MOSAICS INVOLVING ARISTAPEDIA, A HOMEOTIC MUTANT OF *DROSOPHILA MELANOGASTER*

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Received October 21, 1963

A homeotic mutant alters an appendage of a segmental series from its typical form to that of some other member of the series. One mutant usually classified as homeotic is aristapedia (BALKASCHINA 1929), which transforms the arista of the antenna into a tarsus-like structure (Figure 1). Most mutants that have been studied by means of genetic mosaics can be considered late acting in that they appear to alter the competence of cells to respond to preexisting inducing fields or "prepatterns" (STERN 1954; TOKUNAGA 1961). Because of the drastic nature of the transformation, it has been suggested (HANNAH-ALAVA 1958) that homeotic mutants alter prepatterns. The study reported here presents evidence that the homeotic mutant, aristapedia, alters the response of antennal cells rather than altering the prepattern.

MATERIALS AND METHODS

Genetic mosaics were produced by X-ray induced somatic crossing over. An insertional translocation, T(1;3)05 has a small segment of the third chromosome (section 88 A-C to 92) bearing the wild-type allele of aristapedia (ss^a , 3-58.5) inserted close to the tip of the X chromosome which also carries the wild-type allele of yellow (y , 1-0.0, bristles and hairs yellow). The deficient third chromosome is marked by Dichaete (D , 3-40.4, wings spread). For a more complete description of the translocation and mutants see LEWIS (1951) and BRIDGES and BREHME (1944).

Males of the constitution T(1;3)05, D/ss^a were crossed to y/y ; ss^a/ss^a females. First instar larvae were exposed to 1200r of 250 kvp X-irradiation, allowed to mature, and adult females were examined for antennal mosaicism. Somatic crossing over involving the X chromosome is usually proximal to the inserted piece of chromosome 3 and produces homozygous ss^a tissue which is identifiable by the presence of yellow bristles and hairs while wild-type tissue has black bristles and hairs. It should be noted that the chief limitation of this technique is the inability to detect small mosaic patches without bristles or hairs or to determine the precise boundary between mutant and wild-type tissue since the cuticle is yellowish in both.

RESULTS

Examination of 1057 duplication-bearing (non- D) females disclosed 46 mosaic antennae. No mosaicism was observed on the heads of 1368 D females indicating that the heterozygous deficiency is cell lethal, at least in the head.

Figure 1 illustrates a mosaic head bearing a wild-type antenna with its feathery

¹ Operated by Union Carbide Corporation for the United States Atomic Energy Commission.

FIGURE 1.—A head bearing a wild-type antenna (right) and a mosaic antenna (left). In this and other figures wild-type bristles and hairs are in solid black and homozygous ss^a bristles and hairs that are yellow in the fly are in outline. Segment numbers are indicated on the wild-type antenna. 40 $\times$.

arista (right) and the tarsus-like appendage which is formed when most of the distal antennal tissue is homozygous ss^a (left). This antenna was almost as tarsus-like as the appendage found on nonmosaic ss^a homozygotes in spite of the juxtaposition of wild-type tissue in segment 2 and ss^a tissue in segments 3 to 6.

Table 1 divides the 46 recovered mosaics into five categories. The divisions are somewhat arbitrary since Types 4 and 5 can be considered variants of Type 3. The ensuing description applies only to segments 4 to 6 since no mosaicism for segment 2 was observed and segment 3, while frequently modified by mosaicism (Figures 1 to 3), shows less contrast between the two genotypes than the distal segments.

Ten of the mosaics (Type 2) were similar to Figure 1 in that they had no detectable wild-type tissue distal to segment 2. Segments 4 to 6 were, in general, strongly segmented but showed more variability than a sample of ten nonmosaic ss^a antennae, suggesting that there was nondetectable mosaicism.

Except for two mosaics (Type 1), which had little tissue beyond segment 3, the combined length of the distal segments fell within a narrower range than

TABLE 1

The frequency of occurrence of five types of mosaic antenna among 46 antennal mosaics

Type 1	Type 2	Type 3			Type 4	Type 5
Little tissue distal to segment 3	Uniformly tarsus-like (Figure 1)	Tarsus proximally, arista distally			Tarsus with aristal filaments (Figure 3)	Tissues intermingled (Figure 4)
		Mostly tarsus	Half tarsus, half arista (Figure 2)	Mostly arista		
2	10	8	10	5	6	5

FIGURE 2.—A mosaic antenna which is tarsus-like proximally and arista-like distally. 90 $\times$.

might have been expected, about twice the width of the distal part of segment 2 regardless of the proportion of mutant and wild-type tissue. The majority of mosaics, Type 3, were tarsus-like proximally and arista-like distally. The composition of these mosaics varied from mostly tarsus to mostly arista with Figure 2, half tarsus and half arista, midway in this series.

No mosaic antennae had appreciable tarsus development distal to a thin arista (Figure 4). When tissues of the two genotypes were intermingled to an even greater degree as in Types 4 and 5 (Figures 3 and 4), *ss^a* tissue was usually thick when proximal, and when thick, strongly segmented with short bristles while wild-type tissue tended to be thinner, more chitinous, and weakly segmented with long filaments.

DISCUSSION

The development of distinct tarsal segmentation in tissue that is predominantly *ss^a* appears to depend on the degree of dilution with wild-type tissue. Where this is not apparent (Figure 1) or slight (Figures 2 and 3, proximal portion of segments 4 to 6) segmentation is distinct. The formation of arisal filaments, however seems to depend only on the presence of a continuous band of wild-type tissue running proximal to distal in a mosaic antenna. This is apparently so in Figure 3

FIGURE 3.—A mosaic antenna in which wild-type tissue forms regular arisal filaments although surrounded by tissue which is *ss^a*. 100 $\times$.

FIGURE 4.—Segments 4 to 6 of an antenna in which tissues of the two genotypes are quite intermingled. 156 $\times$.

but not in Figure 4, where the tip of the antenna is undoubtedly *ss^a*, a condition which has prevented the development and separation of all but the bottom filament. Segmentation in the proximal part of segments 4 to 6 of this particular antenna is weak compared with the same region in Figures 1 to 3. Close examination reveals traces of internal pigmentation suggesting that considerable wild-type tissue lies internally in this region. One interpretation is that owing to the diffusion of substances from one tissue to another, autonomy is not complete. A simpler explanation is suggested by a comparison of antennal development in the two genotypes.

The development of *ss^a* and wild-type antennal disks has been described by VOGT (1946). At 72 hours of development, the antennal disks of the two genotypes are quite similar except for the thickness of the "end knob" which is one third again as thick in the mutant as in the wild-type disk. At the time of prepupa formation the mutant end knob is more than twice the thickness of the wild-type. Subsequently, in the *ss^a* disk, segmentation occurs within the thickened end knob while in the wild-type disk the end knob grows distally to a long, thin point out of which the arista forms. Since the two alleles control, among other things, growth rates of antennal cells, it is possible that the primary effect of dilution of *ss^a* tissue by wild-type tissue is to limit the thickness of the antenna. This may then weaken tarsal segmentation if segmentation is, in turn, dependent on thickness as is suggested by the mosaics. The studies of VOGT also suggest that the failure to recover mosaic antennae with appreciable tarsal development distal to an arista may be attributable to the necessary development of the arista from a terminal outgrowth.

If the mutant and wild-type alleles of *aristapedia* control antennal prepatterns, an antenna which is predominantly one type of tissue should have the corresponding prepattern. A small amount of tissue of the other genotype would be expected to conform to the prepattern set up by the tissue which predominates. Instead of this, the two genotypes exhibit a high degree of autonomy. Small patches of wild-type tissue form long filaments characteristic of an arista even when surrounded by *ss^a* tissue. Conversely, *ss^a* tissue forms tarsus-like segments with wild-type tissue both proximally (in segments 2 and 3) and distally. It is unlikely that the *aristapedia* alleles could be controlling antennal prepatterns, but because the antennal disk was wild-type until the first instar when mosaicism was induced, it might be questioned whether an *aristapedia* allele, acting from the time of fertilization, does not affect the antennal prepattern. Using a temperature sensitive

aristapedia allele, ss^{a-f} , VOGT has shown that the antennal disk is not determined until the third instar. At this time, the appropriate temperature treatment can induce the ss^{a-f} disk to form a tarsus, an arista or intermediate types much as Table 1, Type 3 and Figure 2. Therefore, in the present study, mosaicism induced in first instar larvae was at a time the antennal disks first became visible, at least 24 hours before they became determined. The aristapedia alleles clearly do not control antennal prepatterns.

The evidence suggests that the aristapedia alleles control the competence of cells to respond to what is, in this experiment, an invariant prepatter. Not only do aristapedia and, to a lesser extent, wild-type antennae show evidence of segmentation, but mosaic antennae with the two types of tissue intermingled often have a periodicity of structure indicative of underlying segmentation (Figure 3). Also, the length of a mosaic antenna usually falls within definite limits and is not merely an arista added onto a tarsus (Figure 2). These observations suggest that the aristapedia alleles control the competence of antennal cells to respond to a prepatter which is more general than either of the two mutant phenotypes resembling the primitive, unspecialized arthropod appendage, a segmented appendage of a definite length.

At this point it should be mentioned that serial homology of the eyes and antennae with other appendages is not supported by studies of their innervation (FERRIS 1950). It is impossible, at this time to weigh one type of evidence against the other, but a developmental pathway which has persisted throughout the course of insect evolution, phenotypically concealed until revealed by mutation, may have been less susceptible to modification by selection than patterns of innervation. Mutants are known which produce limb-like appendages from segments now bearing organs which are farther from limb morphology than the antennae. Tetraltera and podoptera transform the wings into segmented appendages (GOLDSCHMIDT 1945), and even the eyes can be caused to bear segmented appendages by erupt (GLASS 1944) and ophthalmopodia (BRIDGES and BREHME 1944).

The normal segmentation pattern of the tarsi involves at least seven known loci (WADDINGTON 1962). Moreover, the characteristic effect of each of these genes is produced on developing leg material in any body segment (e.g., the antennal tarsus). It may be that these genes also control the response of imaginal disk cells to a prepatter which is even more general than suggested above, lacking even the characteristic of segmentation, but it is also a distinct possibility that the "appendage prepatter" is polygenically controlled and that seven of the controlling genes are known.

There is additional evidence that the appendage prepatter is quite general in the recent report of LEWIS (1963). It is accepted that the Diptera are descended from four-winged ancestors, a condition which is duplicated by the mutant bithorax. LEWIS, using the duplication described above but a different technique of producing mosaics, has reported that bithorax is also autonomous. This can be interpreted to mean that bithorax does not control a wing prepatter but controls the response of metathoracic cells to a prepatter which is, perhaps, closer to

podoptera. Also, KROEGER (1959) has presented evidence that the fore- and hindwings of the moth *Ephestia* have identical prepatterns.

The production, by mutation, of limb-like appendages in all these body segments suggests that there is a relict developmental pathway producing an appendage prepatter which is common to all the imaginal disks. The appendage prepatter was probably expressed with comparatively little modification by other genes in some remote ancestor of *Drosophila* which bore similar segmented appendages on all body segments. Its expression has been modified in various ways—profoundly in the eyes, wings and antennae, to a lesser degree in the legs—by striking, gene controlled alterations of the competence of cells to respond to this prepatter during the course of insect evolution.

SUMMARY

Evidence is presented from a study of antennal mosaics that the mutant and wild-type alleles of a homeotic mutant, *aristopedia*, do not control "antennal" prepatterns but control the competence of antennal cells to respond to an "appendage prepatter."

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