SPECIAL ARTICLES

THE ULTRACENTRIFUGAL CRYSTALLIZATION OF TOBACCO MOSAIC VIRUS PROTEIN

WHEN the clear juice pressed from plants[1] infected with tobacco mosaic virus is centrifuged at 25,000 r.p.m. (maximum field = 40,000 gravity), a pellet separates at the bottom of the tube. Under the polarizing microscope this solid is obviously crystalline. Tests show it to contain all but a small fraction of a per cent. of the virus initially present in the juice; we have accordingly sought to determine whether such crystals, obtained by high-speed centrifuging, are the same as those of the chemically[2] prepared virus protein.

One of the most distinctive characteristics of a crystalline species is its x-ray diffraction pattern. That of the chemically purified virus protein is known[3] to be well defined and to consist of many sharp lines. The x-ray method has therefore been chosen to compare the crystals made chemically and by ultracentrifugal precipitation. To do this, 5 cc portions (1) of plant juice clarified by filtration through Hyflo Supercel followed by freezing and a subsequent low-speed centrifuging and (2) of solutions of the chemically purified virus protein were centrifuged for three hours in an air-driven ultracentrifuge.[4] The quantity head containing the tubes of liquid resembled one recently described[5]; it had previously been used for the concentration of pneumococcic antibodies in serum.[6] Crystalline pellets from the bottom of the tubes after centrifuging were mounted directly in the holder of a gas-tight x-ray camera and attached to the usual diffraction outfit[7] installed in a cold room maintained at 36° C. Desiccation or deliquescence of the sample was prevented by filling the camera with helium containing an appropriate amount of water vapor. Measurements of the spacings and estimates of the relative intensities of the principal diffraction lines obtained from crystalline masses prepared (1) by chemical means, (2) by ultracentrifuging a solution of the chemically purified virus protein and (3) by ultracentrifuging the clear plant juice itself are listed in Table I. As the data suggest, the three patterns are indistinguishable one from another.

TABLE I

OBSERVED SPACINGS AND RELATIVE INTENSITIES OF X-RAY DIFFRACTIONS FROM CRYSTALLINE TOBACCO MOSAIC VIRUS PROTEINS

Intensities	Specimens of lines from samples crystallized by Ultra-centrifugation		
	Chemical means*	From solution of purified protein	From plant juice
Faint	37 A.	35 A.	36 A.
Very faint	28	....	....
Medium	20.8	20.7	20.6
Very faint	16.2 }	14.5	14.6
" "	14.2 }		
Strong	11.0	11.2	11.1
Faint	10.2	....	....
Medium	9.2	9.2	9.2
"	7.44	7.43	7.43
Faint	6.5	6.4	....
Very faint	5.7	5.7	....
Faint	5.44	5.45	5.44
Medium (probably not single)	4.95	5.02	4.94
"	4.71	4.72	4.71
"	4.44	4.46	4.46
Faint	4.08	4.10	4.10
Medium	3.88	3.88	3.88

* Data of reference 3. The camera used in photographing the ultracentrifuged samples did not have a sufficient radius and resolving power to record the spacings larger than 40 A.

CONCLUSION

From these observations it is apparent that a crystalline virus protein can be obtained directly by ultracentrifuging the juice of plants infected with the tobacco mosaic disease. The x-ray patterns of this crystalline material and of the protein prepared from the juice by chemical means are indistinguishable; the two substances, must, therefore, be substantially identical.

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A RELATION BETWEEN SEED ATTACHMENT AND CARPEL SYMMETRY AND DEVELOPMENT IN PRUNUS[1]

IN connection with more detailed studies of developmental morphology of deciduous fruits[2,3] and of the effect of embryo development upon fruit development,[4] a relation has appeared between carpel development and the position of seed attachment which is of some interest both in relation to fruit development and to the general topic of symmetry.

The observations cover several hundred specimens from different varieties of peach (*Prunus Persica* Batech), plum (*P. domestica* L., *P. insititia* L., *P. salicina* Lindl., and *P. americana* Marsh.), the sweet cherry (*P. avium* L.), the sour cherry (*P. Cerasus* L.), the apricot (*P. Armeniaca* L.), and the nectarine (*P.*

[1] We are indebted to W. M. Stanley, of the Department of Animal and Plant Pathology of the Rockefeller Institute, for the plant materials used in this study.

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[6] R. W. G. Wyckoff, SCIENCE, 84: 291, 1936.

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[2] H. B. Tukey, *Proc. Am. Soc. Hort. Sci.*, 30: 209–318, 1933.

[3] H. B. Tukey, *Proc. Am. Soc. Hort. Sci.*, 31: 125–144, 1934.

[4] H. B. Tukey, *Bot. Gaz.*, 98: 1–24, 1936.