

such that one end of the chain of cells was firmly attached, while the other end was free to move about in a liquid menstruum. The spiral twisting was so rapid that it could be directly observed. It seems that the success in producing twisting filaments under these conditions strengthens the assumption that they form when under stress produced by rotational growth.

Spores of *B. mycoides* were heavily seeded into tubes of liquid 2 per cent. nutrient agar, and thin smears were made from the seeded agar on sterile glass slides resting within petri plates. After solidification of the film of agar, nutrient broth was poured into the petri plate to a depth just sufficient to completely cover the slide and smear. After 24 hours' incubation, the spores had germinated and the tendrils of cells had grown out into the nutrient broth. Spirally twisted loops were present in great numbers. This method of producing subjects for study has the disadvantage of rendering impossible examination with the oil immersion objective, since the slides must be examined while submerged.

A second procedure gave specimens more suitable for close examination. Plates of 2 per cent. nutrient agar were spot inoculated and a colony of two to three centimeters in diameter was allowed to develop. A small rectangular section of the agar, taken near the edge of the growing colony, was transferred to a glass



FIG. 1

slide and a cover slip was pressed tightly down upon the growing cells. The cover glass was sealed to the slide by generous amounts of vaseline, and nutrient broth was injected under the cover slip into the space between the agar block and the vaseline seal. After one to two hours the chains of cells had grown out into the broth and at the intersection of the agar with the broth, spiral twists could be observed forming. The subsequent observations herein reported were made on subjects prepared in this manner.

It was not possible to observe rotational growth by direct microscopic examination of the filament of growing cells. Neither has it been possible to facilitate direct observations of rotational growth by the attachment of particles of silica, charcoal or kaolin to the bacterial cells. It has been possible to determine the direction of rotation of the loop within spirally twisting filaments. Loop rotation is a direct function of the filament rotation. Account was taken of the inverted image given by the compound microscope.

Hundreds of loops of both left (counter clock-wise)

and right (clock-wise) spiral strains of *B. mycoides* have been studied and, without exception, loops of left spiral strains rotate from left to right (loop away from the observer, ends of the filament toward the observer), and of right spiral strains from right to left.

The production of new cells in filaments of *B. mycoides* occurs most profusely at the end of the filament. Hence, it is probable that any rotational growth would be most pronounced in the young developing cells which are farthest removed from the attached end of the filament. If this is true, a loop rotation (loop away from observer, ends of the filament toward observer) from left to right would indicate a filament rotation from right to left, when the growing tip is away from the observer and the attached end of the filament is toward the observer. Likewise, a loop rotation from right to left would indicate a filament rotation from left to right under the same condition.

Since left spiral strains invariably give loop rotation (loop away from observer) from left to right, it seems probable that the filament rotation (growing tip away from observer) from right to left is responsible for the left-hand (counter clock-wise) spirals produced by these strains on solid culture media. With the right spiral strains, a filament rotation from left to right is probably responsible for the production of right spiral colonies.

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CARCINOGENICS AND GROWTH STIMULATION¹

ALL stimulants to growth do not result in cancer, but it seems apparent, as Loeb *et al.*² have said, "all causes of cancer directly or indirectly stimulate growth." Goldstein³ has suggested acceleration of bacterial reproduction as a microbiological test for carcinogenic hydrocarbons. Hammett and Reimann⁴ have shown that methyl cholanthrene, a carcinogenic, enhances the production of new growth from anlagen in *Obelia geniculata*. The same authors⁵ found proliferation activity of *Obelia* to be stimulated by the carcinogenic 1:2:5:6 dibenzanthracene.

Our studies, employing planaria (*Euplanaria dorotocephala*) show 1:2:5:6 dibenzanthracene to stimulate both regeneration of cut segments and reproduction of whole animals. In a test period of over a

¹ Published with the permission of the Medical Director of the Veterans' Administration who assumes no responsibility for the opinions expressed or the conclusions drawn by the authors.

² L. Loeb, E. L. Burns, V. Sontzeff and M. Moskop, *Am. Jour. Cancer*, 30: 47-53, 1937.

³ S. Goldstein, *SCIENCE*, 86: 176-177, 1937.

⁴ F. S. Hammett and S. P. Reimann, *Am. Jour. Cancer*, 25: 807-808, 1935.

⁵ S. P. Reimann and F. S. Hammett, *Am. Jour. Cancer*, 23: 343-349, 1935.

month there were approximately 45 per cent. more animals in the test than in the control jars. With triphenyl benzene, whose carcinogenic activity has been recently questioned, the whole animals were similarly stimulated while the segments failed to react.

With glutathione, a known tissue growth stimulant, the results were similar to those obtained with dibenzanthracene. Derivatives of glutathione, as glutamic acid, glycine and cysteine, produced no evident stimulation of growth in the segments or increase in the number of planaria.

Allantoin and preserved larval extract did not stimulate growth or reproduction of the cut or uncut animals.

Complete details of the technique, histology of the treated specimens and further results will be presented when the study is completed.

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THE EFFECT OF FAST NEUTRONS ON DRY SEEDS

THE experiments discussed in this paper had a two-fold purpose: (a) to determine whether dry seeds left on the outside of the cyclotron "tank" would receive enough stray neutrons to produce a cumulative harmful effect preventing their subsequent germination; and (b) to determine whether changes in external morphology, similar to those produced by x-rays,^{1, 2} radium and radium salts, would occur in plants grown from the neutron-bombarded seeds.

Plants with very small seeds were chosen for these experiments in order that a large number might be used at one time in the limited space available for bombardment by the cyclotron.

Seeds from each of the selected species and varieties were put into small (2.3 × 0.8 cm) gelatin, medical capsules which proved to be most satisfactory for handling the tiny seeds. More than 500 seeds of the *Oenotheras*, for example, were placed in a single capsule without crowding.

The capsules were in turn placed in a small (8 × 8 × 6 cm) lead box whose 1 cm. thick walls were lined inside with a 2 mm layer of paraffin. The purpose of the lead was to filter out all emanations from the cyclotron except the neutrons, and that of the paraffin to increase the effect of the neutron bombardment within the seeds. A lead cover 1 cm thick was

anchored to the box after the capsules bearing the seeds had been placed within.

The whole was then laid on a small metal shelf attached to the outside of the cyclotron "tank" so as to be close to the bombarding chamber, but at a distance of about 60 cm from the target. In this way the seeds could absorb only stray emanations from the cyclotron, whenever it was in operation, during the three months in which these experiments were conducted.

Seeds of the various kinds were removed from the capsules from time to time and put on wet filter paper in Petri dishes to obtain germination counts. Then the germinating seeds were planted in soil in order to obtain seedlings and mature plants upon which to observe possible morphological changes due to the neutron bombardment.

All exposures were made on the University of Michigan cyclotron through the cooperation of Professor J. M. Cork and Dr. R. L. Thornton, of the physics department.

Normal germination for all plants used in these experiments is above 90 per cent.

Table 1 shows three things: (a) that as exposure time increases the percentage of germination in some plants decreases, whereas (b) in others there is little or no appreciable effect, and (c) that there is a wide range of susceptibility to neutrons as has already been found for a large number of x-rayed plants by Johnson.

TABLE 1

Species studied	Weeks subjected to emanations	Percentage germination
<i>Oenothera franciscana</i>	1	95.0
<i>Oenothera franciscana</i>	2	82.5
<i>Oenothera franciscana</i>	9	15.4
<i>Oenothera blanda</i>	2	94.8
<i>Oenothera blanda</i>	9	51.9
<i>Echinocereus papillosus</i>	1	16.8
<i>Rhipsalis rhombica</i>	2	29.2
<i>Neomammillaria multiceps</i> ..	2	99.5
<i>Antirrhinum</i> sp. (1)	6	5.0
<i>Antirrhinum</i> sp. (2)	6	75.0
<i>Antirrhinum</i> sp. (3)	6	23.0
<i>Myosotis</i> sp.	6	18.7

The morphological variations in the plants grown from treated dry seeds were numerous, and it is noteworthy that often they are found to be similar in more than one species. This same phenomenon occurs in x-rayed plants. A brief summary of the variations and their relative frequencies are discussed in the following paragraphs.

A condition which was observed to be most common was the decrease in size of plants grown from rayed seeds as compared with normal plants. They were not only shorter, but the stems and leaves were smaller and weaker.

In *Antirrhinum* the cotyledons were invariably found to be covered with numerous white dots or

¹ J. H. Lawrence and E. O. Lawrence, *Proc. Nat. Acad. Sci.*, 22(2): 124-133, 1936.

² R. E. Zirkle and P. C. Aebersold, *Proc. Nat. Acad. Sci.*, 22(2): 134-138, 1936.