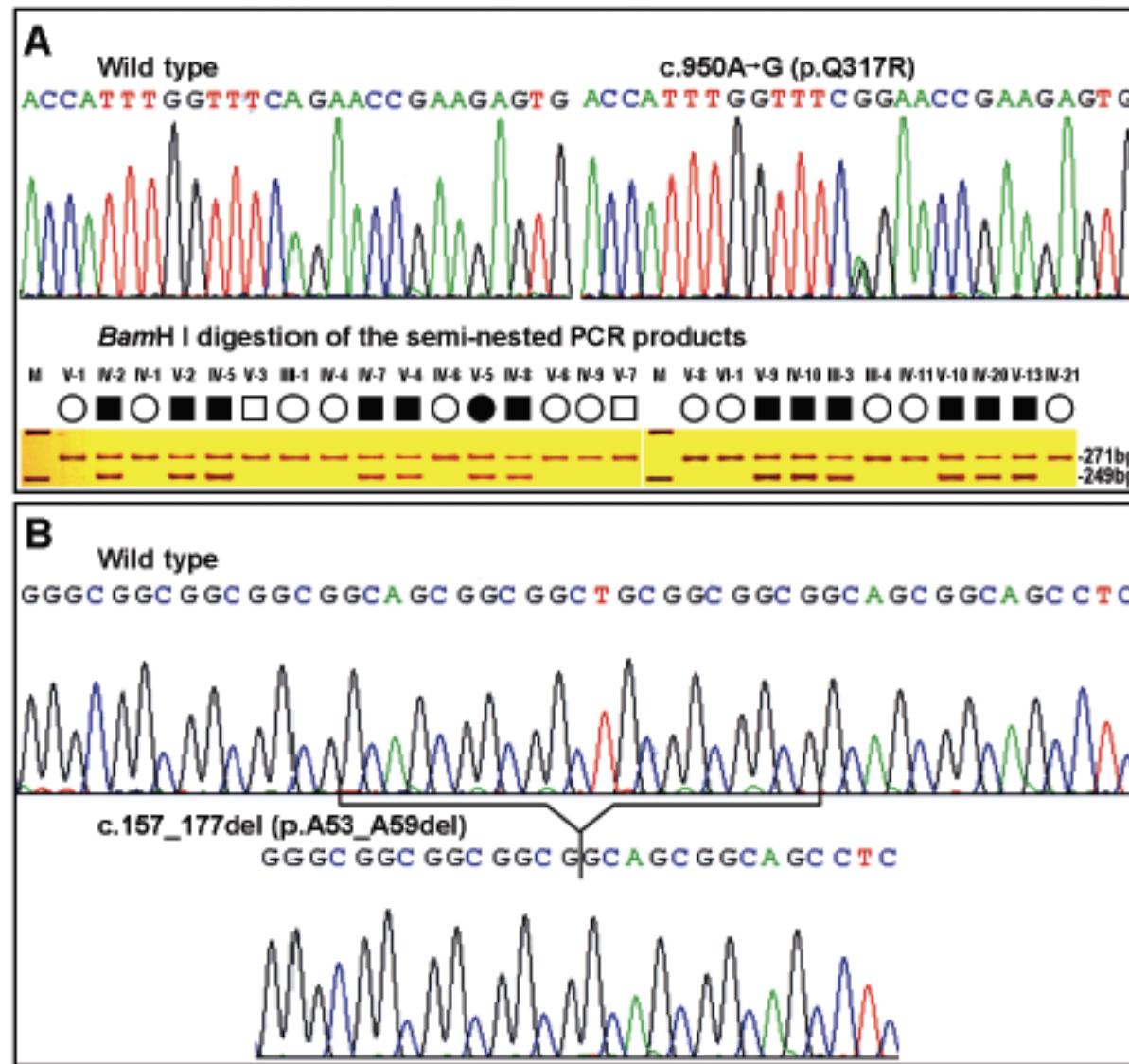


Figure 4. HOXD13 missense and polyalanine contraction mutations in study families.

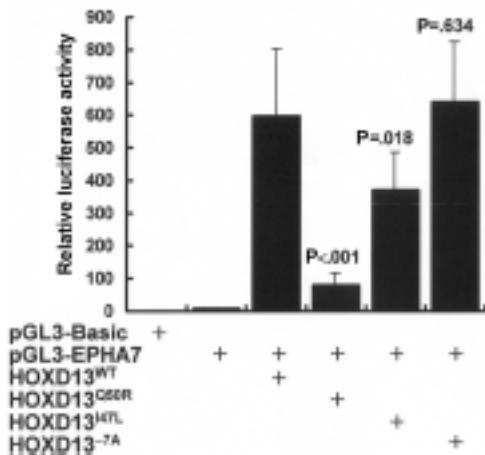
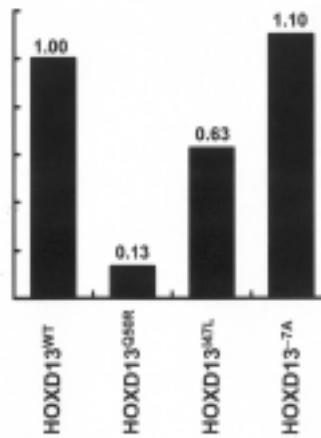
A**B****C**

Figure 5. Transcriptional activity of wild-type and mutant HOXD13 proteins at the human EPHA7 promoter.

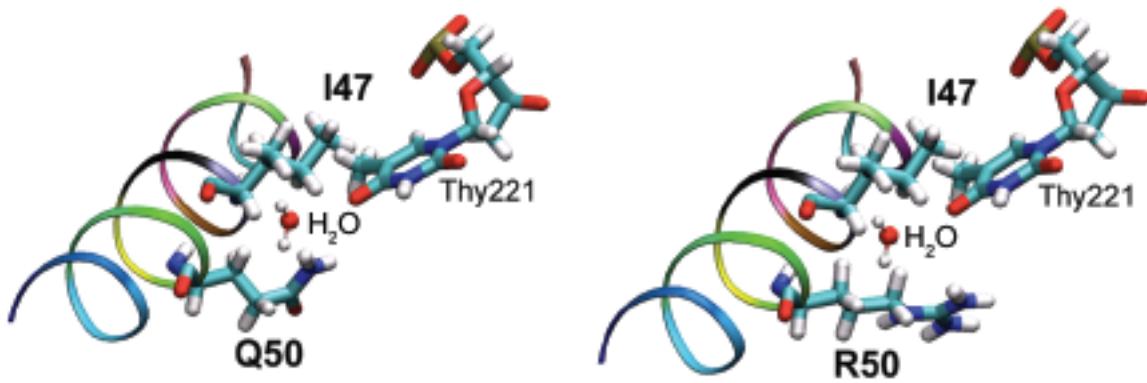
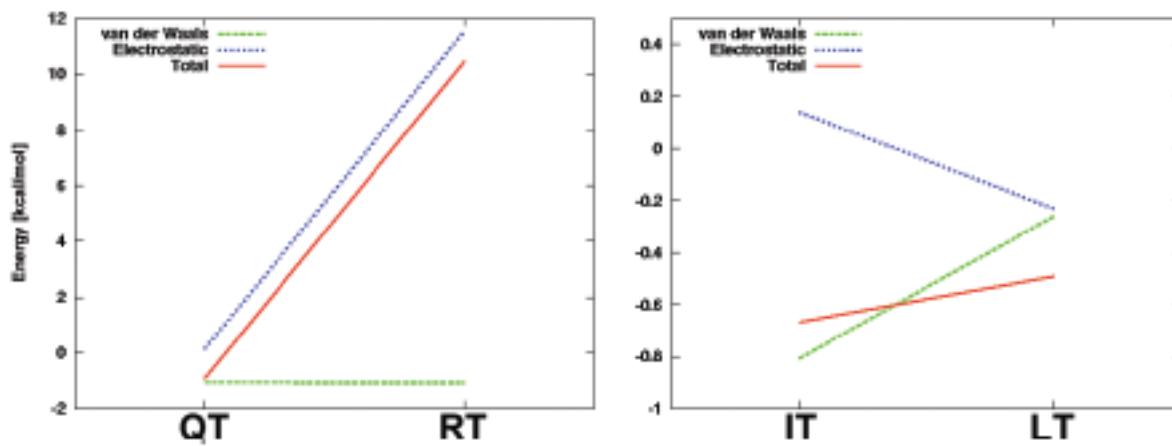
A**B**

Figure 6. Interaction between HOXD13 and DNA.