

sensitive chromophoric group having a strong red fluorescence in ultra-violet light at 365 m μ .

The following procedure distinguishes pure cultures of *D. desulphuricans* and its *aestuarii* variant from *D. orientis*: a fully grown culture in 10 ml. of medium C of Butlin, Adams and Thomas⁶ was centrifuged and the cells resuspended in 1 ml. water (final cell density 1.5–3 mgm. dry wt./ml.). One drop of 20 per cent sodium hydroxide was added and the suspension inspected at once in light at 365 m μ . Twenty strains of the *desulphuricans* group were tested and showed a brilliant red fluorescence; three strains of *D. orientis* did not, nor did six strains of *Cl. nigrificans*. Large amounts of ferric sulphide in the culture interfere, but may be removed by coarse filtration.

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Dec. 12.

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Separation of Specific and Non-Specific Effects of Chemicals on Plants

IN working with the effects of chemicals on plants, it is often difficult to determine which of the effects obtained is specific to the chemical applied. For example, 2,4-dichlorophenoxyacetic acid (2,4-D) has been shown to cause numerous effects on plants. A recent review by Woodford *et al.*¹ discusses this subject in detail. It has been shown that 2,4-dichlorophenoxyacetic acid causes varied tissue changes, affects phosphorus metabolism, increases respiration, decreases photosynthesis and, in general, affects a great many functions of the plant.

Although it is true that these effects do occur on plants, it is by no means certain which, if any, of these reactions is of primary importance, and which are secondary. The living plant represents a dynamic, balanced system, and any change which affects one function of the plant will necessarily affect a great many others. Some of these changes will be associated with any type of killing mechanism; others will be specific. The data presented in many of the reports in the literature are not sufficient to establish this point. Frequently, there is no mention of whether there has been gross damage to the experimental plants, and the reader may suspect that the changes reported are simply those characteristic of damaged or dying plants.

One way of tackling the problem of which changes are of a non-specific nature is to use a control which has been allowed to die without outside chemical influence. We have attempted to do this in our experiment by digging up some of the plants, washing off the roots, and letting them lie on the greenhouse bench for the duration of the experiment. A similar technique was used by Loustalot *et al.*² in field experiments.

Bean plants (*Phaseolus vulgaris* L.) were grown in the greenhouse in a mixture of sand and peat soil

Table 1. CHANGES OF PROTEIN IN TREATED BEAN ROOTS (MGM. ALCOHOL INSOLUBLE NITROGEN PER PLANT)

2,4-D per acre (lb.)	Days after treatment		
	1	4	18
0 (untreated)	2.64	2.54	2.80
1	2.90	2.58	2.51
2	2.71	—	1.60
4	2.51	2.30	1.30
0 (uprooted)	2.58	1.92	1.91

Table 2. CHANGES IN FREE AMINO NITROGEN IN TREATED BEAN ROOTS (MGM. PER PLANT)

2,4-D per acre (lb.)	Days after treatment		
	1	4	18
0 (untreated)	0.109	0.073	0.076
1	0.111	0.104	0.057
2	0.095	—	0.039
4	0.102	0.077	0.029
0 (uprooted)	0.114	0.090	0.006

until they were three weeks old. The beans were then selected for uniformity and treated. Foliage sprays of 1, 2, and 4 lb. per acre acid-equivalent of 2,4-dichlorophenoxyacetic acid (amine salt in water solution) were applied. There were four replicates of each treatment, and the plots were completely randomized in the greenhouse. The roots were harvested 1, 4, 11 and 18 days after treatment. Epinastic responses in the young leaves were visible on the first day after all treatments. The plants were dead in the 4 lb. treatment in 18 days. At this time, plants treated with 2 lb./acre were very stunted and malformed and those treated with 1 lb./acre apparently were recovering. Total protein content (alcohol-insoluble nitrogen) free amino-acid nitrogen, and individual amino-acids were determined as reported elsewhere^{3,4}. A brief summary of the results is given in Tables 1 and 2. The trends for individual amino-acids were similar to those for total free amino-nitrogen. A fuller account of these results is in preparation.

Although pronounced changes occurred in the proteins of the plants following 2,4-dichlorophenoxyacetic acid treatment, and these changes appeared superficially to be correlated with the amount of 2,4-dichlorophenoxyacetic acid applied, it is obvious that equally drastic changes occurred in the plants that were uprooted and not treated with 2,4-dichlorophenoxyacetic acid. It appears, therefore, that these changes in the proteins and the amino-acids occurred as a result of damage or death, and not directly as a result of 2,4-dichlorophenoxyacetic acid treatment *per se*. Our results suggest, therefore, that the effect of 2,4-dichlorophenoxyacetic acid on protein metabolism is of secondary importance. They also emphasize the value of this type of control. Without this control, it would have been possible to draw exactly opposite and erroneous conclusions.

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