study on fresh weight changes, fully expanded mature leaves were harvested from both phenotypes and kept in the dark for 24 hr. One cm discs were than cut from the leaves with a cork borer and these discs were washed for 1 hr in running tap water, blotted dry and weighed. The discs were placed in 6 cm petri dishes containing Kuraishi's solution without kinetin and kept in the dark at 22 C. Samples were removed and weighed after 8, 16 and 24 hr. The results of this experiment are presented in Figure 5. It appears that the leaves from green plants showed a greater increase in fresh weight than the tan plants. This data would tend to substantiate the hypothesis that the green plants contains higher endogenous levels of some kinetin-like substance.

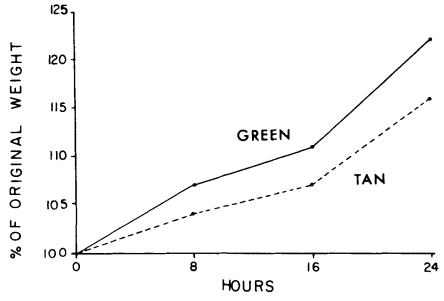

Fig. 5. Fresh weight increase of leaf discs of green and tan phenotypes with time.

Literature Cited

1. Anonymous. 1967. Amchem 66-329, a new plant growth regulator. Information Sheet 37. Amchem Products, Inc., Ambler, Pa.
2. Chatterjee, S. K. and A. C. Leopold. 1964. Kinetin and gibberellin action on abscission processes. *Plant Physiol.* 39:334-337.
3. Honma, S., J. C. Bouwkamp, and M. J. Stojianov. 1968. Inheritance of dry pod-color in snap beans. *J. Heredity* 59:243-244.
4. Kuraishi, S., and S. Okumura. 1956. The effect of kinetin on leaf growth. *Bot. Mag.* (Tokyo) 69:300-306.
5. MacKinny, G. 1941. Absorption of light by chlorophyll solution. *J. Biol. Chem.* 140:315-322.
6. Osborne, D. J., and S. E. Moss. 1963. Effect of Kinetin on senescence and abscission in explants of *Phaseouls vulgaris. Nature* 200:1299-1361.

Differentiating Diploid from Tetraploid Seedlings of *Asparagus officinalis* L.[1]

J. Howard Ellison and Elito S. Tiangco
Rutgers University, New Brunswick, New Jersey

Abstract. **Tetraploid asparagus seedlings were produced from diploid seeds by means of colchicine treatment. Stomata length and cladophyll width were reliable characters for identifying diploids and tetraploids, but number of cladophylls per whorl and cladophyll length were unreliable.**

Efficient identification of tetraploid seedlings is a problem in asparagus. Braak and Zeilinga (1) claimed to distinguish tetraploid from diploid asparagus by the longer cladophylls and darker green color of the former, but did not present supporting evidence. The present paper reports, primarily, an attempt to determine characters in asparagus (stomata length, number of cladophylls per whorl, or length and width of cladophylls) that would conveniently and reliably distinguish diploids from tetraploids.

Diploid progenies of 5 crosses were used. The method of colchicine treatment was a slight modification of the one described by Braak and Zeilinga (1). Seeds were sterilized by soaking in a concentrated solution of sodium hypochlorite for one minute, washed with distilled water and then incubated in petri dishes on moist filter paper at 30 C for 4 to 7 days until radicles emerged. When radicles were 4 to 6 mm, seed were put in vials containing a 1.5% aqueous solution of colchicine which was placed inside a desiccator under vacuum for 20 minutes. The seeds were soaked for an additional 10 minutes under normal atmospheric pressure and then washed before sowing in peat pots.

Chromosome counts were made from root tips, pollen mother cells, and apical buds from young shoots. A hot-water bath was useful in stimulating root development of some seedlings.

In each case, the specimens were fixed in Carnoy's fixative, modification III (2), between the hours of 10 AM and 2 PM to insure the presence of numerous dividing cells. The staining procedure was adapted from Snow's squash preparation (4) using alcoholic hydrochloric acid-carmine as stain. After 24-hour fixation, the materials were washed with 3 changes of 70% alcohol, allowing at least one hour for each change. The tissues were drained in bibulous paper and then soaked in a specially prepared solution of aceto-carmine for 36 to 48 hours. The specimens were rinsed with 70% alcohol and squashed between the slide and cover slip. Only plants with 80% or more tetraploid cells in tissues examined were included as tetraploids. Varying numbers of tetraploids from the different crosses and treatment dates were obtained.

Samples of cladophylls for stomata measurement were gathered from the tenth whorl from the tip of one of the main stalks. Four or five cladophylls from each plant were fixed, using a combination of methods suggested by Clarke (3) and by Speckmann *et al.* (5). They were placed in vials with 70% alcohol and put in a bath of boiling water until most of the chlorophyll was extracted, transferred to vials containing 85% lactic acid and boiled for approximately 10 minutes, and finally stored in lactic acid for microscopic examination.

The guard cell lengths from 5 stomata of each of the 2 cladophylls from each plant were measured in sub-units of 2.5μ. Cladophylls were counted from 10 successive whorls of the central portion of the main shoot of each plant. One randomly selected cladophyll from each of the 10 whorls was measured for length in millimeters and for width in microns. The data analyzed were composed of means of 10 measurements.

Stomata length and cladophyll width of colchicine induced tetraploids were considerably larger than untreated diploids (Table 1). There was no overlap in ranges of diploids and tetraploids in stomata length and cladophyll width among individual plants. The data are means of five progenies, and there was no progeny x ploidy interaction; thus the results were not presented by progenies.

Chromosome counts were made from root tips from each plant but not from apical buds from all plants or from pollen mother cells of all male plants (not all of the plants had flowered). However, apical buds were studied from enough plants to indicate complete agreement with root tip counts, and there was complete agreement between root tip ploidy and stomata length and cladophyll width. We believe this is good evidence that no chimeras were involved.

The suitability of using stomata or cladophyll width or both in differentiating asparagus tetraploids

[1]Received January 12, 1970. Paper of the Journal Series, New Jersey Agricultural Experiment Station, Department of Horticulture and Forestry.

from diploids seems to be justified. Some small but statistically significant effects of seedling age were manifested among the tetraploids. Nevertheless, these effects were overridden by the large differences between diploids and tetraploids. In the population of seedlings in this study, which included five different progenies, no plant would have been misclassified in the basis of either stomata length or cladophyll width.

The assurance provided by using either of the two above characters in detecting colchicine induced tetraploids is greater than that reported by Speckmann *et al.* in rye grass (5), where overlap in stomata length was recorded. Although there were some differences between $2n$ and $4n$ seedlings in number and length of asparagus cladophylls, there was so much variation in the patterns as to make these two characters unsuitable for screening purposes. These results, regarding length of cladophylls, do not support the observation of Braak and Zeilinga (1).

Table 1. Characters used in classifying asparagus diploids and colchicine induced tetrapolids.

Character	Ploidy level			
	Diploid		Tetraploid	
	mean	range	mean	range
Stomata Length (μ)	21.3	(18.8 - 24.0)	34.2**	(25.8 - 46.8)
Cladophyll width (μ)	265.2	(217.0 - 295.5)	382.2**	(309.5 - 488.0)
Cladophyll length (mm)	18.1	(11.9 - 26.0)	21.0	(9.3 - 38.2)
Cladophyll number per whorl	6.1	(4.6 - 8.4)	5.0	(2.4 - 11.5)

** Significant from diploid at 1% level.

Literature Cited

1. Braak, J. P., and A. E. Zeilinga. 1957. Production of a colchicine-induced tetraploid asparagus. *Euphytica.* 6:201-212.
2. Brooks, R. M., M. V. Bradley, and T. I. Anderson. 1963. Plant microtechnique manual (mimeographed). University of California, Davis. 70p.
3. Clarke, J. 1960. Preparation of leaf epidermis for topographic study. *Stain Tech.* 35:35-39.
4. Snow, R. 1963. Alcoholic hydrochloride acid-carmine as a stain for chromosomes in squash preparations. *Stain Tech.* 38:9-13.
5. Speckmann, G. J., J. Post Jr., and H. Dijkstra. 1965. The length of stomata as an indicator for polyploidy in rye grass. *Euphytica.* 14:225-230.

The Inheritance of Mature Fruit Color in *Capsicum pubescens* R. & P.[1]

Gaafar M. El Hassan and Paul G. Smith

University of California, Davis

Abstract: **Red and yellow mature fruit color in *Capsicum pubescens* is controlled by a single gene with yellow (y) recessive to red (y^+).**

In mature fruits of *Capsicum annuum* L. yellow color (y) is recessive to red (y^+) (1,2,3,4,5,7). This report deals with the inheritance of red and yellow color of *C. pubescens* R. & P., a cultivated species of South American origin. The yellow (or orange yellow) color of *C. pubescens* is different from the lemon yellow of *C. annuum* L. referred to by Kormos (3), but may be similar to his orange color. Both the yellow (or orange) and the lemon yellow fruit colors are known in *C. pubescens, C. pendulum, C. chinense* and *C. annuum.*

A yellow-fruited, self-compatible line of *C. pubescens* (SA265) was crossed with a red-fruited self-incompatible line (SA359) of this species in the greenhouse. All the F_1 plants were red fruited and self-compatible. F_2 and backcross generations were obtained by selfing F_1 plants and crossing them to SA265 plants, respectively. Plants in the two segregating generations were scored for fruit-color with results show in Table 1. The Chi square values indicated that yellow and red mature fruit colors of *C. pubescens* are controlled by a single gene with yellow (y) recessive as in other species of *Capsicum.*

Table 1. Segregation for mature fruit color of a cross involving *C. pubescens.*

Generation	Total no. of plants	Mature fruit color		Expected ratio	X^2	P
		Red	Yellow			
P_1(SA359)		all				
P_2(SA265)			all			
F_1		all				
F_2	42	33	9	3:1	0.127	$>.70$
BC(F_1 x SA265)	21	13	8	1:1	0.782	$>.30$

Because of the high degree of cross sterility of *C. pubescens* with other cultivated species (6) no attempt has been made to determine whether the genes in the different species are allelic. The yellow color of mature fruits in *Capsicum* has been observed only once in wild material of *C. chinense* but not in the wild forms of the other species. Perhaps the presence of yellow mature fruits in these 4 species is a product of parallel evolution which occurred following demostication.

Literature Cited

1. Atkins, W. R. G. and G. O. Sherrard. 1915. The pigments of fruits in relation to some genetic experiments of *Capsicum annuum* L. *Sci. Proc. Roy. Dublin Soc.*, N. S. 14:328-335.
2. Khan, A. R. and M. Munir. 1954. Inheritance studies in the common chillies (*Capsicum annuum* L.). *Proc. Sixth Pakistan Sci. Conf., Karachi*, 1954. Part III, pp. 14-16 (Abstr.).
3. Kormos, J. 1954. (Inheritance of carotenoid pigment.) *Ann. Inst. Biol. Tihany* 22:253-259.
4. Shaw, F. J. F., and A. R. Khan. 1928. Studies in Indian chillies. I. The types of Capsicum. *Indian Dept. Agric. Hem., Bot. Ser.* 16:52-82.
5. Smith, P. G. 1950. Inheritance of brown and green mature fruit color in peppers. *Jour. Hered.* 41:138-140.
6. Smith, P. G. and C. B. Heiser. 1957. Breeding behavior of cultivated peppers. *Proc. Amer. Soc. Hort. Sci.* 70:286-290.
7. Webber, H. J. 1911. Preliminary notes on pepper hybrids. *Ann. Rep. Amer. Breed. Assoc.* 7:188-199.

[1]Received for publication November 24, 1969.