Effects of 2,4-Dichloro-5-iodophenoxyacetic Acid and Its Derivatives as Plant Growth Regulators

P. J. Linder, J. W. Mitchell, and J. W. Wood

Bureau of Plant Industry, Soils and Agricultural Engineering, and
Bureau of Agricultural and Industrial Chemistry,
U. S. Department of Agriculture, Beltsville, Maryland

Radioactive iodine has been useful in studying the absorption and translocation of growth-regulating substances by plants (*2, 4*). Work in this field has thus far been limited to the use of radioactive 2-iodo^{131}-3-nitrobenzoic acid. A series of growth-regulating substances of the phenoxy type closely related to 2,4-dichlorophenoxyacetic acid (2,4-D)[1] have now been synthesized and some of them tagged[2] with I^{131} (*3*). These new compounds consist of 2,4-dichloro-5-iodophenoxyacetic acid (2,4-D-5-I) and 14 derivatives. They offer promise of extending the usefulness of radioactive isotopes in learning how growth regulators are absorbed and translocated, and in studying the physiological mechanism involved when plants respond to these chemicals.

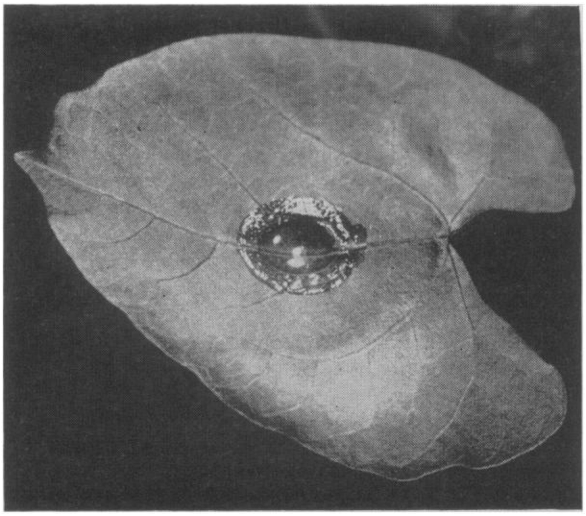

FIG. 1. Method of treating a known area of leaf with a measured amount of liquid containing a plant growth regulator, by enclosing the area with a lanolin ring.

Plant responses to stable 2,4-dichloro-5-iodophenoxyacetic acid and its derivatives are described here, and their effects are compared with those resulting from the use of an equimolar amount of 2,4-D. The use of 2,4-D-5-I^{131} and its derivatives as radioactive tracers will be described in a later paper.

2,4-Dichloro-5-iodophenoxyacetic acid was applied separately to the leaves, stems, and roots of kidney bean plants. As controls, comparable plants were similarly treated with an equimolar amount of 2,4-D. Both 2,4-D-5-I and 2,4-D brought about stem curvatures when applied to three plant parts. It was evident that 2,4-D-5-I was absorbed at about the same rate as was the 2,4-D, whether applied to the leaves, stems, or roots.

[1] The abbreviation 2,4-D is used to denote only the parent acid.

[2] Radioactive 2,4-D-5-I^{131} was originally synthesized in the Bureau of Agricultural and Industrial Chemistry; later it was obtained on contract from Texas Research Foundation.

Translocation of 2,4-D from leaves to stems of bean plants is associated, either directly or indirectly, with the translocation of carbohydrates from the leaves. Tests were made to determine whether the translocation of 2,4-D-5-I from leaves was also related to the movement of photosynthate. In this experiment, a ring of lanolin was stamped on the upper surface of the primary leaves of bean seedlings. During this stage of development, carbohydrate reserves in leaves of bean seedlings are relatively low (*1*). The ring was placed along the midrib and approximately 1 cm from the petiole attachment. Approximately 0.5 cm² of leaf surface was thus enclosed. One-hundredth of a milliliter of 95% ethyl alcohol containing 2% of Tween 20 as a carrier and 10 μg of 2,4-D-5-I was applied evenly to the enclosed leaf area with the aid of a 0.1-ml pipette (Fig. 1). Other comparable leaves were treated in a similar manner with an alcohol-Tween mixture containing an equimolar amount of 2,4-D. In this way, the growth regulators were confined to a known position on each leaf and covered a known area. Following treatment, the alcohol evaporated rapidly, leaving a thin layer of Tween 20 in which the growth regulator was dissolved. Control leaves treated in a similar manner with the alcohol-Tween mixture alone showed no injurious effects.

TABLE 1

COMPARISON OF THE INHIBITING EFFECT OF 2,4-D-5-I WITH THAT OF 2,4-D WHEN APPLIED TO LEAVES

Treatment	Percent reduction in terminal bud growth
2,4-Dichlorophenoxyacetic acid	
Untreated control	
0.5 μg per 0.01 ml	38
1.0 " " " "	66
2.0 " " " "	80
3.0 " " " "	91
2,4-Dichloro-5-iodophenoxyacetic acid	
2.0 μg per 0.01 ml	11
5.0 " " " "	18
10.0 " " " "	33
20.0 " " " "	57
40.0 " " " "	77

Following treatment, one half of the plants treated with 2,4-D-5-I and half of those treated with 2,4-D were placed in darkness, and the remaining plants of each group were grown under artificial illumination. Neither 2,4-D-5-I nor the 2,4-D was translocated from the leaves of the plants kept in darkness. Both compounds were translocated readily from leaves of illuminated plants as was indicated by the development of marked stem curvature. These results indicate that the translocation of both 2,4-D-5-I and 2,4-D from leaves took place under conditions favorable for the production and translocation of photosynthate.

2,4-D-5-I and 2,4-D were compared for growth-inhibiting effect. Various concentrations of each acid were applied within lanolin rings on primary leaves of bean plants in the manner described above. During the following 6-day period, 2,4-D was more effective in inhibiting bud growth than was 2,4-D-5-I (Table 1).

The herbicidal properties of the 2,4-D-5-I were compared with those of 2,4-D. 2,4-D-5-I was dissolved to make a concentration of 1000 ppm in water containing 0.5% of Tween 20. An equal concentration of 2,4-D was prepared in the same manner. The mixtures were sprayed separately on two groups of bean seedlings. Within 6 days, all plants treated with the 2,4-D were dead and there was no apparent increase in the size of the plants. In contrast, plants sprayed with 2,4-D-5-I survived for 11 days following treatment. During this period there was extensive cell proliferation, mainly in sensitive tissues of the stems. Terminal bud growth was, however, completely checked by 2,4-D-5-I, and the plants finally died. Thus, 2,4-D-5-I was somewhat less toxic to bean plants than was 2,4-D under the test conditions, since the latter compound killed the plants more quickly and caused less cell proliferation.

of both 2,4-D and 2,4-D-5-I dissociate within the plant to give two effective phenoxy components for each molecule.

Dissociation of the cupric salt of 2,4-D-5-I was not indicated by results of the present tests. It should be stressed that the activity of a compound is determined by such factors as rate of absorption and translocation as well as by dissociation.

In other experiments three additional derivatives of 2,4-D-5-I, namely the triethanoleamine salt, the *n*-butyl ester, and the isopropyl ester, were tested for growth-regulating activity. These had growth-regulating properties similar to the other iodo compounds tested.

In summary, 2,4-dichloro-5-iodophenoxyacetic acid was found to have plant growth-regulating properties. Fourteen derivatives that included salts and esters of this acid also brought about plant growth-regulating effects. The ethyl ester was the most effective of these iodo compounds under the test conditions. 2,4-Dichlorophenoxyacetic acid was more active, both from the standpoint of growth inhibition and with respect to herbicidal properties, than

TABLE 2

GROWTH-INHIBITING EFFECTS OF 10 MICROGRAMS OF 2,4-D COMPARED WITH THAT OF EQUIMOLAR CONCENTRATION OF 2,4-D-5-I AND DERIVATIVES

Compound	Mg used per 10 ml for equimolar concentration	Average fresh wt terminal growth (g)	Percent inhibition of terminal growth
Untreated control	0.00	1.93	00
2,4-Dichlorophenoxyacetic acid	5.00	0.16*	92
Ethyl 2,4-dichloro-5-iodophenoxyacetate	8.49	0.51*	74
Calcium 2,4-dichloro-5-iodophenoxyacetate	16.55	0.54*	72
2,4-Dichloro-5-iodophenoxyacetamide	7.83	0.65*	66
Morpholine 2,4-dichloro-5-iodophenoxyacetate	9.82	0.68*	65
Methyl 2,4-dichloro-5-iodophenoxyacetate	8.17	0.73*	62
Ethylammonium 2,4-dichloro-5-iodophenoxyacetate	8.85	0.79*	59
Ammonium 2,4-dichloro-5-iodophenoxyacetate	8.24	0.81*	58
Potassium 2,4-dichloro-5-iodophenoxyacetate	8.70	0.83*	57
2,4-Dichloro-5-iodophenoxyacetic acid	7.85	0.85*	56
Diethylammonium 2,4-dichloro-5-iodophenoxyacetate	9.50	0.85*	56
Sodium 2,4-dichloro-5-iodophenoxyacetate	8.35	0.91*	53
Cupric 2,4-dichloro-5-iodophenoxyacetate	17.09	1.00*	48

* Significantly less than the control at 1% level, 0.65 g being required for significance at this level.

The growth-inhibiting effect of 2,4-D-5-I and 11 of its derivatives was compared with that of 2,4-D at equal molar concentrations (Table 2), the treatments being applied to bean seedlings as previously described.

The relatively high activity of the calcium salt is thought to be due, in part at least, to dissociation of the compound within the plant. An experiment with 2,4-dichlorophenoxyacetic acid was carried out to test this theory. By using methods similar to those described above, it was determined that the activity of 2,4-D in inhibiting terminal growth of bean plants was approximately equal to that of the calcium salt of 2,4-D when compared on an equivalent weight basis. Applied at an equimolar concentration the activity of the calcium salt was roughly twice that of 2,4-D, which indicates that the calcium salts

were any of the iodo compounds tested. Evidence was obtained indicating that the calcium salt of 2,4-dichlorophenoxyacetic acid and 2,4-dichloro-5-iodophenoxyacetic acid dissociate within the plant. Although 2,4-dichlorophenoxyacetic acid differs from the iodo compounds with respect to degree of herbicidal activity, the method of absorption and translocation of these compounds appears to be similar.

References

1. MITCHELL, J. W. and BROWN, J. W. *Bot. Gaz.*, 1946, **107**, 393.
2. MITCHELL, J. W. *et al.* *Science*, 1947, **106**, 395.
3. WOLFE, W. C. *et al.* *J. org. Chem.*, 1949, **14**, 900.
4. WOOD, J. W., MITCHELL, J. W., and IRVING, G. W., JR. *Science* 1947, **105**, 337.