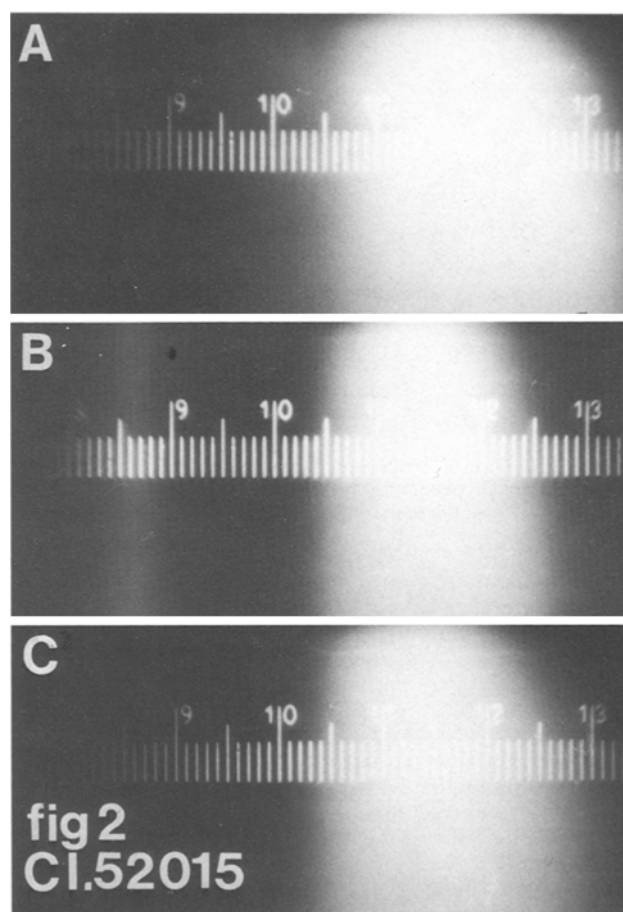


Figure 1. Spectra obtained from Ponceau de Xylidine: *A* Spectrum of the stain solution, *B* spectrum of the Ektachrome slide, *C* spectrum of the Kodachrome slide.

Figure 2. Spectra obtained from methylene blue: *A*, *B*, *C* as for figure 1.



was used, without standardization of its numerical scale by sodium light. The spectra were photographed using a Wild stereomicroscope. We have always studied and compared successively the spectra of the colored solution in the flask and the Ektachrome and the Kodachrome slides prepared as above. Results are shown in figures 1 and 2 for Ponceau de Xylidine and methylene blue.

For the red stains (Ponceau de Xylidine and acid fuchsin), the 3 spectra were exactly similar; no difference was observed between the 2 films. For methylene blue, the Ektachrome transparencies were seen to exhibit a strong additional red line (fig 2, *B*). This abnormal line was not present on the spectrum of the Kodachrome slide; this last spectrum could be superimposed exactly upon that of the stain solution.

This simple and reliable technic gives permanent colored filters with spectra exactly similar to those of the stains.

- 1 Acknowledgments. Authors are greatly indebted to Dr J. C. Healy for his kindness in supplying the spectroscope and for useful advice and discussions and Mr H. Crawford for reviewing the manuscript.
- 2 Plastic flasks from: Greiner and Son, rue de l'Industrie, 67240 Bischwiller, France; or: Falcon, 1950 William Drive, Oxnard, California 93030, USA.
- 3 Chappard, D., and Laurent, J. L., *Experientia* 35 (1979) 708.

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A method for semi-permanent temporary salivary gland chromosome squashes in Diptera¹

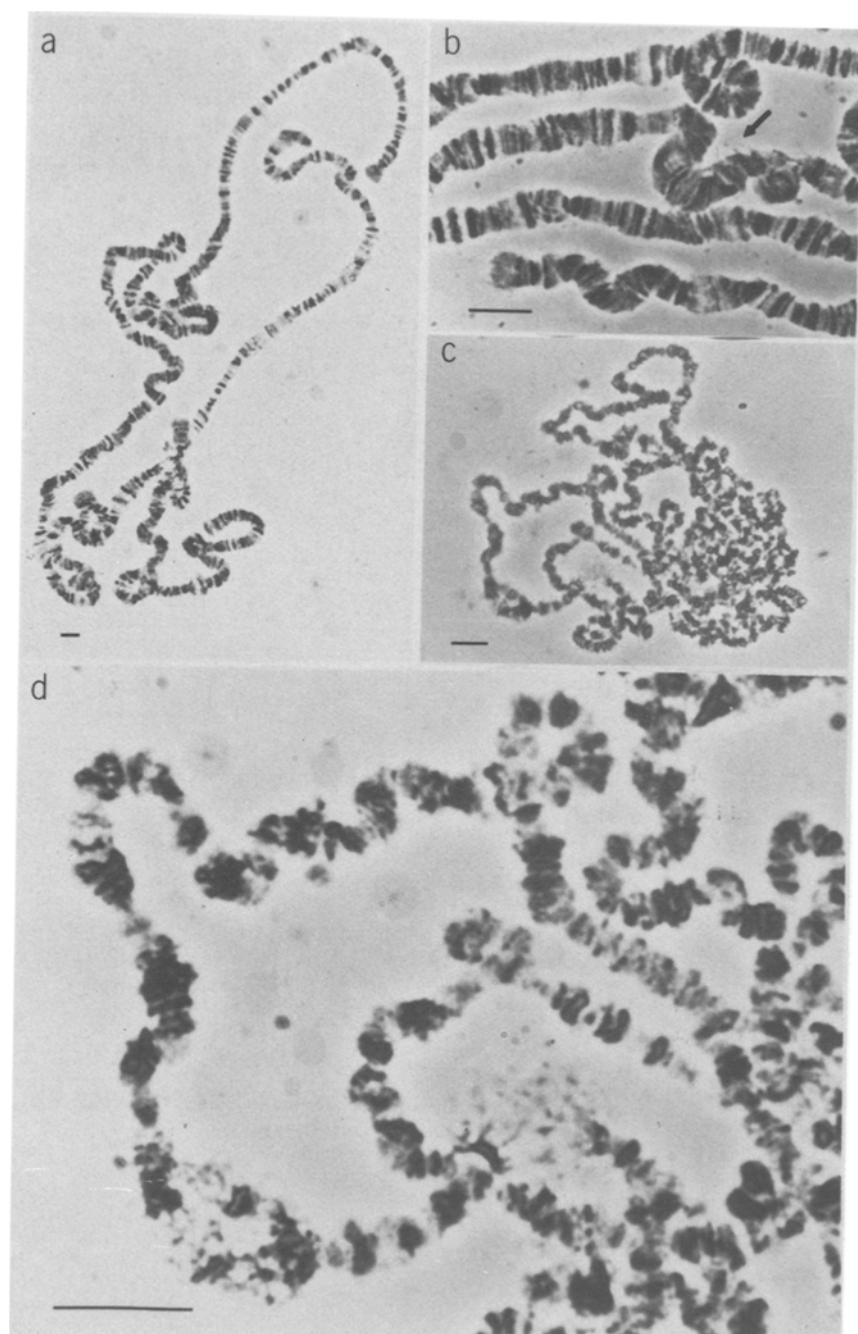
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Department of Biology, University of California-Los Angeles, 405 Hilgard Avenue, Los Angeles, (California 90024, USA), October 22, 1981

Summary. A method to preserve the temporary preparations of Dipteran salivary gland chromosomes during longer periods has been described. Departures from the usual procedures are: absence of heat treatment, destaining of cytoplasm with lactoacetic acid and the use of 'rubber cement' as the sealing material. This procedure preserves the banding details, ectopic pairing and clear background for exceptionally longer periods.

Since the time of Bridges³, temporary squash preparation of salivary (polytene) gland chromosomes have been the most useful way to study the banding pattern of normal and rearranged chromosomes in *Drosophila*, *Sciara* and

other Dipterans. Several improvements of this method⁴⁻⁹ have been made to increase the resolution of the bands against the background, but no attempt has been reported, except one¹⁰, to prolong the life of temporary squash



Salivary chromosomes of Diptera prepared following the reported technique. Notice the cytoplasm free background and the banding detail. Salivary chromosomes of *Drosophila melanogaster* (a) and *Culex pipiens pipiens* (c) in a cytoplasm free background. Banding details in *Drosophila* (b) and in *Culex* (d). Arrow indicates ectopic pairing sites. Scale bar 10 μ m.

preparations. However, methods have been devised to make the preparations permanent¹¹, which doubtlessly decrease the 'freshness' of the temporary preparation.

Procedure and results. We checked most of the existing temporary methods used in *Drosophila* to find some simple way to make squash preparations of comparable quality and longer life. Finally, we came up with a simple and inexpensive temporary method where the preparation can be stored for a longer time with excellent banding detail including ectopic pairing and clear background (see figure), which is reported in this communication. This method is also suitable for *Culex pipiens pipiens* salivary gland chromosomes (fig., c and d).

The major modifications of the mostly used temporary

method⁸ are: a) No temperature treatment, b) washing of excess stain with 45% acetic acid, c) treating and squashing of dispersed cells in lacto-acetic acid at room temperature for destaining of the cytoplasm and d) use of 'rubber cement' as the sealing material to allow refilling of dry edges for longer life as and when required.

The complete procedure is as follows: 1. Clean slides in 95% alcohol and keep them dust free. 2. Dissect out a pair of gland in 0.7% NaCl. 3. Transfer the gland to the pre-cleaned slide and fix in a drop of aceto-alcohol (1 part glacial acetic acid:3 parts absolute ethyl alcohol) for 5 sec. 4. Put a drop of 2% lacto-aceto-orcein⁸ on the gland and stain for 20 min (older stain must be filtered before use). 5. Wash out the stain with 45% acetic acid. 6. Wash the gland with lactoace-

tic acid (1 part 85% lactic acid:1 part glacial acetic acid) at least twice and place a square cover glass over the gland. 7. Break the gland by tapping the coverglass several times with a needle, being careful not to move the coverslip; keep the slide untouched for 2-3 min to allow cytoplasm to destain in lacto-acetic acid. Squash with moderate pressure under VWR bibulous paper or Whatman filter paper; for mosquito salivary gland chromosomes squash with as least pressure as possible. 9. Seal the edges of the coverslip with Carter's rubber cement (The Carter's Ink Co., Cambridge, Mass. 02142, USA). 10. Store the slides in the refrigerator or a place with adequate (80%) humidity. Check slides at 3-month intervals to see whether edges are dried out; if so rub-off sealing cement, put a drop of lacto-acetic acid near the dry area on the slide, wait until the space is filled with the solution and reseal.

This method is now being used routinely in our laboratory to make long lasting temporary preparations of X; Y translocations of *Drosophila*. We found it is useful to retain preparations for comparison with new material or as additional questions arise. The clear background and banding details with ectopic pairing of 15-month-old preparations are still of high quality with no noticeable change.

- 1 The author wishes to express his appreciation to Dr John R. Merriam for invaluable suggestions throughout the course of this work and for his comments on the manuscript. The author also likes to thank Drs R. Barr and P. Guptavanij of UCLA Public Health for the supply of *Culex* larvae for chromosome preparations. This work was supported by US Public Health Service Research Grant No. AG-01871 to JRM.
- 2 Reprint request should be addressed to Zoological Survey of India, 34 Chittaranjan Avenue, Calcutta 700012, India.
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Announcements

Prizes

The executive committee of the Swiss Chemical Society will announce the winners of the **Werner- and Paracelsus Prizes** at its spring conference in 1984. Nominations and applications for these prizes must be submitted to the jury by May 31st 1983. The address of the secretary to the Swiss Chemical Society is: O. Rohr, CIBA-GEIGY AG, R-1047.1.04, 4002 Basel.

This year's **Henry E. Sigerist Prize**, which is sponsored by the Swiss Historical Society for Medicine and Natural Sciences, will be awarded on the 14th and 15th of October in Delémont and Porrentruy. Students, doctoral candidates and research assistants at Swiss technical schools and universities wishing to compete are asked to submit their entries (dissertations or manuscripts published or completed in 1982) not later than May 13th 1983, to the president of the jury: Prof. Dr. med. Carl Haffter, Medizinhistorische Bibliothek, Klingelbergstrasse 23, CH-4031 Basel.

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Full Papers (in-depth reports not exceeding 4-6 printed pages)

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Short Communications (1-2 printed pages)

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Authors are requested to specify under which section heading they would wish their communication to appear:

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