However, in some individuals with metastases responses were somewhat less, irrespective of whether or not irradiation had been administered. This finding is in general agreement with earlier studies on the universal serologic reaction (*6*).

The data just presented failed to confirm the findings of Evans (*4*), who reported a marked depression of production of H agglutinins following heavy irradiation of individuals receiving a single injection of *S. paratyphi.* However, Evans' titers were in the order of 10^2 times as high as our own, indicating a far more sensitive test system. Negative results in the present investigation may have been due to a masking of irradiation injury by a protracted course of immunization (*7*). Furthermore, it would appear that antibody production may not necessarily parallel resistance to infection (*8*). At any rate, the observed lack of effect on agglutinin formation would not appear to warrant a conclusion that immune defenses were in no way affected by irradiation.

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Changes Induced by Indoleacetic Acid in Nucleic Acid Contents and Growth of Tobacco Pith Tissue[1]

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Studies in this laboratory on interactions of indoleacetic acid (IAA) with purines and pentoses in organ differentiation (*1*) suggested an effect of auxins on nucleic acid metabolism. To investigate this possibility, the effects of IAA on the nucleic acid contents of tobacco pith tissue were examined.

Stems were taken from 3- to 4-ft plants of *Nicotiana tabacum,* Wisc. 38, grown in a greenhouse. After leaves were cut off, the stems were cleaned thoroughly with a cotton wad wetted with 95% ethanol. The bark was then stripped from the stems, leaving a core of pith enclosed within a jacket of vascular tissue. Borings were made longitudinally through 3-cm segments of this pith core with a 7-mm diameter cork borer, and the cylinders of pith so obtained were cut into disks 3 mm thick. After the disks from several stems had been mixed together, they were planted on 50 ml of sterilized medium in 125-ml flasks, 5 to a flask. Sterile tissue cultures were obtained in this way without direct exposure of internal tissue to antiseptics.

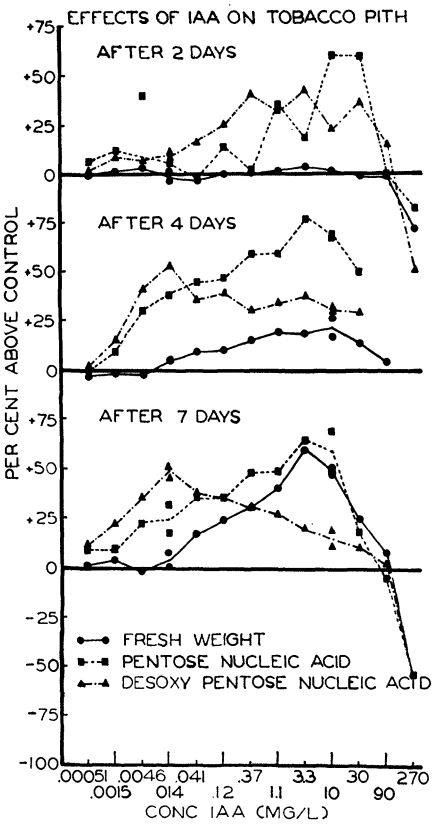

FIG. 1. Changes as per cent of controls in fresh weight, DNA content, and PNA content in excised tobacco pith tissue disks cultured on a sucrose agar medium with serial concentrations of IAA for 2, 4, and 7 days.

The increase in fresh weight of this pith tissue, cultured on an IAA-sucrose-mineral-agar medium, is a function of the supplied IAA over a wide concentration range (*2*). However, as the growth during the first 10 days is not enhanced by the presence of the mineral nutrients (*3*), a simplified medium, containing only Bacto agar, 1%; sucrose, 2%; and IAA was used for these experiments.

At regular intervals, duplicate samples of at least 5 disks each were taken and dispersed in cold 70% ethanol in a Potter-Elvehjem homogenizer. The homogenate was extracted by the method of Ogur and Rosen (*4*), which is a differential perchloric acid extraction procedure for separation of plant pentose nucleic acid (PNA) and deoxyribose nucleic acid (DNA). Quantities of nucleic acid were estimated by measuring optical densities of extracts at 258 mμ in a Beckman spectrophotometer, and they are reported as optical densities of PNA or DNA extracted from one

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disk, dissolved in 2 ml of solvent, and measured in a 1-cm quartz cuvette.

After a number of experiments had shown definite changes in NA contents in response to supplied IAA, effects of IAA over a wide concentration range were explored in two experiments. Figure 1 shows increases over controls of PNA, DNA, and fresh weight with IAA concentration and time. Figure 2 permits com-

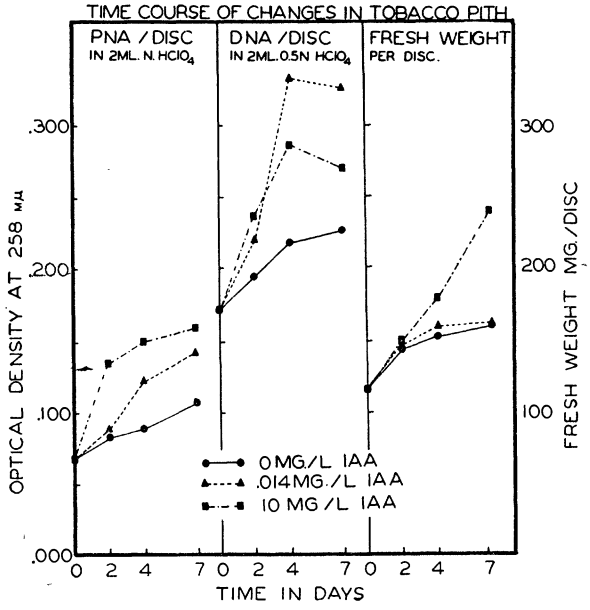

FIG. 2. Fresh weights, DNA content and PNA content of tobacco pith tissue disks cultured on sucrose agar media with 0, 0.014, and 10.0 mg/l IAA.

parison of absolute changes in PNA, DNA, and fresh weight in control disks and disks treated with 0.014 and 10.0 mg/l of IAA. The results are: (*a*) both PNA and DNA contents in the IAA treated pith tissue rise above control values before an increase in fresh weight is apparent; (*b*) nearly maximal effects of IAA on PNA and DNA are reached within 4 days; (*c*) whereas DNA increases most at *ca.* 0.014 mg/l of IAA, the optimal concentration for PNA increase is at least a hundredfold higher; (*d*) with duration of the experiment, fresh weight increase becomes proportional to PNA increase over a wide range of IAA concentrations. The average ratio of PNA to fresh weight at 7 days is 1.08 with an average deviation of 0.08; and (*e*) material fixed and sectioned shows a correlation of cell division with DNA increase at low concentrations of IAA. At concentrations of IAA optimal for cell enlargement and PNA content, no cell divisions are found in spite of increases in nuclear material (*5*).

It should be noted that these changes result from utilization of products in tissues not supplied with nitrogen, phosphate, or organic growth factors. In the presence of such factors that permit rapid proliferation and continuing growth of the tissue, higher nucleic acid contents are found.

Inasmuch as striking zonation of cell division and elongation exists in the plant axis, these differential effects of IAA concentration on the nucleic acids are of special interest because of their morphogenetic implications.

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The Polarographic Investigation of Some Commercially Available Chlorophyllins

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Although the New and Nonofficial Remedies (N.N.R.) and Association of Official Agricultural Chemists' (A.O.A.C.) methods are being used extensively at present for the assay of the chlorophyllins and chlorophylls, they are not as reliable as would be desired. In an attempt to find a more reliable method for assaying these compounds, the use of the polarographic method was investigated.

This method has previously been applied to the study of some extracted chlorophylls by Van Rysselberghe *et al.* (*1*). They reported that the chlorophylls give a reduction wave, $E_{1/2}$ vs. saturated calomel electrode (S.C.E.) = −1.88 v, and believe it to be a hydrogenation reaction of the double bond between carbons 2 and 3 of the phytol side-chain in the chlorophyll molecule. Kolthoff and Lingane, however, state that the actual electrode process involved is not known (*2*). Van Rysselberghe's group also reported that no reduction wave was obtained, except for a negligible residue of the phytol wave, for the lithium and tetramethylammonium chlorophyllins they had prepared. No polarographic waves have been reported for the commercial chlorophyllins.

The instrument used in the present investigation was the Sargent Model XXI Visible Recording Polarograph. All potentials were measured directly against the saturated calomel electrode. The dropping mercury capillary had the following characteristics: at a pressure of 26 cm of mercury, the drop time (t) on open circuit in 0.1 M KCl at 20° C was 2.86 sec, the weight of mercury dropping/sec (m) was 3.511 mg, and $m^{2/3}t^{1/6}$ was 2.752.

The electrolysis cell of Lingane and Laitinen was used (*3*). Dissolved oxygen was removed from all solutions by passing oxygen-free nitrogen through the sample for 15 min. A constant temperature water bath was used to maintain the temperature at 20° ±1° C. All samples were examined over a potential range of 0.0 to −2.0 v.

Of the 14 commercial chlorophyllins used in this in-