(about 2 mv. less than for H_2O) at 5 per cent. D_2O. For higher D_2O content E^0 is an almost linear function of the composition of the solvent.

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THE INACTIVATION OF CRYSTALLINE TOBACCO-MOSAIC VIRUS PROTEIN

THE isolation of a crystalline protein possessing the properties of tobacco-mosaic virus has been described.[1] It has now been found that treatment of this active protein with hydrogen peroxide, formaldehyde, nitrous acid or ultra-violet light produces inactive native proteins that, although slightly altered, retain certain chemical and serological properties characteristic of the virus protein. These inactive proteins do not cause the mosaic disease nor the production of a high molecular weight protein on inoculation to Turkish tobacco plants, and do not produce local lesions on inoculation to *Nicotiana glutinosa* L.

Although the inactive proteins are native and may be taken into solution and crystallized, their solutions are more opalescent than are those of active protein, and they tend to denature more readily than does active protein. The optical rotation of solutions of protein previously treated with ultra-violet light, hydrogen peroxide or formaldehyde is practically unchanged, while solutions of protein inactivated by nitrous acid possess a considerably lower laevo rotation than before treatment. The isoelectric point of protein inactivated by ultra-violet light or by hydrogen peroxide is practically unchanged, while that of protein treated with formaldehyde or nitrous acid is shifted towards the acid side. The amino-nitrogen content of protein inactivated by means of hydrogen peroxide or formaldehyde is considerably lower than that of active protein, and, as would be expected, the protein treated with nitrous acid contains practically no amino-nitrogen. A preliminary determination of the sedimentation constant of protein inactivated by ultra-violet light, kindly made by Dr. Wyckoff and Mr. Biscoe, indicates that no marked change has occurred in the molecular weight; the more diffuse boundary they observe, however, is indicative of a decreased molecular homogeneity. Under the microscope the crystals of the inactive proteins are indistinguishable from those of active protein, and in a preliminary analysis of oxidized protein, kindly made by Drs. Wyckoff and Corey, no conspicuous difference in the x-ray diffraction pattern was found. Mixtures containing varying amounts of active and inactive protein may be prepared, crystallized and recrystallized. The crystals are indistinguishable from those of active protein, but they possess an activity which is slightly less than that which would be proportional to the amount of active protein that they contain. Crystalline protein possessing any desired activity less than that of the regular active protein may be prepared by mixing the active and inactive or by partial inactivation of active protein using any of the four methods mentioned.

In a typical experiment, inactivation of a 1 per cent. solution of virus protein occurred after standing for 5 hours at 27° C. with 5 per cent. formaldehyde at pH 7, 5 per cent. hydrogen peroxide at pH 7, or with 2 per cent. sodium nitrite at pH 3. The amino-nitrogen content was found to have been decreased 60 per cent., 60 per cent. and 99 per cent., respectively, as a result of the treatments. Irradiation of a 0.5 per cent. solution with the full light of a laboratory mercury vapor lamp for 8 hours caused inactivation. The preparations were dialyzed against water at pH 7 immediately after the treatments and were then tested for virus activity.

The sera of animals injected with virus preparations give a precipitate when mixed with a solution containing as little as 10^{-5} gm per cc of inactive protein, and the serum of an animal injected with a solution of inactive protein gives a precipitate when mixed with solutions containing but 10^{-5} gm. per cc. of either active or inactive protein. Thus the precipitin reaction, which has been used as a measure of virus activity,[2] may not be used unreservedly for this purpose, for in the case of inactive protein there is no correlation between precipitin titer and virus activity. Positive precipitin reactions between anti-sera to virus preparations and apparently inactive material have been reported.[3] The serum of an animal injected with protein inactivated by ultra-violet light has a neutralizing effect on tobacco-mosaic virus, which appears to be similar to that previously reported for the sera of animals injected with sap from mosaic-diseased plants.[4]

Vigorous treatment of the virus protein, such as denaturation by means of acids, alkalis or heat, oxidation with potassium permanganate, chromic acid or chloramine-T, or prolonged treatment with concentrated nitrous acid, causes not only loss of virus activity, but also loss of the characteristic properties of the protein, and it is only by means of comparatively mild treatments that inactive native protein, retaining

[1] W. M. Stanley, SCIENCE, 81: 644, 1935; *Phytopath.*, 26: 305, 1936.

[2] T. Matsumoto and K. Somazawa, *Jour. Soc. Trop. Agr.*, 2: 223, 1930; *Ibid.*, 6: 671, 1934; J. M. Birkeland, *Bot. Gaz.*, 95: 419, 1934; K. Starr Chester, *Phytopath.*, 25: 702, 1935; E. T. C. Spooner and F. C. Bawden, *Brit. Jour. Exp. Path.*, 16: 218, 1935.

[3] T. Matsumoto and K. Somazawa, *Jour. Soc. Trop. Agr.*, 3: 24, 1931; F. C. Bawden, *Brit. Jour. Exp. Path.*, 16: 435, 1935; F. C. Bawden and N. W. Pirie, *ibid.*, 17: 64, 1936.

[4] Helen A. Purdy, *Jour. Exp. Med.*, 49: 919, 1929; Kenneth S. Chester, *Phytopath.*, 24: 1180, 1934.

many of the characteristic chemical and serological properties of virus protein, may be obtained. As a whole, the preliminary results indicate that only slight changes occur in the protein molecule on inactivation by the four methods mentioned. Although there is always a possibility, as with any apparently pure substance, that the crystalline tobacco-mosaic virus protein may consist of two closely related components, one active and the other inactive, the available evidence indicates that the virus activity is a specific property of this high molecular weight protein. It appears likely, therefore, that the slight changes in the protein, which result from treatment with formaldehyde, hydrogen peroxide, nitrous acid or ultra-violet light, cause it to lose its ability to infect susceptible plants.

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SUPERIOR INFLUENCE OF THE MOTHER ON BODY SIZE IN RECIPROCAL HYBRIDS

IN previous papers[1] it has been shown that in rabbits and in mice, when races of unlike body size are reciprocally crossed or reciprocally backcrossed, the maternal group of larger body size produces offspring of larger body size. In other words, the mother has greater influence than the father on the body size of the offspring. This might be supposed to be due either to cytoplasmic influence of the egg or to an influence exerted by the mother during gestation. The latter alternative seems to be excluded in the case of some amphibian crosses recently described by Käte Pariser,[2] in which a similar difference is found between reciprocal crosses produced by subspecies of Triton of different body size, but in which the development of the young takes place outside the body of the mother. The crosses made by Pariser were studied primarily with reference to the sex ratio and problems of sex determination, but incidentally they throw light on size inheritance.

The superior influence of the mother is shown with especial clearness in the reciprocal crosses between *Triton palmatus* and *Triton alpestris*. The mean body lengths of metamorphosed individuals of the respective parent species are, *T. palmatus* 26.0 mm, and for *T. alpestris* 37.2 mm. Hybrids produced by *T. palmatus* mothers have a body length of 25.3 ± 0.4 mm, whereas those produced by *T. alpestris* mothers average 29.1 ± 0.3 mm. The difference between these means, 3.8 ± 0.5 mm, is nearly 8 times its probable error, and so, highly significant. It follows that the cytoplasm of the *alpestris* egg at the time of fertilization must contain sources of growth energy much superior to those found in the cytoplasm of the *palmatus egg*. Whether it is legitimate to explain their presence there as a result of previous activity of maternal nuclear material remains to be demonstrated, if indeed this further question is capable of experimental solution. But at any rate an immediate effect of the maternal cytoplasm is clearly shown.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

A LOW COST ELECTROCARDIOPHONE FOR TEACHING PURPOSES

MANY teachers, particularly those teaching physiology and physical diagnosis, recognize the electrocardiophone as an extremely valuable instrument with which to demonstrate heart and respiratory sounds to a large group of students. Until just recently, however, such equipment has been both complicated and costly, keeping many from enjoying its advantages. Recently a new type of microphone has appeared on the market which has opened the field for a simple and inexpensive electrocardiophone. The cost should not run over fifty dollars for the entire instrument.

It is the purpose of this article to describe such a unit, the outstanding features of which are simplicity, compactness and low cost, and which will do almost anything which the more complicated instruments will do.

The basis of this electrocardiophone is the crystal type microphone as sold under the Brush patents. This microphone operates on the piezo-electric principles as defined by Curie in 1880. Thus, if crystals which exhibit pyro-electric properties are subjected to compression or tension, opposite charges of electricity appear at the ends of the crystal; thus a small alternating voltage is generated between two metal places glued at opposite ends of the crystal. The material used for these crystals is Rochelle salts. When a sound is impressed on the crystal the bending strain will set up a voltage between the ends. This voltage is then applied to the grid of a pre-amplifier tube. No polarizing voltage or magnetic field is needed and no input transformer is used. The audio output is almost as large as that obtained from a highly damped carbon microphone. There is no background noise, and the frequency response is good enough. Several carbon microphones were tried and found to be less satisfactory, since the vibrations caused by body movements produced a good deal of rattle and

[1] *Proc. Nat. Acad. Sci.*, 20: 621–625, December, 1934; *Genetics*, July, 1936 (in press).

[2] *Rev. Español de Biol.*, 5: 11–93, 1936.