

Enzymatic Degradation of Desoxyribose Nucleic Acid by Crystalline Desoxyribonuclease

As has been shown in this laboratory, the dielectric increment per gram per litre of desoxyribose nucleic acid in aqueous solution as sodium salt is proportional to molecular weight. Thus, enzymatic degradation of poly-desoxyribose nucleic acid by Kunitz's¹ crystalline desoxyribonuclease (supplied by the Worthington Biochemical Laboratory, Freehold, New Jersey) may be followed by determinations of dielectric constant.

The method used—Fürth's ellipsoid method adapted to high frequencies—is not suited for following the action of the enzyme under its optimal conditions. By increasing the enzyme concentration, however, sufficiently high activity was obtained. The substrate used was sodium desoxyribose nucleic acid isolated from calf thymus by Hammarsten's procedure². Generally, a 0.025 per cent solution containing 0.0001 *N* magnesium ions and 0.005 per cent gelatine has been investigated at a temperature of 20° C. The enzyme concentration has been varied between 0.01 and 0.1 mgm. per ml.

As is shown in the accompanying diagram, four different stages of breakdown are obtained. In the first (I), the increment decreases almost linearly with time. Determinations at various frequencies within the anomalous dispersion region have shown that the average molecular weight decreases with the increment as expected. After 10–20 min. the increment becomes almost stable (stage II). When using high enzyme concentrations there is a sharp change from stage I to II, and the level of the increment which characterizes stage II is in inverse ratio to the enzyme concentration. At low enzyme activities the boundary is diffuse and the increment decreases slowly during stage II. Stage III is characterized by a strikingly rapid decrease of the increment to a considerably lower level. The time when this decrease begins depends upon the enzyme concentration and can take place from 10 min. to several hours after the beginning of stage II. Stage IV constitutes the ultimate condition from a dielectric point of view, there being no detectable change of the increment during 24 hr.

Our experimental conditions are suboptimal and not directly comparable with the conditions usually employed³⁻⁶. A certain correlation is, however, possible. The diagram shows schematically the variation of the relative values of acid precipitability, pH, high-frequency resistivity and viscosity during the different stages.

The genuine 'depolymerase' effect, namely, disaggregation of desoxyribose nucleic acid⁷, appears in

stage I. The almost stationary condition found in stage II may be explained as being due to a splitting off of smaller components—having little effect on the increment—from the desoxyribose nucleic acid. The peculiar rapid change of the increment in stage III seems to be due to an acceleration by the reaction products of the nuclease activity. To investigate this, further additions of desoxyribose nucleic acid have been made at different times. Additions made during stage II lead to mainly additive increment and the enzyme activity remaining unchanged. Additions in the beginning of stage IV result in the new initial stage being at least as rapid as when the enzyme is added to unchanged substrate, while stage II nearly disappears. It cannot be excluded, however, that in stage III a different enzyme is concerned. In stage IV, which is inaccessible to dielectric investigation, important changes occur, with the appearance of dialysable products.

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¹ Kunitz, M., *Science*, **108**, 19 (1948).

² Hammarsten, E., *Biochem. Z.*, **144**, 383 (1924).

³ McCarty, M., *J. Gen. Physiol.*, **29**, 123 (1946).

⁴ Greenstein, J. P., *J. Nat. Cancer Inst.*, **7**, 29 (1946).

⁵ Zittle, C. A., *Arch. Biochem.*, **13**, 191 (1947).

⁶ Zamenhof, S., and Chargaff, E., *J. Biol. Chem.*, **178**, 531 (1949).

⁷ Jungner, G., Jungner, I., and Allgén, L.-G., *Nature*, **163**, 849 (1949).

'Vegetative Hybridization'

MUCH has been written about the experiments and scientific conclusions of Lysenko and his collaborators. There has, however, been almost no attempt at an actual critical confirmation of the Russian results. Thus, although the experiments on 'vegetative hybridization' have formed a large part of the basis for certain theories suggested by Lysenko, there has, as yet, been no published account in Great Britain of a repetition of any of these investigations. In view of this lack of information, it was decided to repeat one of the Russian experiments on 'vegetative hybridization' in the tomato, and the present communication summarizes the results in the year of grafting.

Among the Russian investigators, Avakjan and Jastreb¹ reported changes in tomato grafts, including one in which a variety with red fruit was used as stock and grafted with a pale yellow variety as scion. The scion produced fruits which were red, pink, yellow, or yellow with pink stripes. Some of these colour changes were transmitted to the progeny. Glušenko² also reports changes in both grafted plants and their progeny. An example of a change in the year of grafting is his account of a tomato variety with ordinary tomato leaves grafted on to a tomato variety with potato-type leaves. The developing scion is described as producing both tomato and potato-type leaves.

In the description of actual grafting technique, Lysenko and his co-workers have latterly emphasized the need for taking the graft components at different ages. When one plant is grafted on to another, the plant used as scion, the characters of which it is desired to change, should be very young, whereas

