9. The use of pectinase for root tip squash preparations.

Considerable difficulty has been encountered in attempting squash preparations of maize root tips by means conventional for many other plants. Tips squashed directly in carmine or orcein stain following fixation in acetic alcohol and hydrolysis in N HCl yield solid clumps of tissue from which individual cells are not easily freed. Furthermore, the cell walls are fragile and commonly rupture when flattened. Pectinase (Nutritional Biochemicals Corp., Cleveland, Ohio) used according to the procedure described by Östergren and Heneen (Hereditas 1962) for other grasses alleviates the principal difficulties. Partial dissolution of the middle lamella permits ready dispersion of the meristematic region into small groups or individual cells and cell walls are softened such that whole cells may be flattened intact. The following simplification of Östergren and Heneen's technique has been found satisfactory for chromosome counts and tentative identification of several chromosomes. Other of their suggestions may be desirable for detailed study of chromosome morphology.

1. Pretreat tips for four hours at room temperature in 0.002 M aqueous 8-hydroxyquinoline.

2. Fix overnight in 3:1 acetoalcohol (Fixed roots may be stored for at least several weeks at -15°C).

3. Hydrolyze 8 minutes at 60°C in Normal HCl.

4. Either stain with Feulgen reagent for 2 hours then treat with 5% pectinase in distilled water for 2 - 2 1/2 hours at room temperature and squash in 45% acetic acid or treat directly in pectinase and squash in propiono-carmine.

K. V. Satyanarayana
J. L. Kermicle