ters that contain water molecules that act as "double donors" [see figure 2 of Pribble and Zwier (2)]. This is consistent with theoretical calculations for the larger water clusters, which predict that the lowest energy structures are no longer the ring structures formed entirely from single-donor water molecules [again, see figure 2 in (2)], but also include double-donor waters.

Understanding the fundamental interactions between a molecule and water is the cornerstone of our understanding of aqueous solutions. The observations and conclusions drawn by Pribble and Zwier (2), if they prove to be correct, add new insights into our understanding of the forces between benzene and water, a classic example of a hydrophobic interaction. This work also illustrates the synergism that has developed between experimental and theoretical studies in modern chemical physics. The information from each is in some measure incomplete; but combined, they can provide definitive answers to long-standing chemical questions. The experimental studies of Pribble and Zwier will likely stimulate theoretical studies of benzene-water clusters to provide further validation of and insight into the conclusions drawn by these authors.

References

1. S. S. Xantheas and T. H. Dunning Jr., *J. Chem. Phys.* **98**, 8037 (1993); *ibid.* **99** 8774 (1993).
2. R. N Pribble and T. S. Zwier, *Science* **265**, 75 (1994).
3. G. Scoles, Ed., *Atomic and Molecular Beam Methods*, vol. 1 (Oxford Univ. Press, New York, 1988).
4. R. E. Smalley, L. Wharton, D. J. Levy, *Chem. Phys.* **66**, 2750 (1977).
5. W. R. Bosenberg and D. R. Guyer, *J. Opt. Soc. Am. B* **10**, 1716 (1993).
6. R. H. Page, Y. R. Shen, Y. T. Lee, *J. Chem. Phys.* **88**, 4621 (1988).

Genes on the Wing

H. Frederik Nijhout

Discovering the mechanisms by which structure emerges from an initially homogeneous field of cells continues to be one of the fundamental problems in developmental biology. There are many such mechanisms—generally collected under the term "pattern formation." Among the problems in the pattern formation field are such disparate questions as: What determines the location where gastrulation will begin in a spherical embryo, the points where limb buds will develop along the flank of an embryo, the pattern of five bones in an initially featureless paddle-shaped hand primordium, the division of a cylindrical embryo into segments, or the placement of an eyespot on the blank, uniform epithelium of a butterfly wing? Occasionally structure is organized around irregularities that are set up by external events such as fertilization, gravity, or graded distributions of chemicals that the mother incorporates into an egg cell, but in many cases such singularities or prepatterns are unknown. The mechanisms of pattern formation are generally believed to require interactions among molecules that specify the fates of cells. Some of these molecules are the direct products of genes, others alter the expression of genes, while yet others are metabolites, the products of biochemical reactions.

Recent years have witnessed a revolution in our ability to detect and visualize the distribution of gene products in the tissues of developing organisms. With these new eyes, we can see molecular markers of early developmental events that were previously invisible and inaccessible. Much of this work has focused on the early embryonic development of the fruit fly, *Drosophila melanogaster*, in which we now know more about the early molecular events of development than in any other species of plant or animal. For instance, we know that segmentation in the *Drosophila* embryo is preceded by a complex and dynamic pattern of gene expression, in which half a dozen or more genes become expressed in bands and rings whose positions indirectly define those of the future body segments (1). Segment specification is followed by an almost equally complex pattern of gene expression, different in each segment, that presages the development of the characteristic morphology of each segment. In the imaginal disks of *Drosophila* embryos and larvae, there are likewise patterns of gene expression that correspond to the future wing margin, appendage tips, and to dorsal, ventral, anterior, and posterior compartments (1, 2).

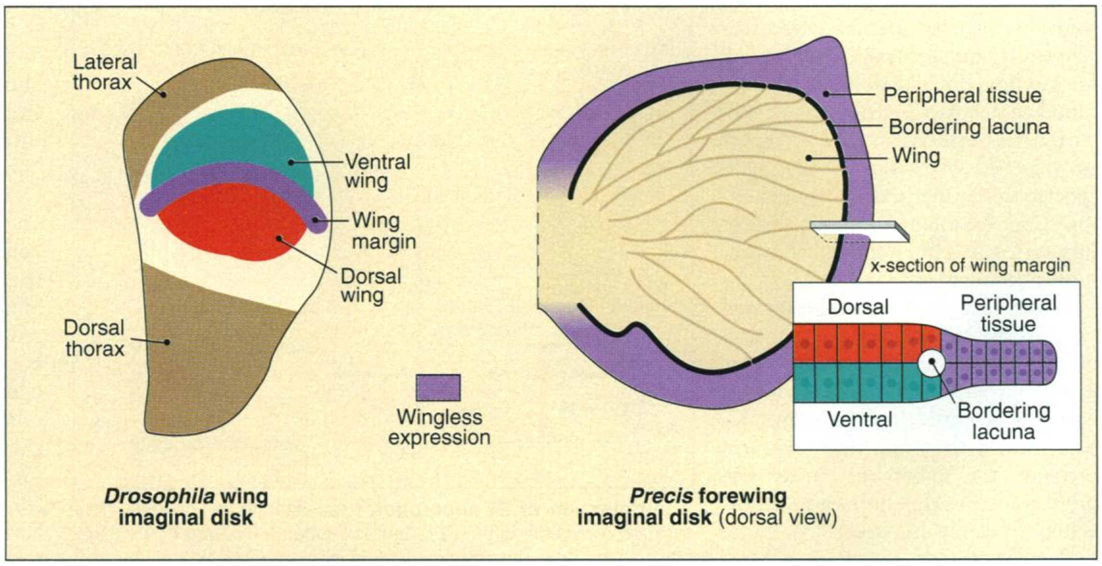

Fig. 1. Wing development in fruit flies and butterflies. In insects with complete metamorphosis, all adult appendages develop during larval life as internal imaginal disks. Imaginal disks of the higher Diptera, like *Drosophila*, are complex structures that undergo substantial morphogenesis in their metamorphosis to the adult forms. The wing imaginal disk, for instance, contains tissues that will make a large portion of the thorax, and the wing portion is folded open, so that the distal wing margin is represented by a line that runs in an arc across the center of the disk. The wing imaginal disk of butterflies is much simpler, consisting of two flat monolayers of cells, appressed to each other, which will form the dorsal and ventral surfaces of the wing. In butterflies, only the central portion of this epithelial sheet will form the adult wing, while the peripheral tissue undergoes programmed cell death during metamorphosis.

This treasure trove of molecular markers for early developmental events could only have been developed in a species such as

---

The author is in the Zoology Department, Duke University, Durham, NC 27708–0325, USA.

Drosophila, where the genetics are extensive and rich. Fortunately, it is now also possible to use *Drosophila* sequences to visualize the expression of homologous genes in other animals. In a paper in this issue of *Science*, Carroll and co-workers (3) report a series of exciting new results that describe expression of homologs of the patterning genes for *Drosophila* imaginal disks in the developing wings of the butterfly *Precis coenia*. Some of these genes are expressed in patterns similar to those in *Drosophila*, suggesting ancient and conserved developmental mechanisms. Others are expressed in unique, new patterns that predict the subsequent development of the color pattern of the adult butterfly, and these provide one of the nicest examples to date of how the application of "*Drosophila* technology" to other species can lead to the discovery of new roles for old genes.

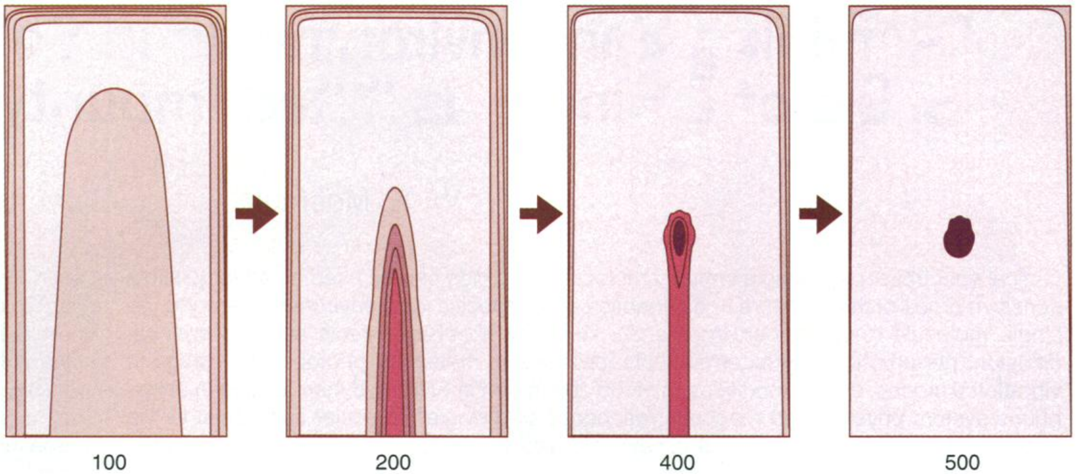

Fig 2. Computer simulation of wing development in butterflies. A reaction-diffusion model for color pattern formation on butterfly wings (*9, 10*), in which the rectangle represents a single wing cell (the area bounded by wing veins) and the colors represent the concentration of a reactant.

In spite of the dramatic difference in morphology of the wing disks of *Drosophila* and *Precis* (Fig. 1), topologically equivalent portions of the disks express the same kinds of genes. The *apterous* gene, for instance, is expressed only in the cells that will become the dorsal surface of the wing, even though the anatomical location of the dorsal cells differs significantly in the two species. In the *Drosophila* imaginal disk, these cells lie in the same plane as those that will form the ventral surface, while in *Precis* they are on opposite faces of the disk. The *scalloped* and *decapentaplegic* genes have similar patterns of expression in both species. The *invected* gene, a close relative of *engrailed*, is expressed only in the posterior compartment of the wing disk in both species.

Some genes, however, have unique patterns of expression in *Precis*. In late larvae of *Drosophila*, *wingless* is expressed in a narrow arc along the presumptive wing margin in the middle of the wing disk; in butterflies it is expressed in all the peripheral cells that will not become part of the wing but that undergo programmed cell death during metamorphosis. In *Drosophila*, *wingless* expression is not associated with cell death (*4*), but the *wingless* gene product is required to restrict the expression of *apterous* to the dorsal wing surface and is involved in specifying the proximo-distal axis of the disk and the margin of the future wing (*4–6*). The wing margins in the two species are anatomically and developmentally very different (*6, 7*), and it will be interesting to determine whether ontogeny of the margin through cell death of the peripheral tissues is a newly derived evolutionary feature in the Lepidoptera or whether it is primitive and has been lost in the Diptera.

The most interesting patterns of expression are those of *Distal-less*. In *Drosophila*, *Distal-less* marks the embryonic primordium of imaginal disks and is also expressed in the portions of the larval disks that will form the most apical structures (*2*). In *Precis* larval disks, *Distal-less* marks the center of a presumptive eyespot in the wing color pattern. The cells at this center act as inducers or organizers for development of the eyespot: If these cells are killed, no eyespot develops; if they are excised and transplanted elsewhere on the wing, they induce an eyespot to develop in an ectopic location around the site of implantation (*8, 9*). Carroll and co-workers (*3*) show that the pattern of *Distal-less* expression in *Precis* disks changes dynamically in the course of the last larval instar. It begins as broad, wedge-shaped patterns centered between wing veins. These wedges gradually narrow to lines, and a small circular pattern of expression develops at the apex of each line. This same progression of expression was predicted by a theoretical reaction-diffusion model for determination of the induction sources for color pattern formation (*9, 10*). In most cases of patterned gene expression during development, all the interesting events (from the viewpoint of pattern formation) are already over when the pattern is first detected, and interpretation of how the pattern of gene expression is regulated depends on interpretation of the altered patterns of various mutants. Having a dynamic pattern that closely matches the predictions of a theoretical model (Fig. 2) is relatively rare in developmental biology (although there are a number of good post hoc models that explain various aspects of patterned gene expression).

What remains to be explained is why only a single circle of *Distal-less* expression eventually stabilizes on the larval wing disks studied by Carroll and co-workers, while clearly two eyespots develop on the adult wings. One explanation may be that before a wing disk secretes the adult cuticle it must secrete a pupal cuticle whose pattern contains a single large pigmented spot at the location of the circle of *Distal-less* expression. This pupal spot is a precursor of the large posterior eyespot that develops on the adult wing. It will be interesting to see whether a later developmental stage of the *Distal-less* pattern will stabilize circles at the locations of both adult eyespots.

References

1. P. A. Lawrence, *The Making of a Fly* (Blackwell, London, 1992).
2. J. A. Williams and S. B. Carroll, *BioEssays* **15**, 567 (1993).
3. S. B. Carroll *et al.*, *Science* **265**, 109 (1994).
4. J. A. Williams, S. W. Paddock, S. B. Carroll, *Development* **117**, 571 (1993).
5. K. D. Janda, C. G. Shevlin, R. A. Lerner, *Science* **259**, 490 (1993).
6. J. P. Couso, S. A. Bishop, A. Martinez Arias, *Development* **120**, 621 (1994).
7. C. E. Dohrmann and H. F. Nijhout, *J. Res. Lepid.* **27**, 151 (1988).
8. H. F. Nijhout, *Dev. Biol.* **80**, 267 (1980).
9. _____, *The Development and Evolution of Butterfly Wing Patterns* (Smithsonian Press, Washington, DC, 1991).
10. _____, *Proc. R. Soc. London. Ser. B* **239**, 81 (1990).