virus protein solution. 0.2 cc of 0.1 M NaCN and 0.1 cc of toluene were added to all tubes which were then incubated at 37° C. Immediately and at intervals samples were withdrawn for formol titration. At the end of the experiment an equal volume of 20 per cent. trichloracetic acid was added to the remaining solutions, and the amino nitrogen was determined on the filtrates by the Van Slyke method.

Table 1 shows that the papilloma tissue itself undergoes autolysis, but in the presence of the virus protein more carboxyl groups are liberated. Since the virus alone remains stable, this increase in carboxyl groups may be attributed to the hydrolysis of the virus by the papilloma tissue. The results of the amino nitrogen determination on the trichloracetic acid filtrate confirm this; 53 per cent. of the total virus protein nitrogen was liberated as free amino groups. Under exactly similar conditions the virus was not hydrolyzed by papilloma tissue from cottontail rabbits, although the tissue alone showed some autolysis. The virus was also not hydrolyzed when it was incubated with normal domestic rabbit epidermal cells[9] treated in the same way.

It appears that the papilloma virus protein is hydrolyzed by some factor, presumably an enzyme, in domestic rabbit papilloma tissue which was absent in the cottontail rabbit wart tissue thus far studied. In the experiment shown in Table 1 the weight of papilloma tissue was not more than 0.2 gm, and the quantity of virus changed was 1.05 mg. Other experiments gave similar results, and the rate of hydrolysis was not increased by doubling the virus protein concentration. Crystalline horse serum albumen and the macromolecular component of normal chick embryo[10] were not hydrolyzed by the domestic rabbit papilloma tissue. The indication is that the catalyst is specific for the virus protein or similar compounds.

The results demonstrate a mechanism by which the papilloma virus may be degraded as fast as it is formed in domestic rabbit papilloma tissue. It is possible, though no evidence of it was seen here, that such a mechanism may operate in lesser and varying degree also in cottontail rabbits to account for variations in the virus content, especially in experimentally induced growths.[4] The findings are not incompatible with the antigenicity of domestic rabbit wart material, for the antigen could well be a non-infectious, insoluble and possibly partially denatured degradation product of the virus, somewhat analogous to the degraded virus antigen of equine encephalomyelitis vaccines.[11] The possibility exists that a similar mechanism accounts for the absence of virus from the carcinoma based on the papilloma of domestic rabbits,[12] as well as that which derives from papillomas of cottontail rabbits.[13] The cells of the latter may acquire virus-degrading factors or enzymes in the carcinomatous change. It is an obvious possibility that such factors may prevent the recovery of a causative agent from neoplastic growths other than those associated with the papilloma virus.

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THE RESPIRATION OF ELODEA

MEASUREMENTS of respiration on higher aquatic plants are seldom performed. The present investigation, originally to determine the relation of cation intake to gaseous metabolism, provided a better insight into the metabolic implications of a hydrophytic existence. Slight modifications of the Fenn-Ledebur[1] microrespirometer allowed simultaneous measurement of carbon dioxide production and oxygen consumption of excised leaves of *Elodea canadensis* Michx. with facility. A barium hydroxide solution was used to absorb the carbon dioxide, and its conductance was measured at intervals by the use of the Kohlrausch bridge method. Experiments averaged three hours in length and were performed in the dark.

Respiratory quotients were very high (average 8.4), suggesting the possible occurrence of anaerobiosis, possibly in connection with or in addition to the utilization of chemically bound oxygen. Presence of previously stored substrates rich in chemically bound oxygen would decrease the amount of free oxygen necessary for complete combustion, would lower the apparent oxygen consumption, and thus cause the R.Q. to rise. Substances like oxalates and citrates would serve well as such substrates; in fact, calcium oxalate has been identified in cell vacuoles of elodea leaves.[2] Migration of oxalic, citric or other acids to

[9] For this, layers of cells were thinly shaved from the ears and abdominal skin. The results are of dubious significance for the amount of cells thus obtainable is minute.

[10] A. R. Taylor, D. G. Sharp, D. Beard and J. W. Beard, SCIENCE, 94: 613, 1941.

[11] D. G. Sharp, A. R. Taylor, H. Finkelstein, D. Beard and J. W. Beard, *Proc. Soc. Exp. Biol. and Med.*, 43: 650, 1940.

[12] P. Rous and J. W. Beard, *Jour. Exp. Med.*, 62: 523, 1935.

[13] J. G. Kidd and P. Rous, *Jour. Exp. Med.*, 71: 469, 1940.

[1] Wallace O. Fenn, *Am. Jour. Physiol.*, 84: 110, 1928. J. Frhr. v. Ledebur, *Mikrochemie*, Pregl-Festschrift (Sonderband), 253, 1929.

[2] Daniel Mazia and Jean M. Clark, *Biol. Bull.*, 71: 306, 1936.

the surface of the tissue would liberate carbon dioxide from the carbonate-incrusted leaves, also apparently increasing the R.Q. According to Gregory and Sen,[3] the rate of liberation of carbon dioxide may indicate the rate at which the protein cycle, involving deamination and protein synthesis, is taking place.

Chemical storage of oxygen is obviously of advantage to the plant, since it is not only subjected to widely varying degrees of oxygen tension in its aquatic environment, but photosynthetic activity in the winter months is apt to be curtailed. In this connection it was interesting to note the change in gaseous metabolism as the rainy season (1940–41) drew to a close. From January to mid-March the oxygen consumption rose about threefold, the carbon dioxide production neither rising nor falling conspicuously. This change was presumably conditioned by an increase in illumination, causing an increase in photosynthetic activity, in turn causing storage and subsequent breakdown of carbohydrates, and in this manner involving a shift to aerobic oxidation. It is conceivable that the oxygen consumption may also have been affected by the cessation of dilution of the pond water by rain and commencement of concentration of ions by evaporation loss. Further pursuit of these questions should throw more light on the physiology and ecology of such forms.

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ON THE SIZE AND SHAPE OF THE TOBACCO MOSAIC VIRUS PROTEIN PARTICLE

THE proposition that the tobacco mosaic virus protein is molecularly dispersed in aqueous solution has been staunchly defended by Stanley and his coworkers. In response to a paper by Bawden and Pirie,[1] who originally demonstrated that this virus protein has a marked tendency to aggregate, Loring, Lauffer and Stanley[2] announced that the virus protein "has a molecular weight of at least 46 million, a length of at least 430 millimicrons, and an effective diameter of about 12 millimicrons," and that aggregation, if any does occur in the virus protein solutions, is of little significance. Essentially this same point of view has been maintained consistently by Stanley and his coworkers, although in more recent papers, in particular those relating to the research with the electron microscope,[3,4] they speak of a "marked tendency of the particles to aggregate." These workers have avoided consideration of the various anomalies shown by solutions of this virus protein and they consider the size and shape of the protein particles arrived at from ultracentrifuge and viscosity studies as valid and accurate. The electron microscope photographs are cited as substantiating evidence, and the conclusion is reached from a consideration of these photographs that the length of the protein molecule is 280 millimicrons. This conclusion is reached notwithstanding the fact that there are many particles shown in the electron microscope photographs that are considerably shorter than 280 millimicrons.

One might be expected to suppose that all molecules of a given species would be of the same size. In the case of a protein preparation having a heterogeneous size distribution one would suspect either that the preparation is impure or that association or aggregation has occurred, in which case the size of the various particles would be simple multiples of some basic unit. Fortunately in the case of the electron microscope photographs of this virus protein, the sizes of the various particles may be measured, albeit the accuracy is not of a high order. It is impossible, for example, to be sure that all the particles are lying flat, and measurements made from a half-tone reproduction are not particularly satisfactory. Although the errors in the estimations of the lengths of the virus protein particles from these and similar photographs are large, a length distribution curve for 159 particles occurring in the photographs published by Stanley and Anderson[3] and Anderson and Stanley[4] is presented in Fig. 1.

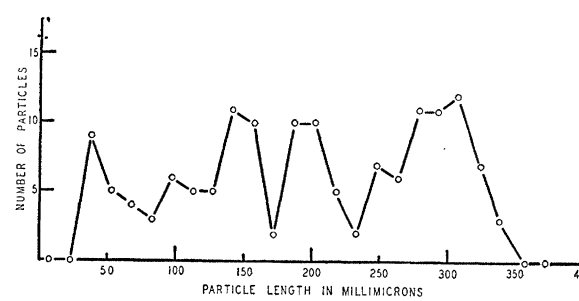

FIG. 1. Length distribution curve for particles of tobacco mosaic virus protein. Data taken from electron microscope photographs.

The class length along the abscissa is 15 millimicrons. Note that a bunching occurs at lengths in the regions of 300, 190, 150, 100 and 37 millimicrons. These lengths are essentially in the ratio of 8 : 5 : 4 : 3 : 1.

The implications of the orderliness indicated above are obvious.

An added item of interest is the observation by Melcher, Schramm, Trurnit and Friedrich-Freksa[5] that the electron microscope photographs of their

[3] F. G. Gregory and P. K. Sen, *Ann. Bot.*, n.s. 1: 521, 1937.
[1] *Proc. Roy. Soc.*, B 123: 274, 1937.
[2] *Nature*, 142: 841, 1938.
[3] *Jour. Biol. Chem.*, 139: 325, 1941.
[4] *Jour. Biol. Chem.*, 139: 339, 1941.
[5] *Biol. Zentr.*, 60: 524, 1940.