

and Illinois A with an inbred line of Luces Favorite. Individual tetraploid plants induced in this three-way hybrid by the heat treatment technique of Randolph² were mass pollinated during two subsequent generations and from this tetraploid strain samples of grain from 10 ears selected at random were taken for analysis. Diploid sister plants of the original tetraploids were intercrossed in a similar manner for two generations to provide a comparable diploid strain for comparison with the tetraploid.

For the fractionation and determination of the carotinoids the procedure of Kuhn and Brockmann³ was adopted with certain modifications. The pigments were extracted directly from the corn meal with anhydrous methyl alcohol, saponified and fractionated

with petroleum ether. For determining concentration we used a photoelectric colorimeter equipped with Corning glass filters 428 and 585 and calibrated against standard solutions of crystalline beta carotin.

We attribute the observed differences between diploid and tetraploid yellow corn to quantitative rather than qualitative gene differences, since the comparison was made between strains having a common origin and an essentially identical genetic constitution. There is a possibility that tetraploid corn will be of practical importance due to its increased vitamin A activity.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

SIMPLIFIED SCHAEFFER SPORE STAIN

BEGINNING students of bacteriology frequently find it difficult to follow the original Schaeffer¹ technique, which calls for heating a flooded slide over an open flame, without breaking the slide. Consequently, the following technique was first worked out for beginning students in bacteriology at the Agricultural and Mechanical College of Texas, and since then, over a period of time and with a variety of cultures, it has given better results than the technique originally described. In addition, the method of drying and the time of staining have been modified.

A simple, inexpensive steam bath, perhaps a tin can or beaker of proper diameter or a metal tray about three inches deep and two inches wide, on an asbestos centered wire gauze, is used to heat the slides.

(1) Smears, prepared from spore suspensions, are dried for staining by laying the slide on the table top near the base of the burner used for heating the steam bath.

(2) Dried slides are placed across the steam bath until definite droplets of water collect on the bottom of the slide.

(3) The slides are then flooded with 5 per cent. aqueous malachite green and left on the steam bath for one minute.

(4) Stained slides are removed from the steam bath with the thumb and index finger of one hand and dropped into cool water. This is done by spanning the length of the slide. The overhanging ends of the slides are cool enough to do this without danger of burns.

(5) The slides are thoroughly rinsed and, while still

wet, are counterstained with 0.5 per cent. aqueous safranin for thirty seconds and again rinsed in cool water.

(6) Rinsed and dried slides are easily examined under the microscope. As in the original Schaeffer staining technique, the spores stain green and the vegetative cells stain red.

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A METHOD FOR FIXING AND STAINING EARTHWORMS

NOTHING furnishes more satisfactory material for classroom work than sections of earthworms properly fixed and stained. In almost every course in general biology or zoology some time is devoted to the histological study of the common earthworm. It is unfortunate that the material used is often poorly fixed and the sections do not show up well. I have found the method given below simple, quick and one which gives excellent results.

Collect several earthworms and rinse off the dirt. Place these in a covered dish and sprinkle a small amount of well-sifted corn meal and powdered agar mixed in equal proportions on the bottom of the dish. Some finely chopped lettuce may also be added. Cover the worms with a moist paper and leave in a cool dark place. Transfer the worms to clean dishes and change the food each day for three days. By this time their alimentary tract should be free of all dirt and grit.

The specimens are fixed by cutting them into pieces about three fourths of an inch long and dropped directly into warm (about 50° C.) Allen's B-15. Fix for twelve hours and then rinse in water and run through 35, 50, 70, 80, 95 and two changes of 100 per cent. alcohol. Leave in each alcohol one hour. Now run

² L. F. Randolph, *Proc. Nat. Acad. Sci.*, 18: 222-229, 1932.

³ R. Kuhn and H. Brockmann, *Zeit. Physiol. Chemie.*, 206: 41, 1932.

¹ Alice B. Schaeffer and McDonald Fulton, *SCIENCE*, 77: 194, 1933.