

A New Trait for Distinguishing *Drosophila azteca* and *D. tolteca* from Other Members of the *D. affinis* Subgroup¹

ABSTRACT: *In male Drosophila azteca and D. tolteca the base of the genital clasper is covered by numerous small hairs. Such hairs are lacking (or nearly so) in other D. affinis subgroup species observed: D. affinis, algonquin, athabasca, helvetica and narragansett. This trait should be useful for distinguishing males of D. athabasca and D. azteca where their geographical ranges approach each other or overlap. Hybrid males from the D. athabasca ♀ × D. azteca ♂ cross have very little hairiness of the clasper base, while males from the reciprocal cross are distinctly hairy. Hybrid males from the D. athabasca ♀ × D. tolteca ♂ cross show a pronounced hairiness of the clasper base. Males from both reciprocal crosses between D. azteca and D. tolteca resemble their parent species in having hairy claspers.*

Sulerud and Miller (1966) reported that males of laboratory strains of *Drosophila athabasca* and *D. azteca* differed in numbers of primary clasper teeth, the former species having eight or more teeth per clasper comb, the latter usually six or fewer. It was proposed that this difference should be useful for separating males of these closely related species, supplementing the difference in mesonotum striping pattern presented by Sturtevant and Dobzhansky (1936) as a basis for separating specimens of these species. However, it was also noted that some wild males from regions of possible proximity or coexistence of the two species (Oregon, northern California, southern Arizona) had seven clasper comb teeth and hence remained doubtful as to species identity.

It has since been found that a laboratory strain of *D. azteca* from southern Arizona (Chiricahua Mountains) contains some males with seven teeth per clasper comb and that several of our strains of *D. athabasca* (Massachusetts, Minnesota, North Carolina, Pennsylvania, Vermont) likewise contain males with seven clasper comb teeth. Thus, the two species definitely overlap re-

¹ Contribution Number 409 of the Department of Zoology of the University of Nebraska.

garding this character. Hence, the attempted identification of males as *D. athabasca* or *D. azteca* on the basis of number of clasper comb teeth would necessarily involve some uncertainty, though the frequency of cases of ambiguity is possibly low.

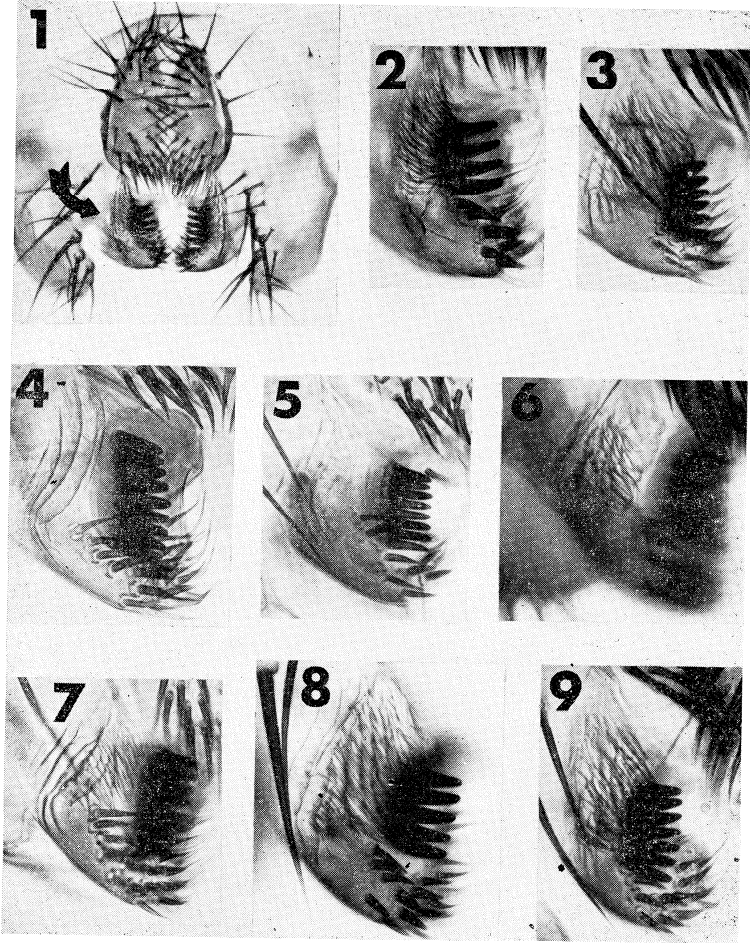
We have recently re-examined the genital arch of males in laboratory strains of seven available species of the *D. affinis* subgroup (*D. affinis*, *algonquin*, *azteca*, *athabasca*, *helvetica*, *narragansetti*, and *tolteca*) and have discovered a trait, the presence of small hairs covering the base of the clasper, that distinguishes two of the species, *D. azteca* and *D. tolteca*, from all the rest. On the basis of these laboratory strains it would appear that this character should serve to distinguish males of these two species from all others and, in particular, would make possible the separation of *D. azteca* from *D. athabasca*, since the base of the clasper is bare (or nearly so) in the latter species.

Specimens of the following *D. affinis* subgroup species strains have been scrutinized for presence or absence of hairiness on the basal region of the clasper in males: *D. affinis* Nebraska (Humboldt); *D. algonquin* Ontario (Owen Sound); *D. athabasca* Alaska (Anchorage, Matanuska Valley), Colorado (Garden of the Gods, Gothic), Idaho (Boise), Manitoba (Churchill), Massachusetts (Amherst), Minnesota (Duluth, Halstad, Lake Itasca, Lake Shamaineau), New Mexico (Raton Pass), North Carolina (Highlands), Oregon (Eugene), and Pennsylvania (Carbon Co., Philadelphia); *D. azteca* Arizona (Chiricahua Mountains), California, (Siskiyou Mountains), and Mexico (Chilpancingo, Mexico City); *D. helvetica* Switzerland (Zürich); *D. narragansetti* Nebraska (Halsey); and *D. tolteca* Bolivia (Cobrico) and Colombia (Medellin).

Two methods were used to prepare specimens for observation. In one, males previously immersed in 95% alcohol were transferred to creosol (24 hours or more); the genital arch was then dissected away and observed in creosol or after transfer to euparal. In the other, males were cleared by brief boiling in 10% KOH, then transferred to glycerine, in which the genital arch was dissected off and observed. Visibility of the clasper region under consideration seemed equally good with either method. Observations were made at 160 \times and 400 \times magnifications with bright field illumination.

Only in strains of *D. azteca* and *D. tolteca* was a pronounced hairiness of the clasper base regularly found. In all remaining species either no hairs could be observed or else no more than a very few widely spaced hairs were visible. In *D. athabasca* there appeared to be some interstrain variation regarding such hairs; definite presence of a few hairs was encountered in strains from Alaska (Anchorage) and Minnesota (Lake Shamaineau), but there were far fewer hairs than in *D. azteca* and *D. tolteca*. The existence of folds and creases in the surface of the clasper made it difficult to be sure of the total absence of small hairs in species other than *D. azteca* and *D. tolteca*.

Drosophila athabasca, *azteca*, and *tolteca* are capable of hybridization in the laboratory in the following five ways: both reciprocal crosses of *D. athabasca* \times *D. azteca* (Sturtevant and Dobzhansky, 1936); both reciprocal crosses of *D. azteca* \times *D. tolteca* (Patterson, 1954); and *D. athabasca* $\text{♀} \times$ *D. tolteca* ♂ (Ensign, 1960). We have repeated these hybridizations and have obtained male hybrids which have been examined for the clasper trait distinguishing *D. azteca* and *D. tolteca* from *D. athabasca*. Figures 1 and 2 illustrate the clasper region in *D. azteca*, Figures 3 and 4 the corresponding part in *D. tolteca* and *athabasca*, respectively. The contrast between the first two species and *D. athabasca* is apparent. Figures 5 and 6 show claspers of male hybrids between *D. athabasca* and *azteca*. The clasper is virtually bare in the hybrid of *D. athabasca* $\text{♀} \times$ *D. azteca* ♂ (Fig. 5) while the male hybrid from the reciprocal cross has a clasper that is quite hairy (Fig. 6). Thus, with



Figs. 1-9. 1.—Genital arch of *Drosophila azteca* (California) male; the arrow indicates the left clasper (left and right claspers did not appear to differ regarding the trait in question—*i.e.*, hairiness). 2.—Enlarged photomicrograph of left clasper of *D. azteca* (same specimen as in Fig. 1). 3.—Left clasper of male *D. tolteca* (Bolivia). 4.—Left clasper of male *D. athabasca* (Oregon). 5.—Clasper of F₁ male *D. athabasca* (New Mexico) ♀ × *D. azteca* (Arizona) ♂; actually the right clasper reversed to correspond to other specimens illustrated. 6.—Left clasper of F₁ male *D. azteca* (Mexico City) ♀ × *D. athabasca* (Oregon) ♂. 7.—F₁ male *D. athabasca* (Colorado) ♀ × *D. tolteca* (Bolivia) ♂. 8.—F₁ male *D. azteca* (California) ♀ × *D. tolteca* (Bolivia) ♂. 9.—F₁ male *D. tolteca* (Colombia) ♀ × *D. azteca* (California) ♂.

All pictures were taken with a Zeiss photomicroscope, Fig. 1 printed at 315X, the others at 800X magnification, reduction to four-ninths of print size.

regard to this trait, male hybrids resemble males of their maternal species. In other respects these hybrid males differ strikingly from males of the maternal species as well as from each other; *D. athabasca* ♀ × *D. azteca* ♂ hybrid males are small-winged dwarfs while *D. azteca* ♀ × *D. athabasca* ♂ hybrid males are unusually large with relatively large wings (Sturtevant and Dobzhansky, 1936). Fig. 7 illustrates a clasper of a male hybrid from the *D. athabasca* ♀ × *D. tolteca* ♂ cross; the presence of an appreciable amount of hair, more than in the maternal species, is evident. Figs. 8 and 9 show claspers of hybrid males from the two reciprocal crosses between *D. azteca* and *tolteca*. Not surprisingly these manifest hairiness similar to that of both parental species.

Patterson and Mainland (1944) suggested, on the basis of morphological traits, that the newly described species *D. tolteca* was closely related to *D. azteca*. Patterson's report (1954) of hybrids in the laboratory between these two species, with fertility of both sexes in *D. tolteca* ♀ × *D. azteca* ♂ hybrids, provides additional evidence of close relationship. We have recently found that female hybrids from the reciprocal cross (*D. azteca* ♀ × *D. tolteca* ♂) sometimes have limited fertility; mated to *D. tolteca* ♂♂ they yielded a few offspring that died as pupae (not necessarily significant since, within these species, cultures with only a few larvae sometimes fail to produce adults). These two species provide the only known case in the *D. affinis* subgroup of fertile hybrids from both reciprocal crosses. The pronounced hairiness of the clasper base, shared by *D. azteca* and *tolteca* but absent in other *affinis* subgroup species, is still additional evidence of a closeness of their relationship.

Acknowledgments.—We are indebted to Dr. Hans Burla (Zoological Institute and Museum, University of Zürich, Switzerland) for providing us with a strain of *D. helvetica*. The senior author is grateful to Dr. M. R. Wheeler and the University of Texas for space and facilities provided him during part of this study and for access to a few additional strains from the University of Texas collection.

REFERENCES

- ENSGIN, S. E. 1960. Reproductive isolation between *Drosophila tolteca* and related species. *Evolution*, **14**:378-385.
- PATTERSON, J. T. 1954. Note on a new case of interspecific hybridization in the *D. affinis* subgroup. *Univ. Texas Publ.*, No 5422:46.
- AND G. B. MAINLAND. 1944. The *Drosophilidae* of Mexico. *Ibid.*, No. 4445:9-101.
- STURTEVANT, A. H. AND TH. DOBZHANSKY. 1936. Observations on species related to *Drosophila affinis*, with descriptions of seven new forms. *Amer. Natur.*, **70**:574-584.
- SULERUD, R. L. AND D. D. MILLER. 1966. A study of key characteristics for distinguishing several *Drosophila affinis* subgroup species, with a description of a new related species. *Amer. Midl. Natur.*, **75**:446-474.
- DWIGHT D. MILLER AND WARREN G. SANGER, Department of Zoology, University of Nebraska, Lincoln 68508. Submitted 3 March 1969; accepted 17 March 1969.

Reprinted from

THE AMERICAN MIDLAND NATURALIST

Vol. 82, No. 2, October, 1969, pp. 618-621

University of Notre Dame Press

Notre Dame, Indiana