Cytological technique

Painter, T.S. Aceto-carmine technique for salivary chromosomes. The writer has worked almost exclusively with temporary aceto-carmine mounts and the commments given below apply only to that type of preparation.

Much difficulty has been experienced in getting a good iron aceto-carmine stain. Powdered carmines, from many different sources, have been tried including the certified product of Coleman and Bell, Grübler's Karmin Rubrum Opticum, and Carmine 40 from several sources. The trouble has been either that the stain would not take or was not selective enough or else it would not keep, at ordinary room temperature here in Texas for more than a few days without going bad. At present we are using some uncertified carmine manufactured by Coleman and Bell and are obtaining a satisfactory stain in the following way. In one flask an excess of carmine is simmered, under a reflex condenser, in 40 per cent acetic acid, for an hour or two, when it is cooled and filtered. In another flask carmine is boiled in 60 per cent acetic acid similarly, and after cooling is filtered. A trace of ferric acetate is added to both stock solutions. From time to time, as needed, the two stains are mixed in equal proportions. We are unable to explain why this procedure gives a good stain but it works.

The salivary glands are dissected out in Ringer's solution (cold-blooded or frog formula) and transferred to a clean slide with a pipette. The Ringer's is quickly removed with a pipette and iron-aceto-carmine is flowed over the glands from one side. After a few moments the first stain is removed and fresh stain is added in considerable excess. The slide is put to one side and allowed to stand until a little of the carmine begins to precipitate around the edge, a matter of 15 to 25 minutes depending on atmospheric conditions. A cover slip is now placed over the glands, and the excess stain removed with a pipette and filter paper. Being careful not to let the coverslip move, the preparation is next blotted with a good deal of pressure, a process which usually frees the individual nuclei from the surrounding cytoplasm. Under a dissecting binocular, the individual nuclei are crushed with a blunt needle, by the pressure applied locally to the cover slip, and then after blotting the slide once more to remove all traces of the stain the coverslip is sealed with vaseline or melted paraffin.

The type of light filter used for the examination of preparations is the BG 7 optical glass filter put out by Zeiss.

Marshak, A. A rapid method for making permanent mounts of Drosophila salivary gland chromosomes. (1) A saturated solution of aceto-carmine is prepared by boiling carmine in a 45 per cent aqueous solution of glacial acetic acid for several hours. A reflux condenser is attached to the flask containing the solution in order to prevent changes in concentration by evaporation. A clear dark red solution is ob-