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Photoaffinity-labeled Auxins

SYNTHESIS AND BIOLOGICAL ACTIVITY[1]

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ABSTRACT

Two light-sensitive analogs of 2,4-dichlorophenoxyacetic acid, namely 4-azido-2-chlorophenoxyacetic acid and 3-azido-5-chlorophenoxyacetic acid, have been synthesized for use as auxin photoaffinity labels. The preparation and biological activity of the compounds are described. Both contain the photolabile azido group; the 2,4-substituted compound shows auxin activity and the 3,5-substituted compound does not. These photoaffinity analogs of 2,4-dichlorophenoxyacetic acid may be useful in the identification of the auxin receptor molecules in plant cells and eventually of the receptor sites within these molecules.

Among plant growth regulators, the auxins are generally distinguished by their promotion of cell elongation (25, 29). They are typified by the naturally occurring IAA, but the synthetic compounds 2,4-D and NAA[3] are often used to produce similar responses as IAA does in plants (31). The problem of the action of auxin in cell elongation can be approached from the point of view of establishing the binding site or sites of auxin and determining the specificity of such binding. The reported isolation of a protein from the nuclei of tobacco and soybeans which binds auxin and stimulates RNA synthesis (23) together with the report that an auxin-binding protein isolated from the endosperm of coconut, plus auxin, stimulated an RNA polymerase system *in vitro* (24) suggests an auxin acceptor located within the cell nucleus. Other work has implicated auxin action initially at the plasma membrane level. Hertel *et al.* (14) detected a specific binding of IAA and NAA, but not of 2,4-D, to plasma membranes obtained from maize homogenates, suggesting the possibility of different receptor sites for polar transport and for elongation (25). Hardin *et al.* (13) have proposed the presence of a regulatory factor for RNA polymerase associated with soybean plasma membrane and specifically released by the auxin 2,4-D but not by the isomeric nonauxin 3,5-dichlorophenoxyacetic acid. These advances, together with the specificity of response shown by the enantiomers of 2-(2,4-dichlorophenoxy)-propionic acid (2, 11, 30) and other auxins (1, 11) suggest that the receptor(s) may be protein and possibly membrane-bound.

One method that has recently been applied to the identification of biological receptor sites is the technique of photoaffinity labeling (18, 21). In this procedure, an analog of a biologically active substance is allowed to bind to the receptor site and is then activated photochemically to produce a chemically reactive species that immediately combines covalently with neighboring structures at the binding region. For example, the reactive species may be a carbene, which is generated from an α-keto diazo compound (26) or diazirine (28), or may be an aryl nitrene which is produced from an aryl azide (21). The use of the triplet of benzophenone has also been suggested as a photochemical probe of biological ligand-receptor interactions (12). Among numerous applications of photoaffinity labeling are the covalent attachment of NAD+ analogs to enzymes (5, 15), the identification of antibody binding sites (8, 10), the labeling of phosphofructokinase with a cAMP analog (9), and the use of photosensitive analogs for study of estrogen binding proteins of the rat uterus (19, 20).

These applications of the photoaffinity technique have inspired the synthesis and testing of analogs of synthetic auxins, corresponding to 2,4-D. We report here the synthesis and biological activity of a pair of analogs which are azidochlorophenoxyacetic acids. Both contain the photolabile azido group; one shows auxin activity and the other does not in the tested concentration range. The use of these photoaffinity analogs may help in the identification of the receptor molecules in plant cells and eventually of the receptor sites within these molecules.

The azido analog of 2,4-D, namely 4-azido-2-chlorophenoxyacetic acid **(I)**, was prepared according to the sequence outlined in Scheme I (II → III → IV → I). As an internal confirmation of the methodology, it was ascertained that treatment of the diazonium salt **IV** with freshly prepared cuprous chloride produced 2,4-dichlorophenoxyacetic acid, identical with an authentic sample.

As a control for the biological testing of compound **I** and for analogy with the 3,5-disubstituted phenoxyacetic acids which consistently show little or no auxin activity (30), 3-azido-5-chlorophenoxyacetic acid (**V**) was prepared as shown in Scheme II (VI → VII → VIII → IX → X → V). It was necessary that the final product be completely free of positional isomeric contaminants which might show activity in the sensitive bioassay pro-

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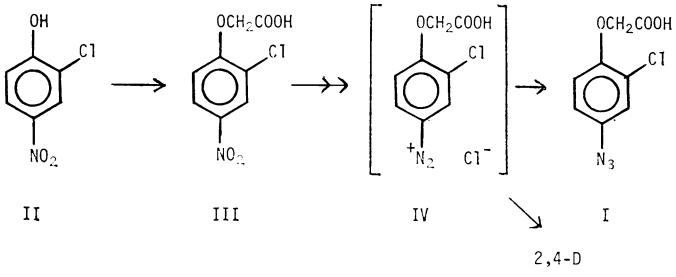
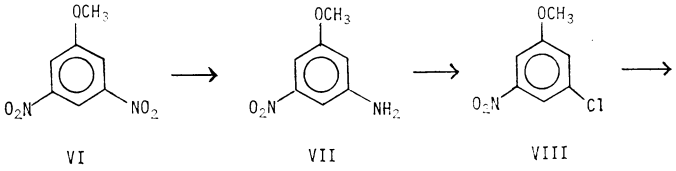
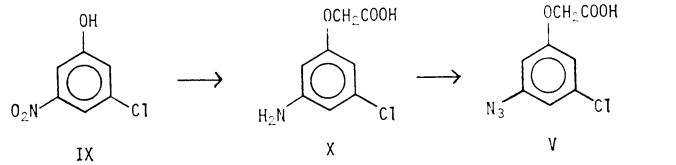
[3] Abbreviation: NAA: 1-naphthaleneacetic acid.



SCHEME I





SCHEME II

cedure capable of detecting auxin concentrations as low as 1 nM. This requirement precluded syntheses involving nitration or chlorination of a disubstituted benzenoid precursor. As a point of entry into the 1,3,5-trisubstituted benzenoid series, 3,5-dinitroanisole (**VI**) was used as the starting material. The synthesis was followed for isomer integrity at each stage by nuclear magnetic resonance.

4-Azido-2-chlorophenoxyacetic acid (**I**) and 3-azido-5-chlorophenoxyacetic acid (**V**) were bioassayed for auxin activity in the pea stem segment (27) and oat coleoptile segment (4) tests. The activities were compared against IAA, NAA, and 2,4-D. In addition, the azido compounds **I** and **V** were tested for possible competition with or sparing of 2,4-D in the same bioassay.

MATERIALS AND METHODS

General. All melting temperatures were determined with a Thomas-Hoover melting point apparatus and are corrected. Proton magnetic resonance spectra were determined on a Varian A-60A or HR-220 instrument. Chemical shift data are given as δ units from tetramethylsilane as internal standard. Infrared spectra were recorded on a Perkin-Elmer 521 grating spectrophotometer as KBr pellets. Ultraviolet spectra were recorded on a Cary Model 15 spectrophotometer, using the appropriate blank. Microanalyses were performed by Joseph Nemeth and staff, University of Illinois, and by Midwest Microlab, Inc., Indianapolis, Ind. Mass spectra were determined with a Varian-MAT CH-5 or 731 spectrometer.

2-Chloro-4-nitrophenoxyacetic Acid (III). This compound was made from sodium bromoacetate with 50% excess 2-chloro-4-nitrophenol (II) (Aldrich) and NaOH in refluxing aqueous solution, purified by acidifications, basifications, and extractions and recrystallized from aqueous ethanol, m.p. 179 to 180 C (179 to 181 C, see ref. 7).

$C_8H_6ClNO_5$
Calculated: C, 41.49; H, 2.61; N, 6.05
Found: C, 41.75; H, 2.55; N, 5.76

4-Azido-2-chlorophenoxyacetic Acid (I). A solution of 500 mg (2.15 mmoles) of 2-chloro-4-nitrophenoxyacetic acid (III) in 95% ethanol was reduced at 23 C with hydrogen (approximately 3 atm) in the presence of 50 mg of 10% Pt/C catalyst. The solvent was removed under reduced pressure, 30 ml of 0.1 N HCl were added to the residue, and the resulting suspension was filtered. The filtrate was chilled in an ice bath, and a solution of 160 mg (2.3 mmoles) of sodium nitrite in 10 ml of H_2O was added slowly. The cold solution was stirred for 30 min, filtered, and to the filtrate (IV) was added 150 mg (2.3 mmoles) of sodium azide in 10 ml of H_2O. When nitrogen evolution subsided, the suspension was extracted with ether. From this point on, the material had to be handled under red light in order to preserve the azido group. The combined ether extracts were dried over anhydrous sodium sulfate, filtered, and the filtrate was evaporated to dryness. The residue was recrystallized from aqueous ethanol to yield 300 mg (61%) of tan crystals, m.p. 128.5 to 129.5 C; UV $\lambda_{max}^{95\% \text{ ethyl alcohol}}$ 294 nm (ϵ 2,850), 256 (11,400); nuclear magnetic resonance ($(CD_3)_2SO$) δ 4.78 (s, 2H), 7.04 (m, 3H); infrared (KBr) ν_{max} 2110 cm^{-1} ($-N_3$), 1735 (C=O); mass spectrum m/e (relative intensity) = 227 (M^+, 5.7), 199 (34.6), 141 (100), 106 (27.1). The compound was stored in a foil-covered vial in the dark.

$C_8H_6ClN_3O_3$
Calculated: C, 42.22; H, 2.66; N, 18.46
Found: C, 42.48; H, 2.74; N, 18.52

2,4-Dichlorophenoxyacetic Acid. When a portion of the diazotized solution **IV** prepared from 4-nitro-2-chlorophenoxyacetic acid (**III**) was treated with freshly prepared cuprous chloride solution, a white solid was obtained. Recrystallization from benzene produced 2,4-dichlorophenoxyacetic acid which was identical with an authentic sample, as determined by infrared spectra and by mixture melting points.

3-Amino-5-nitroanisole (VII). 3,5-Dinitroanisole (**VI**) (Pfaltz and Bauer) was reduced with sodium hydrosulfide in 74% yield according to the procedure of Idoux (17) and purified by sublimation, m.p. 120 C (120 C, see ref. 16); nuclear magnetic resonance ($(CD_3)_2SO$) δ 3.82 (s, 3H); 6.57, 6.87, and 7.09 (apparent triplets, each 1H, J = 2 Hz); 5.60 (br s, 2H).

$C_7H_8N_2O_3$
Calculated: C, 50.00; H, 4.80; N, 16.60
Found: C, 50.02; H, 4.74; N, 16.76

3-Chloro-5-nitroanisole (VIII). 3-Amino-5-nitroanisole (**VII**) was diazotized with nitrous acid and then treated with cuprous chloride, following the method of Marvel and McElvain (22). Purification by sublimation produced colorless crystals in 70% yield, m.p. 99.5 to 100 C (100 C, see ref. 6); nuclear magnetic resonance ($CDCl_3$) δ 3.92 (s, 3H); 7.20, 7.61, 7.77 (apparent triplets, each 1H, J = 2 Hz).

$C_7H_6ClNO_3$
Calculated: C, 44.82; H, 3.22; N, 7.47
Found: C, 45.03; H, 3.24; N, 7.27

3-Chloro-5-nitrophenol (IX). A mixture of 3g of 3-chloro-5-nitroanisole (**VIII**) in 10 ml of conc sulfuric acid was heated 2 hr on the steam bath and was then poured onto ice. The resulting aqueous suspension was made basic with sodium carbonate, extracted with methylene chloride, and the organic layer was evaporated to dryness under reduced pressure. The entire process was repeated on the residue. Purification by sublimation gave 2.5 g (90%) of pale yellow crystals, m.p. 147.5 to 148 C (147 C, see ref. 3); nuclear magnetic resonance ($(CD_3)_2SO$) δ 7.22, 7.52, 7.62 (apparent triplets, each 1 H, J = 2 Hz); 10.83 (br s, 1 H).

$C_6H_4ClNO_3$
Calculated: C, 41.52; H, 2.32; N, 8.07
Found: C, 41.64; H, 2.45; N, 7.88

3-Chloro-5-nitrophenoxyacetic Acid (X). To a solution of 2 g (11.5 mmoles) of 3-chloro-5-nitrophenol (**IX**) and 0.46 (11.5 mmoles) of sodium hydroxide in 5 ml of H_2O stirred at reflux were added dropwise a solution of 15 mmoles of sodium bromoacetate in 2 ml of H_2O. The resulting solution was stirred at reflux for 3.5 hr and then poured into 50 ml of H_2O. The solution was adjusted to pH 1 with HCl, and the resulting solid was extracted with ethyl acetate. The combined organic extracts were dried over anhydrous sodium sulfate and evaporated to dryness under reduced pressure. To remove residual **IX** from the residue, 10 ml of acetic anhydride and two drops of concentrated H_2SO_4 were added. The resulting solution was warmed 15 min, chilled on ice, and poured into 100 ml of ice water. The aqueous suspension was made basic with sodium carbonate, extracted with ethyl acetate, and the organic extracts were dried and reduced to dryness under diminished pressure. The residue was purified by sublimation to give 2.3 g (87%) of colorless crystals, m.p. 139 to 140 C; nuclear magnetic resonance ($(CD_3)_2SO$) δ 4.92 (s, 2H); 7.53, 7.70, 7.82 (apparent triplets, each 1 H, $J = 2$ Hz).

$C_8H_6ClNO_5$
Calculated: C, 41.49; H, 2.61; N, 6.05
Found: C, 41.69; H, 2.67; N, 6.08

3-Azido-5-chlorophenoxyacetic Acid (V). This compound was prepared from 3-chloro-5-nitrophenoxyacetic acid (**X**) by the same procedure described for producing the isomer **I.** Sublimation gave 2.3 g (71% yield), m.p. 138.5 to 140 C; UV $\lambda_{max}^{95\% \text{ ethyl alcohol}}$ 294 nm (ϵ 1,800), 285 (2,210), 253 (9,250); nuclear magnetic resonance ($(CD_3)_2SO$) δ 4.79 (s, 2H), 6.73 (m, 3H); infrared (KBr) ν_{max} 2120 cm^{-1} ($-N_3$), 1755 (C=O); mass spectrum *m/e* (relative intensity) 227 (M^+, 20.6), 199 (52.5), 140 (100), 112 (47.1), 76 (47).

$C_8H_6ClN_3O_3$
Calculated: C, 42.22; H, 2.66; N, 18.46
Found: C, 42.46; H, 2.76; N, 18.30

Bioassay Procedure. Auxin activity was determined on the basis of the elongation of pea stem segments and oat coleoptile segments (4, 27). In the case of the peas, 10-mm segments were taken from immediately below the "hook" of 7-day-old, dark-grown Alaska pea seedlings. Ten (or 12) segments were floated on 6 ml of test solution, pH adjusted to 6, in 5-cm diameter plastic Petri dishes in the dark. All necessary handling of solutions and tissues was done in the dark room with ruby red light. Treatment periods were generally 18 hr, but varied from 5 to 24 hr. In the case of presoak treatments, the tissues were floated as above on the initial test solution for from 1 to 5 hr, rinsed several times in distilled H_2O, and then floated on the final test solutions. Oat coleoptile segments were used for the early assays of compound **I,** 10-mm segments being taken from 5 mm below the coleoptile tip. The conditions of the oat assay were the same as for the pea assay except for the addition of 2% sucrose to all solutions.

2,4-D was used as the standard for the evaluation of auxin activity and for the detection of additive and/or competitive effects. The test compounds 4-azido-2-chlorophenoxyacetic acid (**I**) and 3-azido-5-chlorophenoxyacetic acid (**V**) were supplied at the rate of 0.2, 1, 5, and 25 mg/l. The time periods and concentration ranges were varied to optimize differences between treatments and reproducibility of results.

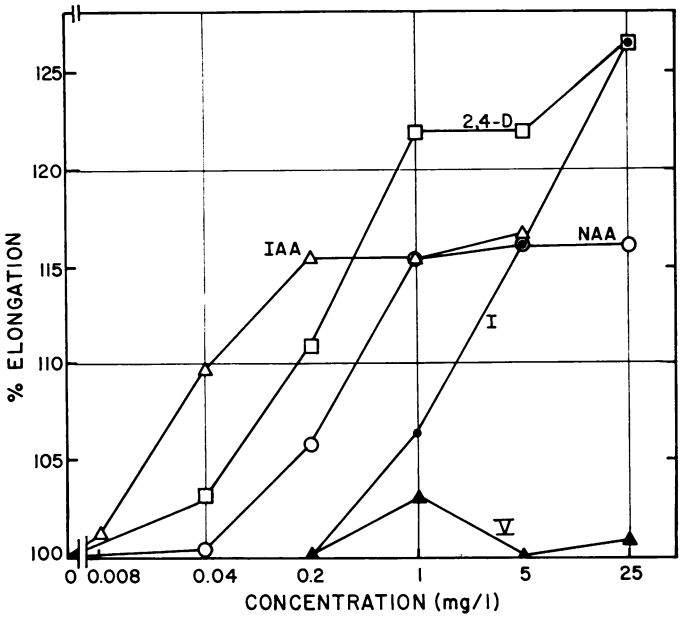


FIG. 1. Effects of compounds **I** and **V** on pea stem elongation as compared with the effects of IAA, 2,4-D, and NAA. Elongation is expressed as percentage of the water controls. Initial length of segments: 10 mm; final length of controls: 10.8 mm. Bioassay period: 19 hr. Data from experiment P-10 (15 Mar. 1973), except for curve V, which is from experiment P-11 (14 Nov. 1973).

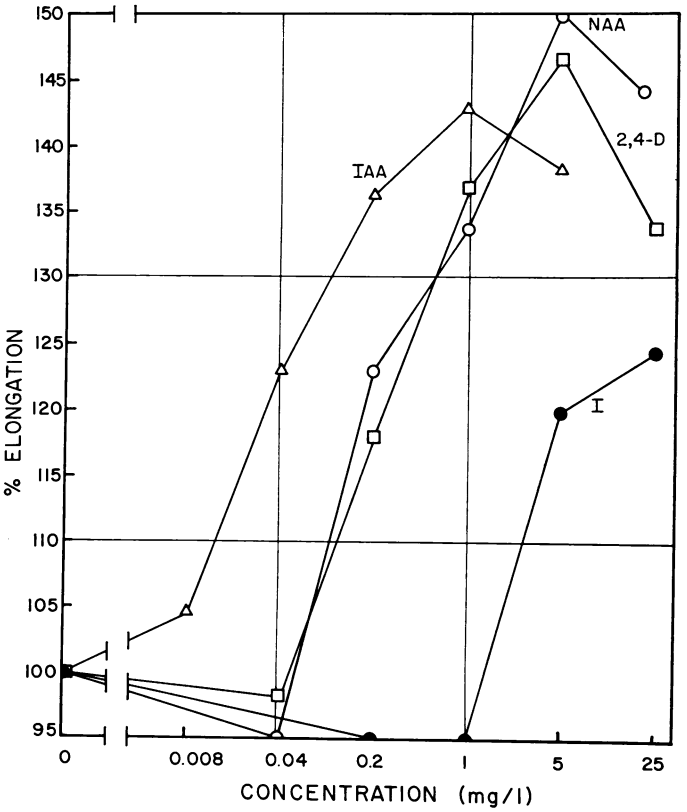


FIG. 2. Effect of compound **I** on oat coleoptile elongation as compared with the effects of IAA, 2,4-D, and NAA. Elongation is expressed as percentage of the sucrose controls. Initial length of segments: 10 mm; final length of controls: 15.2 mm. Bioassay period: 20 hr. Experiment O-16 (14 Mar. 1973).

Table I. *Summary of All Pea Stem Segment Assays for Auxin Activity*

Compound and Conditions	Assays	Activity		Results
		Auxin	Anti-auxin	
	No.			
Presoaked in H_2O, tested with 2,4-D	6			5 assays: presoaking reduced response to 2,4-D
				1 assay: no effect
Not presoaked, tested with **I**	11	+	−	Consistently active at 1 to 25 mg/l (about $\frac{1}{5}$ as active as 2,4-D)[1]
Presoaked in **I**, tested with 2,4-D	2	+	−	Additive effects of **I** and 2,4-D
Not presoaked, tested with 2,4-D + **I**	2	+	−	Additive
Not presoaked, tested with **V**	4	−	−?	3 assays: inactive
				1 assay: slight but consistent inhibition of growth
Presoaked in **V**, tested with 2,4-D	7	−	−	5 assays: same as with water presoaked controls
				1 assay: ≤ water presoaked controls
				1 assay: ≥ water presoaked controls
Not presoaked, tested with 2,4-D + **V**	6	−	−	4 assays: = 2,4-D alone
				1 assay: sl. < 2,4-D alone
				1 assay: > 2,4-D alone

[1] In eight tests on *Avena* sections **I** was consistently active, but about $\frac{1}{25}$ as active as 2,4-D.

RESULTS AND DISCUSSION

This is the first time that a photoaffinity-labeled compound has been shown to have activity in plants and especially in intact tissue. Specifically, 4-azido-2-chlorophenoxyacetic acid (**I**) was found to have auxin activity qualitatively similar to that of 2,4-D in direct comparative tests of IAA, NAA, 2,4-D, and compound **I.**

Auxin Activity Measurements. Comparison of the activities of compound **I,** the 4-azido-2-chloro, and compound **V,** the 3-azido-5-chloro derivative, with the auxins 2,4-D, IAA, and NAA in the promotion of elongation of pea stem segments is shown in Figure 1. Although compound **I** is definitely less active than any of these three commonly used and highly active auxins, it does have moderate activity and if supplied in sufficiently high concentration will give the maximum growth response. Compound **V,** on the other hand, does not give a significant increase in growth over that of the controls. In this particular experiment, **I** appears to be roughly one-twentieth as active as 2,4-D, but in other experiments the difference between the two was less. From a summary of all results presented in Table I, compound **I** was found to be about one-fifth as active as 2,4-D in the pea segment test performed in darkness except for brief exposure to red light as specified.

In comparisons of the activities of compound **I** with those of 2,4-D, IAA, and NAA in the oat coleoptile segment test, it was found to be about one-twenty-fifth as active as 2,4-D, as shown by the results of one test in Figure 2, and this was generally the case in other tests. The highest concentration used was 25 mg/l, so that unfortunately the maximum growth response was not demonstrated in the oat coleoptile tests.

Table II. *Pea Stem Segment Elongation after 16-hr Treatment (P21: 6-27-74)*

In both experiments, the initial length of segments was 10 mm, and the final length of controls was 11.5 mm.

Compound	Concn	Elongation
	mg/l	%
Not presoaked H_2O		100
2,4-D	0.2	106
2,4-D	1	110
2,4-D	5	114
I	5	110
I	25	114
V	5	97
V	25	97

Compound	Concn	Elongation at Concn of 2,4-D (mg/l)		
		0.2	1.0	5.0
	mg/l	%		
Presoaked 1 hr H_2O		104	108	110
I	5	115	116	113
I	25	116	116	113
V	5	109	111	113
V	25	108	110	111

Combined Treatments with 2,4-D and Compounds I or V. An attempt was made to determine the effects of compounds **I** and **V** on the response of pea stem segments to 2,4-D. The data in Table II indicate that some increase in growth was obtained by presoaking with either **I** or **V** as compared with presoaking in water. However, presoaking in water decreased the response to subsequently added 2,4-D, so that the effect of compound **V** was no more than to prevent this decrease, and in six out of seven experiments presoaking in **V** gave no greater elongation than presoaking in water. Presoaking in compound **I** resulted in a greater increase in growth so that at least for the low 2,4-D concentrations (0.2 and 1 mg/l) a slight growth promoting effect was obtained with the presoaking treatment. In treatments with different combinations of 2,4-D and compound **I,** no more than additive effects of the two substances were obtained. In similar combinations with 2,4-D, compound **V** had no significant influence on growth. A summary of all such experiments is included in Table I.

We conclude from the experiments performed thus far that compound **I,** as a moderately active auxin and additive in effect with 2,4-D, may be utilizable in photoaffinity labeling experiments. Compound **V,** as an inactive isomer analogous to 3,5-D, did not influence the effect of 2,4-D on growth in experiments in which combined treatments with the two compounds were used. Accordingly, compounds **I** and **V** have different affinities for binding sites involving growth-regulatory action. It is to be hoped that bound products of photolysis of the two isomers

will permit discrimination between active site-directed binding and less specific binding.

LITERATURE CITED

1. Åberg, B. 1973. Plant growth regulators 31. Some monochloromonomethylphenoxy-acetic and related optically active propionic acids. Swed. J. Agr. Res. 3: 49–62.
2. Åberg, B. 1971. Plant growth regulators 28. The optically active dichlorophenoxyacetic acids. Swed. J. Agr. Res. 1: 23–32.
3. Blanksma, J. J. 1908. Bromuration et nitration de phénols metasubstitués. Recl. Trav. Pays-Bas Belg. 27: 24–41.
4. Bonner, W. D., Jr. and K. V. Thimann. 1949. Experiments on the growth and inhibition of isolated plant parts. Am. J. Bot. 36: 214–221.
5. Browne, D. T., S. S. Hixson, and F. H. Westheimer. 1971. A diazo compound for the photochemical labeling of yeast alcohol dehydrogenase. J. Biol. Chem. 246: 4477–4484.
6. Browne, D. W. and G. M. Dyson. 1931. The inhibitory effect of substituents in chemical reactions. J. Chem. Soc. (Lond.) : 3285–3308.
7. Browne, J. P. and E. B. McCall. 1955. Some chlorinated hydroxyphenoxyacetic acids. J. Chem. Soc. (Lond.) : 3681–3687.
8. Converse, C. A. and F. F. Richards. 1969. Two-stage photosensitive label for antibody combining sites. Biochemistry 8: 4431–4436.
9. Cooperman, B. S. and D. J. Brunswick. 1973. On the photoaffinity labeling of rabbit muscle phosphofructokinase with $O^{2'}$-(ethyl-2-diazomalonyl)-adenosine 3′:5′-cyclic monophosphate. Biochemistry 12: 4079–4084.
10. Fleet, G. W. J., J. R. Knowles, and R. R. Porter. 1972. The antibody binding site. Labelling of a specific antibody against the photoprecursor of an aryl nitrene. Biochem. J. 128: 499–508.
11. Fredga, A. and B. Åberg. 1965. Stereoisomerism in plant growth regulators of the auxin type. Annu. Rev. Plant Physiol. 16: 53–72.
12. Galardy, R. E., L. C. Craig, and M. P. Printz. 1973. Benzophenone triplet: a new photochemical probe of biological ligand-receptor interactions. Nature New Biol. 242: 127–128.
13. Hardin, J. W., J. H. Cherry, D. J. Morre, and C. A. Lembi. 1972. Enhancement of RNA polymerase activity by a factor released by auxin from plasma membrane. Proc. Nat. Acad. Sci. U.S.A. 69: 3146–3150.
14. Hertel, R., K. Thompson, and V. E. A. Russo. 1972. *In vitro* auxin binding to particulate cell fractions from corn coleoptiles. Planta 107: 325–340.
15. Hixson, S. S. and S. H. Hixson. 1973. Photochemical labeling of yeast alcohol dehydrogenase with an azide analog of NAD^+. Photochem. Photobiol. 18: 135–138.
16. Hodson, H. H. and J. S. Wignall. 1926. Preparation of 3,5-dihalogenophenols. J. Chem. Soc. (Lond.) : 2077–2079.
17. Idoux, J. P. 1970. Selective reduction of nitroaromatic compounds. J. Chem. Soc., Sec. C Org. Chem.: 435–437.
18. Katzenellenbogen, J. A. 1974. Affinity labeling of hormone binding sites. Annu. Rep. Med. Chem.: In press.
19. Katzenellenbogen, J. A., H. J. Johnson, Jr., K. E. Carlson, and H. N. Myers. 1974. Photoreactivity of some light-sensitive estrogen derivatives. Biochemistry 13: 2986–2994.
20. Katzenellenbogen, J. A., H. J. Johnson, Jr., and H. N. Myers. 1973. Photoaffinity labels for estrogen binding proteins of rat uterus. Biochemistry 12: 4085–4092.
21. Knowles, J. R. 1972. Photogenerated reagents for biological receptor-site labeling. Accounts of Chemical Research 5: 155–160.
22. Marvel, C. S. and S. M. McElvain. 1941. *o*-Chlorotoluene and *p*-chlorotoluene. *In*. A. H. Blatt, ed., Organic Syntheses, Collective Vol. I, Ed. 2. John Wiley and Sons, New York. pp. 170–172.
23. Matthysse, A. G. and C. Phillips. 1969. A protein intermediary in the interaction of a hormone with the genome. Proc. Nat. Acad. Sci. U.S.A. 63: 897–903.
24. Mondal, H., R. K. Mandal, and B. B. Biswas. 1972. RNA stimulated by indole acetic acid. Nature New Biol. 240: 111–113.
25. Ray, P. M. 1974. The chemistry and biochemistry of plant hormones. Rec. Adv. Phytochem. 6: 93–122.
26. Schafer, J., P. Baronowsky, R. Laursen, R. Finn, and F. H. Westheimer. 1966. Products from the photolysis of diazoacetyl chymotrypsin. J. Biol. Chem. 241: 421–427.
27. Schneider, C. L. 1938. The interdependence of auxin and sugar for growth. Am. J. Bot. 25: 258–270.
28. Smith, R. A. G. and J. R. Knowles. 1973. Aryldiazirines. Potential reagents for photolabeling of biological receptor sites. J. Am. Chem. Soc. 95: 5072–5073.
29. Thimann, K. V. 1972. The natural plant hormones. *In:* F. C. Steward, ed., Plant Physiology, Vol. 6B. Academic Press, New York. pp. 3–123.
30. Wain, R. L. and C. H. Fawcett. 1969. Chemical plant growth regulation. *In:* F. C. Steward, ed., Plant Physiology, Vol. 5A. Academic Press, New York. pp. 231–296.
31. Zimmerman, P. W. and A. E. Hitchcock. 1942. Substituted phenoxy and benzoic acid growth substances and the relation of structure to physiological activity. Contrib. Boyce Thompson Inst. 12: 321–343.