

Drosophila mojavensis baja Mettler 1961. DIS 38:57-58
Drosophila neonasuta Sajjan and Krishnamurthy 1972. DIS 48:56-57*
Leucophenga neoangusta Godbole and Vaidya 1977. DIS 52:24
Stegana subexcavata Vaidya and Godbole 1977. DIS 52:55-56
Zaprionus paravittiger Godbole and Vaidya 1972. DIS 48:135-136

*Starred species were later described again in another journal.

In my opinion, *Drosophila* taxonomists should not publish new species descriptions in DIS--at least until it is formally recognized as a "publication". Further, it is not wise to include new names in articles of a non-taxonomic nature. The Code provides that a new name may be valid if accompanied by a "description"; but a complete, thorough description is not required--the simplest descriptive remark may be enough to validate a new name (e.g., describing the chromosomes, some electrophoretic patterns, etc.). *Drosophila* workers have a rather poor reputation in systematic circles, having used new, unpublished names without regard to the International Code.

Regretfully, the writer is an expert on this subject, having made more than a few of such errors!

Wijsman, E.M. University of Wisconsin, Madison, Wisconsin. The effect of ether on mating behavior in *D. simulans* y w.

In setting up some experiments which involved matings between virgin females and their brothers in *D. simulans* y w, I encountered considerable difficulty with sterility. I decided to test the possibility that the ether that I

was using as an anesthetic was causing this sterility.

I established pair matings using virgin females and their brothers separated by ether, CO₂, or aspirator (no anesthesia), and placed the vials at 25°C. Two weeks later I scored the vials as fertile or sterile. As can be seen in Table 1, ether had a very strong effect on fertility. The hypothesis that anesthesia had no effect on fertility was tested using a 1-tailed Fisher's exact test. Comparison of ether and no anesthesia gave $p < 0.000001$. CO₂ vs. no anesthesia gave $p = 0.18$, which is not significant.

Table 1. Number of vials which were either fertile or sterile when parents were exposed to different types of anesthesia.

Anesthesia	Fertile	Sterile
Ether	4	56
CO ₂	18	8
None	21	4

To determine which of the two sexes was sterilized I repeated the experiment using only one sex which had been exposed to ether. When only the male had been anesthetized high sterility resulted. Anesthetized females mated to non-anesthetized males were fertile.

To determine the cause of sterility I dissected the testes to check for motile sperm and watched the males court females. Males were isolated for 3-4 days after collection with either ether or an aspirator and then placed in empty vials with 3 aged virgin females. Those which had been collected without ether showed normal courtship behavior;

those which had been exposed to ether showed virtually no courtship behavior. Dissection of the testes showed motile sperm. Thus in this strain of *D. simulans*, ether seems to produce almost complete, permanent, behavioral sterility in the males.

Williams, J.M. University of California, Santa Cruz, California. Tumorigenesis in *D. melanogaster* bearing the temperature-sensitive mutation *shibire*^{ts1}.

The imaginal discs of *Drosophila* are single-layered secretory epithelia (Bodenstein 1950; Poodry and Schneiderman 1970) which resemble the ascinar units of vertebrate exocrine glands. This feature has been exploited along with the convenience of in vivo culturing meth-

ods (Hadorn 1963) to characterize the initial morphological and ultrastructural changes occurring in the eye-antenna imaginal disc of *D. melanogaster*.

A temperature-sensitive mutation, *shibire*^{ts1} (Poodry et al. 1973) in *D. melanogaster*

was used to generate information concerning the timing of initiation of tumorous growth and the pattern of cellular proliferation in the neoplasm. Neoplasia in *Drosophila* is well documented (Gateff 1977, 1978); however, none of the previously defined neoplasms of genetic or epigenetic origin have yielded satisfactory data concerning the initial stages of tumorigenesis. The fact that the eye-antenna disc of *shit^{ts1}* is temperature sensitive, transplantable, displays autonomous growth and loss of differentiation capacity has augmented its usefulness in documenting patterns of neoplastic change.

Shit^{ts1} eye-antenna discs were dissected from mature third instar larvae and implanted into the hemocoel of mated 3-4 day old Ore-R female hosts (Ursprung 1967). Host flies were incubated at 29°C (the mutant restrictive temperature) in shell vials containing standard medium. In some experiments these flies were cultured for two weeks. After this time the eye disc had tumorized and began to fill or filled completely the abdominal cavity. The tumorous growth was dissected from the abdomens in buffered ringer solution, fragmented with tungsten needles and reimplanted for second generation growth (one generation = two weeks). Wild-type eye discs do not tumorize or behave similar to *shit^{ts1}* when treated in an identical fashion.

Other eye-antenna discs from *shit^{ts1}* third instar larvae were incubated in vivo for periods ranging from 16 to 22 days. These implants were cultured at 29°C for period between 2 and 10 days and then maintained at 22°C for the remainder of the incubation period. After dissection from the abdomens these implants were measured with a stage micrometer and examined for gross morphological features. Data summarized in Table 1 show that tumorigenesis is initiated within a 48 hour period in these tissues and that continued heat stimulation is not required to maintain tumorous growth. These data indicate that tumorigenesis in this tissue is irreversible. Furthermore, it is noted that the tumors grew to about the same size irrespective of the time cultured at 29°C. This indicates that a maximum pattern of proliferation was established concomitant to the initiation of tumorigenesis. Thus, the neoplasms behave autonomously. This expression is initiated via temperature sensitivity to yield information concerning regulation of gene expression in normal vs. tumorous tissue.

Table 1.

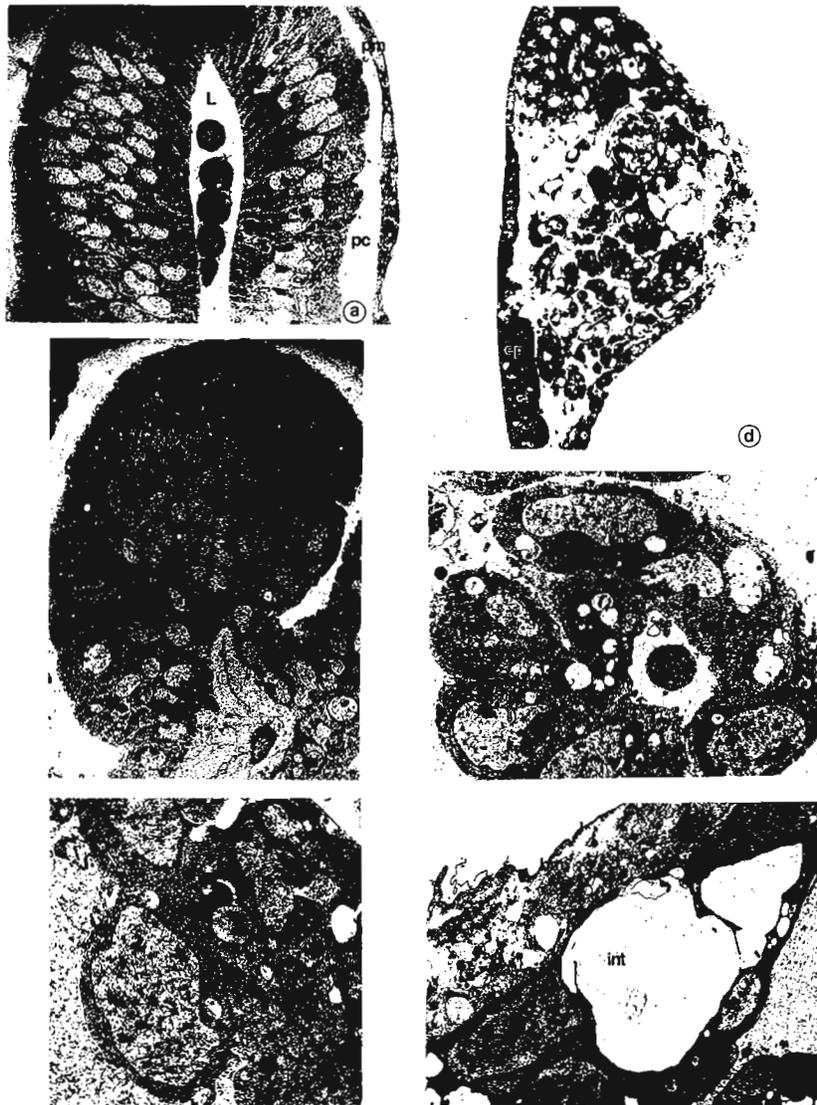
	Group I	Group II	Group III	Group IV	Group V
Days at 29°C	8	10	8	4	2
Days at 22°C	14	10	10	13	14
Total days in culture	22	20	18	17	17
Size* in mm ² x 10 ⁻³	1.2	1.08	1.138	1.21	1.2

*Size of control disc is ~0.85 mm² x 10⁻³.

bulge in what appears to be a solid mass of cells. The arrangement of the monolayer becomes contorted resulting in irregular folds and projections. Some cells lose contact with the basement membrane in areas and aggregate in groups. The predominant columnar appearance of the epithelial cells seen in wild-type discs is not visualized in temperature-sensitive disc epithelia cultured for 6 hours. Instead they become more cuboidal and irregular in shape and show modification to the apical border. The microvilli become shortened and irregular with disorderly microtubular arrangement. Cell-to-cell contacts are interrupted by intercellular spaces and membrane-bound undersides appear intracellularly. The cytoplasm contains numerous ribosomes; many rough ER are present and possibly more mitochondria are present in these cells than in the controls.

Many of the initial morphological aberrations are detectable in implants cultured for longer periods and other abnormalities result as well. The basal lamina of these tumors often form pockets filled with amorphous material, vesiculate particles and dead cell debris. It is often thrown into irregular projections extending beyond the basal surface of the epithelial cells. Multiple cell layers are seen and membranes of juxtaposed cell layers often appear fused. Cells with picnotic nuclei increase in number with continued in vivo cultures as well as cells containing virus-like particles.

Implants ranging from 6 hours to 4 weeks were used in histological and ultrastructural studies. The study of 1 μm sections and thin section electron micrographs reveal that structural changes occur in the disc epithelium within the first 6 hours of in vivo culture. Regions of the epithelium



- a. The cross section of third larval instar eye disc appears as a pseudostratified single layered epithelium with distinct microvillar and basal surfaces. PM, peripodal membrane; PC, peripodal cavity; dv, dividing cell; L, lumen; MC, macrophage-like cells. (1300X)
- b. A section of eye-antenna disc tumors cultured for 6 hrs at 29°C. A mass of epithelial cells infiltrated with dead cells (dc) protrudes into the lumen of the disc. (1000X)
- c. Epithelium with virus-like particles (vlp) present. Intercellular spaces and a cell elaborating microvilli on two opposite sites is seen. (10,000X)
- d. The epithelium (ep) which makes up the cortex varies in thickness but seems to be single-layered. Aggregations of cells and cells organized in monolayers around a central lumina (arrows) are found in the medulla (M). (200X)
- e. An aggregate of cells reminiscent of the ommatidial precursor clusters. No apical/basal distinction is apparent. (5000X)
- f. The basement membrane (bm) is not in contact with the basal surface of the epithelial cells. A massive amount of mitochondria (arrows) and amorphous debris is present between them. Atypical cell morphology and large intercellular spaces (int) are evident. (3300X)

After one generation in vivo the monolayer of epithelial cells becomes rearranged. It appears sponge-like due to intercellular spaces; it also lacks cellular continuity in areas. The basal lamina is often the only structure maintaining the sac-like appearance. In these tumors the outer portion of termed the "cortex" and is comprised of a remnant population of epithelial cells. These cells surround a "medulla" region which is composed of cells arranged in spherical configurations. The cell number in these spheroids vary but are reminiscent of the ommitidial precursor cluster found in the developing eye disc of the wild-type (Waddington and Perry 1960). Thus, it is possible that tumorigenesis did not affect the determined state of this cell population, but did interfere with the differentiation process. A considerable amount of cell debris and amorphous material is found in the medulla.

Autoradiographic studies of tumors incubated with ³H-thymidine for 48 hours showed differential incorporation in areas of the tumor where masses of cells bulge in the epithelium. This indicates that proliferation continues in the epithelium (cortex region) as opposed to the medulla. These features are important in determining basic kinds of cellular interactions which occur in other tumors arising from secretory epithelia and are indicative of a certain pattern of neoplastic change.

References: Bodenstern, D. 1950, *Biology of Drosophila*, Hafner, New York; Gateff, E. 1977, DIS 52:129; _____ 1978, *Science* 200:1449-1459; Hadorn, E. 1963, *Develop. Biol.* 7:617-629; Poodry, C.A. et al. 1973, *Develop. Biol.* 32:376-386; Poodry, C.A. and Schneiderman, H.A. 1970, *Wilhelm Roux Archives* 166:1-44; Urpsprung, H. 1972, *Biology of Imaginal Discs*, Springer-Verlag, New York; Waddington, C.H. and M.M. Perry 1960, *Proc. Roy. Soc. B* 153:155-178.

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Wu, C.K. and P. Smith. Adrian College, Adrian, Michigan. Calcium cyclamate induced lethal effect and genetic damage in spermatocytes of *Drosophila*.

In determining the lethal effect of calcium cyclamate on development, v w f females of the same age were mated individually with three males into five different series according to the concentrations of calcium cyclamate solution in the food media. It is assumed

that, on the average, one female would lay the same number of eggs during the same period of time. It was found that in the treated series, the survival rates were decreasing with increasing concentrations of calcium cyclamate solution in the media, or in other words, the higher the concentration of calcium cyclamate in the medium causes the higher rate of lethality (Table 1). It clearly suggests that calcium cyclamate causes lethal effect on the early development of *D. melanogaster*.

Table 1. Average number of progeny, survival rate and lethality rate from a single female *Drosophila* in media with different concentrations of calcium cyclamate.

	Control	Cyclamate media			
		0.625%	1.25%	2.5%	5%
Male	46.3 ± 9.8	27.2 ± 4.3	27 ± 2.5	10.2 ± 11	0
Female	56.7 ± 4.3	37.2 ± 2.7	35.6 ± 13.2	15.8 ± 9.8	0
Total	103 ± 14.1	64.4 ± 3.8	62.6 ± 13.0	28.6 ± 18.4	0
Survival rate	1.00	0.63	0.61	0.28	0
Lethality	0	0.37	0.39	0.72	1

To estimate the chromosomal damage induced by calcium cyclamate, a doubly marked Y chromosome was used in the experiment. Males of the composition ywf/B^S.Y.y⁺ (y= yellow body; w=

white eyes; f= forked bristles; B^S= Bar eyes of Stone, which is a marker on the long arm of the Y chromosome; y⁺= normal allele of yellow, which is attached to the tip of the short arm of the Y chromosome) were used in this study. Day-old males were collected and transferred to a treatment chamber in which medium mixed with 1.25% calcium cyclamate for about 2 days. Then, the treated males were mated individually with three virgin females of the composition ywf/ywf for a period of 9 days; males treated with 1.25 sucrose mated in the same manner served as the control.

The regular offspring from these crosses are phenotypically yellow, white, forked females and Bar, white forked males. An exchange between the X chromosome and Y^L (the long arm of the Y chromosome) proximal to the B^S marker generates an X chromosome with Y^S and the appended y⁺ marker attached proximally and is recoverable as a phenotypically white, forked female (ywf.