statistically significant (Table 2). Height reduction was equally attributable to a reduction in the number of nodes and in the distance between nodes. Node was defined as the point on the terminal leader from which a side branch originated. Opposite side branches were counted as one node.

EDNA also reduced the dry weight of red cedar tops. Seedlings sprayed with 1000 and 4000 ppm weighed only 86% and 51% as much as the control. Because original dry weights are included, these figures underestimate actual differences in growth.

At 4000 ppm, EDNA reduced daily growth successively more during the first three measurement periods than later (Table 3). After 44 days the inhibitory effect began to lessen but daily growth was still retarded at 93 days. After the respraying at 90 days, growth once again successively decreased. At 1000 ppm EDNA, the inhibition effect wore off after 30 to 60 days and the second spraying failed to reinstate height retardation. Low and medium treatments of EDNA had no significant effect on growth of red cedar. Red cedar leaf color was darker green and more blue for seedlings treated with the very high levels.

For arborvitae high and very high levels of EDNA reduced height growth to 64% and 12% of the control (Table 2). High and very high rates of EDNA reduced dry weight to 91% and 46% of control. Applied at 4000 ppm, EDNA strongly inhibited growth for the entire 90 days before respraying. At the 1000 ppm level (Table 3), EDNA's effect began wearing off after 45 days and was gone after 60 days.

Following the initial spraying, the terminal meristems of arborvitae browned on two of the 15 seedlings given 4000 ppm EDNA. The two plants subsequently died. Low and medium treatments of EDNA had no significant effect on growth. EDNA had no effect on color.

In summary, while EDNA failed to produce hoped for increases in the growth of red cedar or arborvitae it proved to be an effective growth retardant. As such, EDNA may have commercial value in landscaping and nursery production. Where controlled size is desired, spraying may be more economical than shearing, and side effects on color and plant form may be of aesthetic interest. The long term persistence of the chemical and the apparent ease with which it entered the plants are favorable attributes for a retardant. Future work is planned to screen EDNA on other conifers and to conduct field trials on red cedar and American arborvitae.

An Effective Technique for Chromosome Cytology in the Carnation (*Dianthus caryophyllus* L.)

Gene S. Howard and Roger D. Uhlinger[1]

Abstract

A fast, simple consistent technique for carnation chromosome cytology using a modification of Snow's technique and phase contrast microscopy is reported. Sample photomicrographs are shown.

Cytological investigation in carnation has been limited because of small chromosome size and the difficulty of chromosome staining. Because complexity of the techniques published by Mehlquist (2) and Brooks (1) limits their usefulness, we developed this simple and consistent technique. It gives uniform results when used on carnation and on Dianthus hybrids derived from a hexaploid species with 2n=90. We do not recommend pretreatment with colchicine or paradichlorobenzene. Our technique is a modification of the method by Snow (3). Those with light microscopes should try Snow's method.

Alcoholic – Hydrochloric Acid–Carmine Stain (Snow's Formula), modified.

Add 1 ml concentrated HCl to 15 ml distilled water then bring to a gen-

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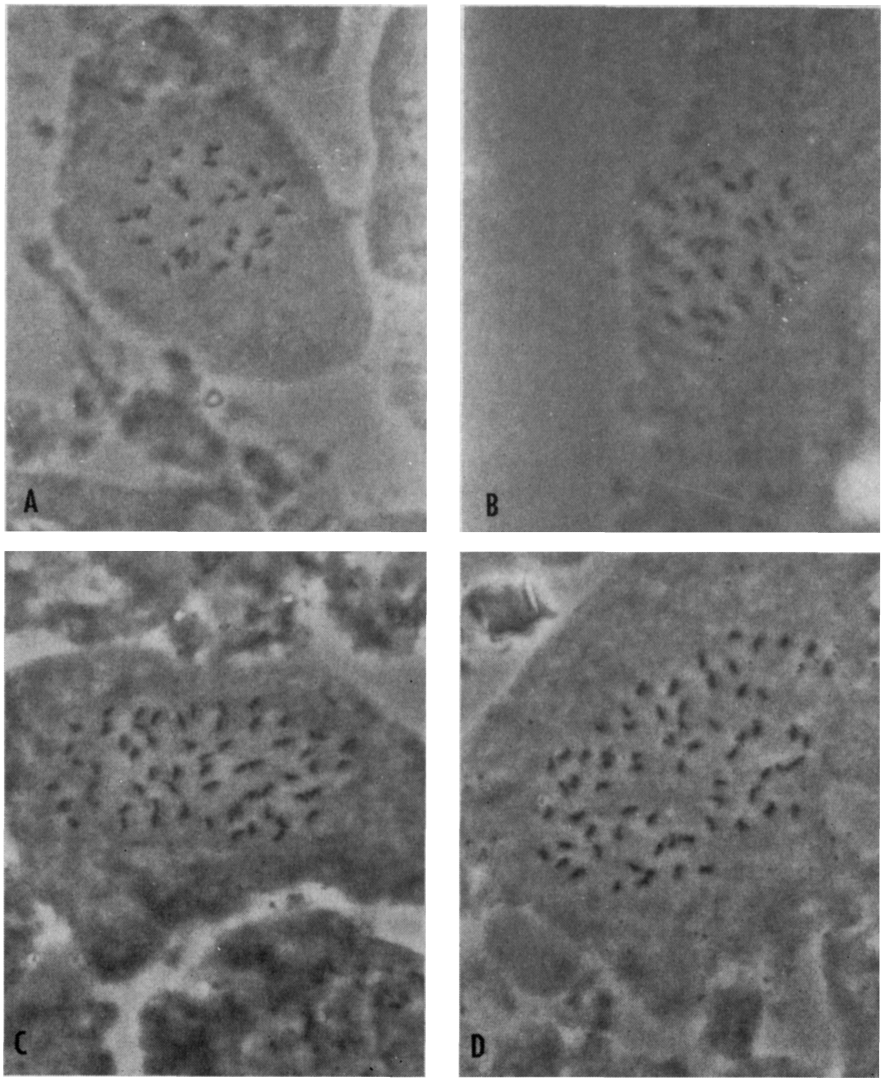

Fig. 1. Characteristic carnation chromosomes, using the technique described. (a) Diploid Littlefield, 30 chromosomes; (b) Diploid Malmaison Giant, 30 chromosomes, (early anaphase showing sister chromosomes); (c) Tetraploid Sir Arthur, 60 chromosomes; (d) Aneuploid (Tetra Sim x Cheyenne), 67 chromosomes.

tle boil. Stir in 1 gm certified carmine (Alum Lake–C. I. No. 75470) while boiling for 10 minutes. After cooling, add 95 ml of 85% ethanol and filter.

PROCEDURE

1. To fix tissue place in McClintock's fixative (1 part glacial acetic acid to 3 parts absolute alcohol), from 1 hour to 8 days.
2. Wash out fixative for ½ hour or longer in 70% ethanol.
3. Store in 70% ethanol for up to 7 days. (The actively dividing cells become somewhat softer when stored, thus less pressure is required to avoid fracturing cells when squashes are made.)
4. Transfer tissue (root tips or anthers) into enough stain to cover for 20 to 40 minutes.
5. Rinse and mount in 45% acetic acid.
6. Seal with clear nail polish for examination, and with thin balsam over the polish for semi-permanent mounts.
7. Use stain again and protect it from evaporation.
8. During mounting spread cells by applying pressure to cover slip with tweezer tips.
9. Heat over an alcohol burner to remove excess acid and assure flattening of cells.
10. Precipitates and debris will be at a minimum.
11. Scan under a *phase contrast* microscope at 250x. Count and photograph at 630x.

This technique should be useful to researchers and students because of the ease of obtaining root tips from carnation cuttings at any time of the year.

Typical examples of carnation chromosome orientation are shown in Fig. 1.

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Procedure for Determining Intercellular Space of Roots and Specific Gravity of Sweetpotato Root Tissue [1]

L. J. Kushman[2] and D. T. Pope[3]

Recent studies showed that at harvest sweetpotato storage roots contain as much as 10 milliliters of intercellular space per 100 milliliters of root; during storage intercellular space increases to the extent that it becomes visible and is classified as pithiness or internal breakdown (*1*). A preliminary report shows that by accounting for the intercellular space tissue specific gravity values can be computed and correlated with dry-matter content in much the same manner as for Irish potatoes (*2*). At harvest intercellular space for each of four varieties was relatively constant for a given variety and differed significantly among varieties (*4*).

A rapid method to measure intercellular space and dry matter would be useful in storage studies and in seedling and variety evaluations in breeding programs. Certain variables which influence the measurements have been evaluated recently (*3*). A procedure which produces consistent results regardless of variety, length of cure, and other variables is described below.

Wash and surface dry roots. Remove all sprouts, rootlets, and foreign matter.

Number each end of each root. A marking pen will work if the root surface is dry.

Weigh each root in air in grams to the nearest 0.1 gram.

Weigh each root in water in grams to the nearest 0.1 gram. It is suggested that a wire pan and a wire cover be suspended from the bottom catch of a Mettler or similar scale with the pan and cover tared in water to show 20.0 grams on the scale. Remove the cover, place the root in the wire pan and replace the wire cover to hold the root under water since many roots will float in water (Fig. 1).

Subtract 20.0 grams from the scale reading to obtain net weight (or buoyancy as negative values). Subtract net weight in water from weight in air to obtain volume of root in milliliters if the root is heavier than water. Add buoyancy to weight in air to obtain volume of root if the root floats in water.

This ignores small errors due to water temperature and dissolved substances or contaminants that modify the specific gravity of the water, small bubbles attached to the root, amount of wire suspending the pan which displaces water and small changes in root volume during handling. Reasonable effort should be made to keep these errors small.

Cut each root in X section and hold under water while subjecting the roots to a vacuum of 27 or more inches of mercury for 20 minutes. Release the vacuum and hold the roots in water for 20 minutes to allow all voids to fill with water.

Pour off water and quickly assemble root halves to match up pieces originally in the same root. Protect the root pieces from rapid drying.

Weigh each root in water again. Since all roots sink in water after evacuation, no tare weight is needed. Subtract the weight in water after evacuation from the original weight in air to obtain root tissue volume.

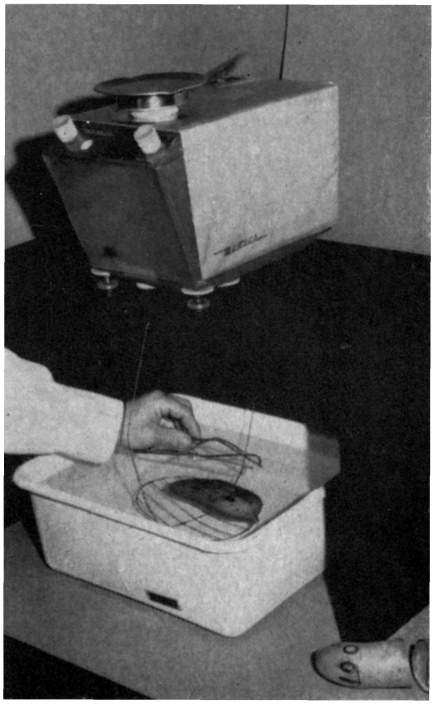

Fig. 1. Placing heavy wire cover on floating sweetpotato to permit weighing in water. Note numbered root in foreground cut in X-section in preparation for evacuation to remove air from intercellular spaces.

Three calculations are possible:

a. root specific gravity equals weight in air divided by root volume;
b. tissue specific gravity equals weight in air divided by root tissue volume;
c. intercellular space per 100 milliliters of root volume equals weight in water after evacuation less weight in water before evacuation (add buoyancy) divided by root volume times 100.

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