TWO WINGS OR FOUR?

By turning a two-winged fly into a four-winged one, Caltech geneticists produce a working model for picturing the genetic control of development

by EDWARD B. LEWIS

BY TAMPERING WITH the genes of the tiny *Drosophila* fly we have constructed a four-winged fly at Caltech. Such a fly is really a contradiction in terms. As every school boy (who has had biology) knows, flies have only two wings. How then does a four-winged fly arise—and what is the significance of such a useless creature?

Be assured that the California Institute has not been under contract to develop and produce such monsters. Instead, the four-winged fly was a by-product of some basic studies of the nature of the hereditary material. Perhaps someday knowledge gained from such studies will help prevent the occurrence of similar kinds of monstrosities among human births.

Why do we use the fly in experiments on heredity? To perform such experiments we need to breed large numbers of individuals in a short time in a small space. The *Drosophila* fly admirably fulfills these conditions; a new generation appears every ten days, and a single pair can produce hundreds of offspring in a small culture bottle on an inexpensive food in a few days.

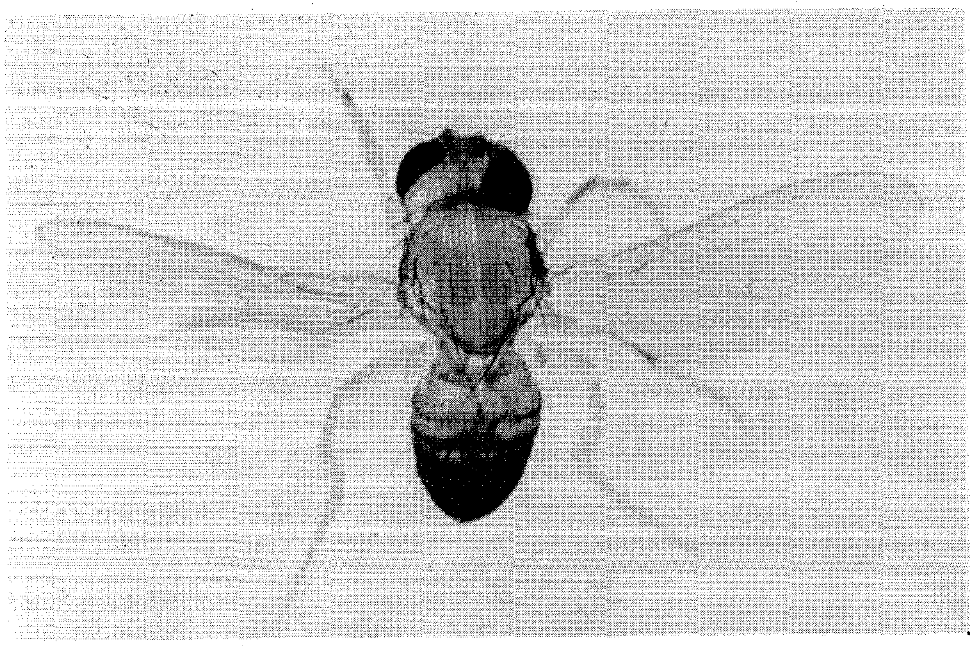
The genetics of this fly is a part of the broad program of genetics that T. H. Morgan and his colleagues initiated at the California Institute in 1928. In fact—in the period from 1910 to 1928—it was Morgan and his group at Columbia University who demonstrated that the genes are arranged in a linear order in the chromosome—much like beads strung on strings.

In 1942, it was discovered at Caltech that there are places in the chromosome where what at first had appeared as a single gene turned out, by finer methods of analysis, to be a cluster of functionally related genes. The genes responsible for the four-winged fly belong to such a cluster—called the "bithorax" cluster.

The normal fly (shown on page 20) has a pair of wings and, behind them, a pair of tiny club-shaped organs—the balancers or halteres. The balancers are thought to provide a gyroscopic action which is used to stabilize the flight of the insect. It is these balancers which are modified into wings in the case of the four-winged flies.

Such flies represent a combination of two extremely rare mutations belonging to the bithorax cluster. The first mutation, a "bithorax" type, was found in 1925 by Professor Curt Stern. It causes an overgrowth of the front half of the haltere so that it resembles the front half of the normal wing.

In 1948, some experiments with x-rays at Caltech produced another kind of mutation that resulted in a



The normal fly has a pair of wings and, behind them, a pair of small club-shaped organs called balancers or halteres.

fly in which the *back* half of the haltere develops an overgrowth resembling the *back* half of the normal wing. This mutant is therefore given the name postbithorax.

We felt that we could best test the validity of these interpretations of the mutant effects by combining the mutants in such a way that both are expressed in the same individual. Then, a fully formed wing should arise in place of the balancer.

To combine the two abnormal mutants required several steps. First, bithorax females were mated to postbithorax males. The offspring of this mating are perfectly normal two-winged flies. They are "carriers," however, of the two mutant genes. These carriers were then allowed to produce offspring which numbered 18,711 flies. Among the 18,711, only three proved to have been cases in which the two mutants were combined together in one and the same chromosome. The final step was to breed together these rare individuals. From this mating, a pure-breeding strain of four-winged flies (shown on page 21) was developed.

The extreme rarity with which the two mutants recombined was expected, since it was already known that these genes are in the same cluster—that is, they are exceedingly close together in the chromosome.

Position of genes

The rule about determining the position of genes can be expressed in another way. If a female inherits a certain genetic defect from her mother, and a quite different defect from her father, the chance that one of her eggs will receive both of these defects depends on how easy it is to get a recombination between the affected genes in the female's maternal and paternal chromosomes. The affected gene in the maternal chromosome must actually be physically recombined with the affected gene in the paternal chromosome by an interchange or "crossover" between these two threadlike structures. The closer the affected genes are to each other, the less often such a crossover occurs. In fact, we use this principle to define the gene: a gene is a unit within which crossovers do not occur.

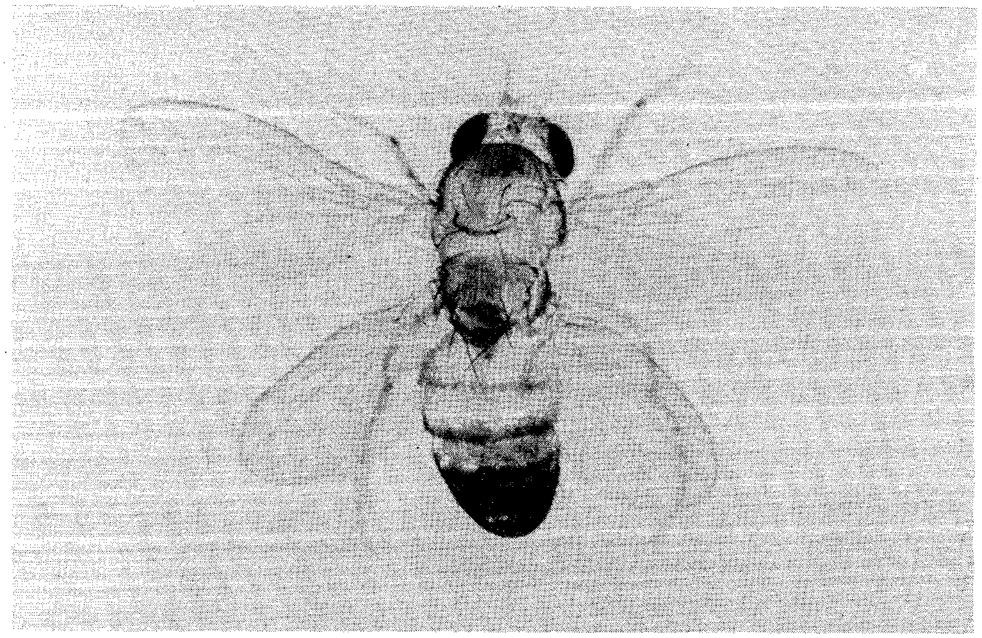
What good is a four-winged fly?

What, if anything, can we learn from a four-winged fly? One thing we hope to learn is how genes affect the development of an organism. We know that the genes are the core of living matter. They provide the information that enables the cells to grow and multiply and develop into the fully formed organism. In other words, they not only account for the transmission of characteristics from the parents to the offspring, but they are also thought to control the whole course of development of the organism from fertilized egg to adult.

In recent years much has been learned about what genes do besides simply making copies of themselves to be handed on from one generation to the next. Thus, each gene seems to control the production of a specific catalyst or enzyme which in turn controls a biochemical reaction. A whole group of investigators in the biology division at Caltech are probing this biochemical aspect of genetics.

Is such a gene-controlled chemistry sufficient to explain how, during its growth and development, an organism acquires its shape and form and elaborate differentiation of parts?

We are not sure of the answer. Instead, as a working hypothesis, we postulate that the development of a living organism is an orderly unfolding in time of many different sequences of biochemical reactions—each ultimately gene-controlled. Curiously, all of the cells of the developing organism seem to contain the same num-



The four-winged fly has fully formed wings in place of the balancers, and a greatly enlarged thorax.

ber and kind of genes. In other words, there appears to be no mechanical sorting out of the genes according to part and function.

If development is to be explained in terms of the action of genes, it becomes necessary to picture it as a gradual and orderly "turning on," so to speak, of systems of genes. The bithorax and postbithorax mutants probably represent part of one such system. Thus, the bithorax cluster is concerned, among other things, with determining whether flies shall have two wings or four wings. We infer this from the fact that, in the presence of the abnormal or mutant genes, bithorax and postbithorax, the normal pathway of development is interfered with and a four-winged fly results.

Now we have good reason to believe that flies evolved from an ancestral type which had four wings. In the evolutionary process, the second pair of wings were reduced to the balancers. From this we speculate that the normal (as opposed to the mutant) bithorax and postbithorax genes must somehow have originated as new genes whose function was to elaborate substances which suppress the potential development of the second pair of wings.

Origin of new genes

Geneticists picture the origin of new genes somewhat as follows: first the "old" gene duplicates; then one of the two identical genes thus formed is free to mutate to a "new" gene having a new function while the old gene is retained to carry out the old function. To be sure, a great many other changes—and perhaps many other new genes besides bithorax and postbithorax—had to arise before the modern fly evolved.

For example, as if to compensate for the reduction of the second pair of wings, the wing-bearing section of the fly underwent an enormous overdevelopment and produced an elaborate pattern of bristles and hairs. The fly shown in the picture above has both wing-bearing sections enormously overdeveloped. This is not surprising, of course, when it is realized that in such a fly there has been no alteration in the systems of genes which are responsible for overdeveloping the wing-bearing region.

What can we infer about the role of the bithorax cluster of genes in normal development? We postulate that, during the course of development of a normal fly, this system of genes is present but effectively inoperative in the wing-bearing section. In the haltere-bearing section, on the other hand, these genes elaborate a series of substances which direct the pathway of that section from wing formation toward haltere formation.

A working model

What determines the essential difference between the wing and haltere-bearing section? We postulate that there is a gradient in the concentration of some chemical substance during the development of the embryo, such that the concentration is relatively much greater in the haltere-forming region than it is in the wing-forming region. It would then be the function of the normal genes of the bithorax cluster to exploit this gradient—to amplify it into an "all-or-none" response. That is, the bithorax cluster would normally be inoperative in the section which ordinarily produces wings, but would be "turned on" (by the presence of greater amounts of the postulated substance) in the section which produces halteres. The normal genes of the bithorax cluster would then elaborate a set of substances which would direct the haltere-forming section to make halteres.

We have a working model for picturing the genetic control of development. Whether it is the correct model or not remains to be seen. In pursuing that model, however, we should make progress in our understanding of the living organism.