

PANTOTHENIC ACID AS A NUTRILITE FOR GREEN PLANTS

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Introduction

Ever since the work of BOTTOMLEY on the stimulating effect of extracts of bacterized peat on the growth of green plants, there has been some interest in the possibility that green plants may require an outside source of specific organic material. The preponderance of opinion (1, 2, 6) seems to favor the idea that, while stimulating effects may exist, the green plant is able to grow without any such outside stimulus and hence is independent in accordance with the classical view.

It has been suggested (6) that the term "auximone," coined by BOTTOMLEY, should be dropped from the literature because the substances are not indispensable and the experimental data from which their existence was inferred were based upon an unfavorable salt balance. It is claimed that auximones, as conceived by BOTTOMLEY, do not really exist. In this connection we again call attention to the advantage in the term *nutrilite* (3), which carries no presuppositions except that the vitamin-like substance functions in nutrition.

Up to the present, so far as we are aware, no substantial clue which will stand scrutiny has been obtained as to the chemical nature of the substances which are presumed to exist and to stimulate the growth of green plants.

Experimental work

The first suggestion that pantothenic acid (4) might be involved in this problem came from a study of the pantothenic acid content of dormant and sprouting potatoes. It was found that while the pantothenic acid is rather uniformly distributed throughout the tissue of dormant potatoes, during sprouting there is a distinct mobilization of the acid toward the sprouting end. This suggested that it might have a growth stimulating function as is the case with yeast.

Preliminary work, conducted about the same time by Dr. W. L. POWERS of the Department of Soils, showed that one of our relatively crude concentrates had a stimulating effect on alfalfa seedlings. The details of these and other tests will be published elsewhere.

Through the initiative of the junior writer, experiments were conducted with an aquatic plant which, so far as we know, has not been used for physiological study. Young plants of *Ricciocarpus natans* were collected from an aquarium and later from a marshy lake, and cultured on

Detmer's solution. The first effects were observed visually. When the young plants grow the two lobes separate as a new lobe grows between them. It was noted that this evidence of growth was much more apparent in cultures to which from 10 to 10,000 parts per billion of a crude pantothenic acid concentrate (calcium salt potency of 70) was added. The best result was obtained with 1 part per million and the difference became definitely noticeable on the third day. In another experiment there was visual evidence (recorded after 12 days) of improved growth when as little as 1 part per billion of a refined preparation (potency 5000) was added. When 10 and 100 parts per billion were added the growths were better than the control but not so good as when only 1 part per billion was present.

In a third experiment, two carefully selected plants of uniform size were grown in each of four dishes. Those to which additions of the refined preparation just described were made showed some increased growth in 5 days; and after 9 days the plants were blotted uniformly between filter papers and weighed in a closed vessel on a micro balance, with the results indicated in table I.

TABLE I
EFFECT OF PANTOTHENIC ACID ON GROWTH OF *RICCOCARPUS NATANS*

	WEIGHT OF PLANTS
Blank	0.002545
1 p. p. billion	0.003210
10 p. p. billion	0.004350
100 p. p. billion	0.003840

It is evident that 100 parts per billion is an overdose for this particular preparation. It should be noted also that while the medium used may not be ideal for the liverwort, the amount of material added is extremely minute, and its effect must therefore be specific.

Further experiments indicated that the liverwort used in these experiments is not a very favorable plant for continuous laboratory culture, because the young plants are available only after sporulation and this event cannot be controlled in the laboratory. The physiological condition of the plants is an important factor. The plants which showed the most striking responses were those obtained from the aquarium. Those taken from the natural habitat were less responsive, and after keeping for a few weeks the plants in one case were apparently unaffected by the more refined preparation, except that 100 parts per billion actually inhibited growth.

Within a few days after these experiments had been completed, a communication was received from Professor W. J. ROBBINS indicating that in preliminary experiments he had obtained positive growth stimulating results with a pantothenic acid preparation which we had furnished. His study utilizing isolated root tips from corn will doubtless be published shortly.

Discussion

These studies with four different plants (including the potato), even though they are not exhaustive, would seem to indicate that pantothenic acid plays a rôle in the growth of green plants generally. This idea is strengthened by the evidence previously presented (4), that pantothenic acid is a constituent of all types of living matter.

To the plant physiologist mainly will fall the problem, which will probably prove an intricate one, of finding precisely how pantothenic acid functions. The writers cannot answer this question and available facilities will probably severely limit our further work along this line. We wish, however, to mention a few observations which are an outgrowth of our limited experience with green plants and our more extended acquaintance with the chemical factors influencing yeast growth.

ORIGIN OF PANTOTHENIC ACID.—The observation has been recorded elsewhere (4) that *Aspergillus niger* produces pantothenic acid as it grows upon a synthetic medium. We have also observed that several bacteria and other molds which can grow upon synthetic media also produce it. It is not surprising, therefore, that the substance is present in soils and especially abundant in those soils which are rich in organic matter. Our experience would lead us to suspect that it is produced neither by higher plants nor by animals. We have conducted careful experiments to prove whether or not it is produced as yeast grows (extremely slowly) in a synthetic medium or one with known low pantothenic acid content. The results indicate that it is not produced. Yeast grown on a medium low in pantothenic acid content contains very little pantothenic acid, as low as one-fiftieth as much as "normal" yeast and even less than that introduced originally into the medium. The medium at the end of the growth period was practically devoid of pantothenic acid. It seems that under these conditions the pantothenic acid present in the mother cells is distributed to the daughter cells, and that growth reaches practically a standstill in the course of a few days, owing to its very low concentration.

IS PANTOTHENIC ACID INDISPENSABLE OR MERELY STIMULATIVE?—In the light of the experiments just cited, one can draw his own conclusions as to the indispensability of pantothenic acid for yeast growth. If the conclusion is that the effect is that of a stimulant, the interest in the stimulating

substance should not be diminished, because no one can question but that under anything approaching natural conditions yeast is always stimulated by pantothenic acid, whether or not it is capable of growing very slowly without that stimulation.

To us the possibility that pantothenic acid is stimulative rather than indispensable to green plants should not lessen interest in it. Under natural conditions plants are doubtless stimulated by the substance because it is found in soils generally and the organisms of the soil are capable of producing more when the available supply runs low, of course, provided that the soil is suitable for the growth of these organisms. Just how long a plant can grow and eke out an existence without a further supply of organic matter beyond that supplied by the seed is not very important, if under ordinary circumstances it obtains specific organic matter from outside sources and grows normally because of this exogenous supply.

ARE OTHER NUTRILITES INVOLVED IN GREEN PLANT STIMULATION BESIDES PANTOTHENIC ACID?—Our experimental evidence with regard to this point is scanty, yet we have observed in work with the liverworts that an extract of potato seemed to have a stimulating effect well beyond that which could be accounted for by its pantothenic acid content alone. Since the physiological condition of the test organism could not be standardized, the results were not of a quantitative nature, and a definite conclusion would therefore be dangerous.

From our work with yeasts we should not be surprised if factors other than pantothenic acid were involved. Not all of the various strains of *Saccharomyces cerevisiae* are stimulated markedly by pantothenic acid alone. Other complementary factors may be necessary to produce even approximately normal growth. In the case of two yeasts, crystalline vitamin B₁ fills this need. What the nature of the complementary substance is in the case of green plants we do not know, but we expect to be able to answer the question as to their existence and nature by further experimentation. From analogy with yeast growth experience, we should expect that plant growth stimulation could be induced much more markedly than has been possible if all the unknown factors were supplied to the growing plant.

EFFECT OF TOXIC FACTORS.—Not only with yeast but in our limited experiments with green plants we have observed the influence of inhibiting substances. In yeast growth experiments we are constantly confronted with such effects. We have natural extracts which are devoid of any ability to stimulate yeast growth because they contain toxic material which is specific for a given yeast. By fractional electrolysis (5) we are able to prepare very active stimulating material from the same extract. Our experiments indicate that such results will carry over to experiments with green plants and will significantly modify results. There is also the possibility

that an unfavorable balance between various complementary growth-stimulating substances may result in growth inhibition. This appears to take place with yeast in some cases.

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Summary

1. Pantothenic acid has been found in preliminary experiments to have a growth stimulating effect on green plants.

2. The questions of whether it is stimulative or indispensable, where it originates in nature, and the probability of the existence of other plant nutrilites, are discussed.

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