

formed ribonucleic acid was thus found in liver, kidney, adrenal cortex, and many epithelia. Large amounts were present in tissues where the neoformation of desoxyribonucleic acid occurred, especially in the crypts of the intestine. In contrast, several organs known to be fairly rich in ribonucleic acid, such as pancreas, salivary glands, and thyroid, did not show a significant amount of newly-formed ribonucleic acid.

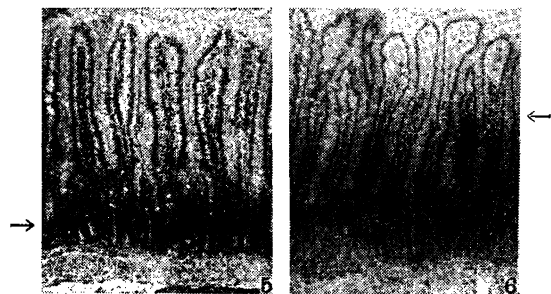


FIG. 3. (5) Duodenum of a rat sacrificed 2 hrs after P^{32} injection. The autographic reaction is located over the cells of the crypts of Lieberkühn, as indicated by the arrow. (6) Duodenum of a rat sacrificed 24 hrs after P^{32} injection. An intense autographic reaction is located over the nuclei in the cells of the villi epithelium; the upper limit of the reaction is indicated by the arrow. A less intense reaction is present in the crypts. ($\times 50$).

Conclusion. Radiophosphorus entering into desoxyribonucleic acid at the time of mitosis may be localized by the "coated autograph" method in tissue sections treated with ribonuclease. The newly-formed desoxyribonucleic acid thus detected is found in the tissues where cell divisions are numerous, e.g. lymphatic and myelogenous tissues, ovarian follicles, intestinal epithelium, etc.

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Protoanemonin as a Mitotic Inhibitor¹

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Experiments are in progress in this laboratory to determine the effect on root tip mitosis of several drugs and other chemicals. Protoanemonin, $\text{CH}_2\text{CH}(\text{CO}\cdot\text{O}\cdot\text{C}\cdot\text{CH}_2)$, has been found to exert striking effects not only on the nuclei of the meristematic cells but also on the mitochondria

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and on the structure of the cytoplasm. It has previously been found to be effective as an antibacterial agent (2).

Seedlings of *Zea Mays* were grown in beakers lined with moistened filter paper, according to the method of Albaum (3), until the roots had reached a length of about 1 cm, and were then transferred to similar beakers in which the filter paper liner was moistened with a solution of protoanemonin. Roots were treated in this way with various concentrations of the drug (10^{-3} M to 10^{-5} M) for various lengths of time (2 hrs to 24 hrs) and then fixed in each of three fixatives: Navashin's for nuclear details, a modified form of Erliki's (5) for mitochondria, and a mixture of chromic sulfate, formaldehyde, and copper hydroxide (4) for cytoplasmic structure. The root tips were dehydrated by the ethyl-normal butyl alcohol schedule, imbedded in paraffin, sectioned at 8 μ , and stained in iron alum-hematoxylin.

The most striking effect of protoanemonin is the disappearance of mitochondria. The Erliki fixation shows them well in untreated root tips, but not in root tips treated for 24 hrs at 10^{-5} M or higher concentrations or for 2 hrs or longer at 10^{-3} M. Cytoplasmic structure is badly disrupted by treatment at the stronger concentrations and for longer periods, as shown by the chromic sulfate-formaldehyde fixation. In the untreated meristem the image after this fixation is that of an even-textured cytoplasm interrupted by many small, sharply outlined vacuoles. After the longer and stronger treatments the cytoplasm is reduced to irregular strands and darkly staining granules. After shorter treatments and at lower concentrations the only effect is some coalescence of the vacuoles.

The nuclear effect of protoanemonin is strikingly different from that of colchicine. Treatment with the latter drug leads to an abnormally high frequency of metaphases. Treatment with protoanemonin at 2.15×10^{-4} M or higher concentrations for 24 hrs or at 10^{-3} M for 4 hrs or longer reduces the frequency of recognizable mitotic stages to a statistically significant degree. After the longer and stronger treatments, all the nuclei of the root tip are in a condition which resembles interphase or prophase. A small proportion of the nuclei superficially resemble late prophase.

In these nuclei the chromosomes are abnormally contracted, as are colchicine-treated chromosomes, but there is no evidence of chromatid separation or of polyploidy, which are characteristic of colchicine. It appears that protoanemonin exerts its inhibiting effect on mitosis at a different stage in the mitotic cycle than does colchicine.

A more detailed report of these results will be published elsewhere (1).

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