

with photoflow and chilling them before putting in the resolving gel.

This research was supported by grant GM 19307 from NIH.

References: Dickinson, W.J. 1971, *Develcp. Biol.* 26:77-88; Grossbach, U. 1965, *Biochem. Biophys. Acta* 107:180-182; Jovin, T., A. Chrambach and M.A. Naughton 1964, *Anal. Biochem.* 9: 351-369; Ward, S., D.O. Wilson and J.J. Gilliam 1970, *Anal. Biochem.* 38:90-97.

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Guest, W.C. and T.C. Hsu. University of Arkansas, Fayetteville, and University of Texas M.D. Anderson Hospital, Houston. A new technique for preparing *Drosophila* neuroblast chromosomes.

The value of *D. melanogaster* larval neuroblast cells for studying somatic metaphase chromosomes has long been recognized and a variety of squash techniques have been developed. Hsu (1971) used brain squashes to obtain metaphases for demonstrating the similarity of *Drosophila* heterochromatin to the heterochromatin in mammalian

chromosomes. However, it is frequently difficult to obtain numerous division figures with well spread chromosomes using conventional squash techniques.

We have recently applied some of the Giemsa banding procedures widely used in studying mammalian chromosomes to *Drosophila* using a modification of the technique developed by Stock, Burnham and Hsu (1972) for obtaining well spread metaphase figures from solid tissues. This procedure, in addition to giving large numbers of metaphase and late prophase figures with well spread chromosomes, permits the slides produced to be handled as air dried preparations.

The procedure is as follows:

1. Twenty-five to thirty larval brains are dissected out in physiological saline or insect Ringer's solution. The brains should be washed several times in saline to remove debris. The physiological saline is replaced with a hypotonic solution. Both 1% sodium citrate and physiological saline diluted with equal parts of distilled water were used with ten to fifteen minutes giving good results. The hypotonic saline is removed with 1 ml tuberculin syringe or with a microcapillary pipette.

2. The ganglia are fixed in methanol acetic acid (3:1) with the fixative being changed several times to insure complete removal of water. The fixed material may be used immediately or may be stored in the fixative. Fixed material stored in absolute alcohol is much more difficult to work with than ganglia stored in the fixative.

3. Slides which have been cleaned thoroughly are warmed to 40-45°C on a hot plate or slide warmer.

4. The fixed ganglia are transferred to a well slide. We used a standard depression slide with a cylindrical well 3 mm deep. Remove as much of the fixative as possible with a syringe or microcapillary pipette. It is helpful to perform these transfers under the low magnification of a stereoscopic microscope. Add approximately 0.2 ml of 60% acetic acid to the well. Under the dissecting microscope the tissue will begin to swell and become translucent. Using a Drummond micropipette (see Stock, et al. for description) agitate the material by drawing the ganglia into the pipette several times. As this is done the ganglia will begin to disintegrate. This is a critical step. If the material remains in the strong acetic acid too long the cells will be distorted or destroyed. Best results have been obtained by adding the acetic acid, agitating briefly, then applying the material to warmed slides while some of the brains are still intact but swollen.

5. The cell suspension and intact tissues are drawn into the Drummond pipette and a small drop placed on the heated slide. Immediately the drop is drawn back into the pipette leaving a thin circular film on the slide containing a monolayer of cells. This process is repeated many times over the entire slide very quickly while the slide is warm. An area that can be covered by a 22 x 40 or 22 x 50 mm cover slip can be spotted on each slide. It is important that the slide be kept warm to accelerate evaporation of the acetic acid.

6. The slides are dried thoroughly before using. While we have used slides made in this manner for banding chromosomes or for staining heterochromatin, the slides can be used in a number of techniques that require a monolayer of air dried cells.

References: Hsu, T.C. 1971, *J. Heredity* 62:285-287; Stock, A.D., D.B. Burnham and T.C. Hsu 1972, *Cytogenetics* 11:534-539.