Phytochemistry, 1974, Vol. 13, pp. 1033 to 1035. Pergamon Press. Printed in England.

PLANT GROWTH INHIBITION BY NAPHTHOIC ACID ESTERS

J. G. BUTA and G. L. STEFFENS

Plant Physiology Institute, Plant Hormone and Regulators Laboratory, Agricultural Research Service, United States Department of Agriculture, Beltsville, MD 20705, U.S.A.

(*Received* 4 *September* 1973)

Key Word Index—*Nicotiana*; Solanaceae; tobacco; *Cucumis*; Cucurbitaceae; cucumber; growth inhibition; naphthoic acids; naphthoate esters.

Abstract—A series of alkyl esters of 1- and 2-naphthoic acids and 1- and 2-naphthaleneacetic acids were synthesized and tested for growth regulating activity in the tobacco bud and cucumber root bioassays. The *n*-propyl ester of 1-naphthoic acid was the most active bud growth inhibitor of the homologous series of alkyl esters (C_1–C_9) tested. The same maximum occurred in a struture-activity comparison of effectiveness of penetration of these compounds measured by the beet-root assay. Unesterified 1-naphthoic acid was the most active growth inhibitor in the cucumber root assay and the inhibition decreased as the alkyl chain length of the esterified acid was increased.

INTRODUCTION

A SERIES of naphthoic acid derivatives have been prepared to study the relationship of the plant growth regulating activity of simple aromatic acids to their structures and lipid solubilities. 1-Naphthoic acid has weak auxin-like activity when tested in pea and tomato bioassays.[1,2] Extensive chemical modifications of the basic structure were made and auxin activity was found to be greatly affected by specific structural requirements.[3] In this report, the effects of modification of lipid solubility by esterification of, primarily, 1-naphthoic acid have been investigated for effects on bud growth and seed germination. The variation in penetration due to differences in lipid solubilities of the various test compounds have also been studied by examination of betacyanin efflux from beet root.

The naphthoic acids were converted to alkyl esters to increase their lipid solubilities and so enhance penetration into sites of growth regulating activity of intact plants.[4] One test system used involved the measurement of axillary bud growth that occurs after decapitation of *Nicotiana* cv. Xanthi.[5] The compounds were sprayed onto plants as emulsions to maximize uptake.[6]

RESULTS AND DISCUSSION

The greatest inhibition of axillary bud growth was obtained with the 3-carbon chain length esters of 1-naphthoic acid. There was a decrease in activity as the alkyl moiety was

[1] MITSUI, T. (1951) *J. Agric. Chem. Soc. Japan* **24**, 465.
[2] FUJITA, T., KASHIMIZU, K., IMAI, S. and MITSUI, T. (1960) *Kyoto Univ. Chem. Res. Bull.* **38**, 76.
[3] FUJITA, T., KAWAZU, K., MITSUI, T. and KATSUMI, M. (1967) *Phytochemistry* **6**, 889.
[4] CRAFTS, A. S. (1960) *Weeds* **8**, 19.
[5] MARTH, P. C. and MITCHELL, J. W. (1964) *J. Agric. Food Chem.* **12**, 61.
[6] BUTA, J. G. and STEFFENS, G. L. (1971) *Physiol. Plant.* **24**, 431.

shortened or lengthened (Table 1). The unsaturated esters (allyl, propargyl) were somewhat less active than the corresponding saturated (propyl) ester. Chain branching of the alkyl ester moiety also decreased activity somewhat. The synthetic auxin, 1-naphthaleneacetic acid, when esterified caused complete inhibition of bud growth and extensive plant deformation. The deformation, primarily epinasty, was not seen with 1-naphthoic acid and derivatives. The isomer, 2-naphthoic acid, was inactive. The 2-naphthaleneacetic acid was similar in effect to the 1-substituted isomer, but less active.

TABLE. 1. GROWTH INHIBITION* BY NAPHTHOIC ACIDS AND THEIR ESTERS†

	R^1H	R^1CH_3	$R^1C_2H_5$	$R^1C_3H_7$	$R^1C_4H_9$	$R^1C_5H_{11}$	$R^1C_6H_{13}$	$R^1C_7H_{15}$	$R^1C_8H_{17}$	$R^1C_9H_{19}$
Axillary bud	37	35	85	93	59	45	40	39	29	29
Cucumber root	90	93	86	69	50	48	43	30	33	15
Beet root	0	0·06	0·08	0·19	0·05	0·01	0·01	0·02	0·01	0·04
	R^2H	$R^2C_3H_7$	R^3H	$R^3C_3H_7$	R^4H	$R^4C_3H_7$	$R^1CH_2CH{=}CH_2$		$R^1CH_2C{\equiv}CH$	$R^1CH(Me)_2$
Axillary bud	100	100	20	48	60	68	71		79	97
Cucumber root	90	90	64	75	64	75	76		72	60

R^1 = 1-$C_{10}H_7CO_2$- (1-Naphthalenecarboxy-)
R^2 = 1-$C_{10}H_7CH_2CO_2$- (1-Naphthaleneacetic-)
R^3 = 2-$C_{10}H_7CO_2$- (2-Naphthalenecarboxy-)
R^4 = 2-$C_{10}H_7CH_2CO_2$- (2-Naphthaleneacetic-)

* Per cent inhibition expressed as control minus test/control × 100—Axillary bud assay: Per cent by weight reduction; Cucumber root assay: Per cent length reduction; Beet root assay: absorbance of betacyanin.

† Concentrations: Axillary bud assay, 2×10^{-2} M; Cucumber root assay, 10^{-3} M; Beet root assay, 10^{-3} M.

Another assay used to study activity was the inhibition of primary root growth of germinating cucumber seeds.[7] In this assay, a constant supply of test compound was in contact with the germinating seed. In contrast to the results from the axillary bud growth tests, the most inhibitory 1-naphthoate found was the free acid and the methyl ester (Table 1). With esterification, the inhibitory activity decreased as the chain length of the ester moiety was increased. Unsaturated or branched chain esters were similar in activity to the corresponding saturated compounds. The structure-activity relationships for the 2-naphthoates, and 1- and 2-naphthaleneacetates in this root assay were similar to those found in the bud inhibition assay.

The results of these assays suggested that the growth regulating activity of the 1-naphthoates was affected by differences in lipid solubility or polarity of the various esters caused by chain length variation. That these naphthoates had different polarities was confirmed by use of TLC procedure that demonstrated differences in partitioning of compounds between lipoidal and aq. phases of various solvent systems.[8] The lipid solubility of the 1-naphthoate esters increased as the chain length of the alkyl portion of the ester moiety was increased. The unsaturated and branched chain esters were slightly more polar than the corresponding saturated linear compounds.

There is a question as to the facility of movement of compounds such as naphthoic acid and esters through plant cuticles and membranes into cells where growth is occurring.[9] The bud assay tests whether movement of compounds is affected by the presence of a cuticle. In the cucumber seedling root growth assay, there would not be any significant lipoidal

[7] READY, D. and GRANT, V. O. (1968) in *Methods of Studying Plant Hormones and Growth Regulating Substances* (MITCHELL, J. W. and LIVINGSTON, G. A. eds.), p. 84, U.S. Department of Agriculture.
[8] BOYCE, C. B. and MILLBORROW, B. V. (1965) *Nature* **208**, 537.
[9] BLUMENFELD, A. and BUKOVAC, M. J. (1972) *Planta* **107**, 261.

barrier to uptake. Cellular pentration by the test compounds was measured as a function of the leakage of betacyanin from *Beta vulgaris* root sections.[10] The results from experiments using emulsions of the 1-naphthoate esters (C_1–C_9) demonstrated a structure-activity relationship similar to that observed in the bud-growth inhibition tests (Table 1). The unesterified 1-naphthoic acid did not cause leakage of betacyanin under these conditions.

The growth inhibiting activities of the longer chain 1-naphthoate esters (C_4–C_9) also appear to decrease as a function of increasing lipid solubility in the cucumber root growth tests. The free acid is the most active inhibitor of the group of compounds in the assay. The decrease in activity seen with the esters may not be due solely to a difference in penetration and the resulting variation in concentration of available compounds. This has been demonstrated with 2,4-D esters utilizing this assay.[11] Differences in the rate of hydrolysis of naphthoate esters could also be occurring in the tobacco bud assay.

No growth regulating activity of 1-naphthoic acid has been reported for intact plants.[12] Esterification modified lipid solubility of the acid such that sufficient biologically active compound penetrated to the sites of activity, and inhibition of plant growth resulted. The optimum growth regulating effect of the acid was achieved by conversion to the propyl ester. This optimal structure-polarity relationship has been found also for both biologically active and contact-type biologically inactive dichlorobenzoate esters.[13,14]

EXPERIMENTAL

The naphthoate esters were prepared by standard methods and confirmed by IR, TLC and GLC. The tobacco bud growth assay involved the spray application of test compounds emulsified with Tween-20 on decaptiated *Nicotiana* cv Xanthi-nc plants.[5] Emulsified preparations were also used in the cucumber root growth assay.[7] The TLC procedure made use of paraffin-coated silica gel plates with 70% Me_2CO–H_2O as the developing solvent.[8] The beet root assay was modified by the use of emulsified compounds at concentrations such that turbidity was minimized.[10]

Acknowledgement—We appreciate the technical assistance of Michael T. Mudd.

[10] SIEGEL, S. M. (1969) *Physiol. Plant.* **22,** 327.
[11] MORRE, D. J. and ROGERS, B. J. (1958) *Proc. N. Cen. Weed Contr. Cong.* 15*th*, p. 3, Chicago.
[12] SINGH, J. P. (1957) *Ind. J. Hort. Sci.* **14,** 145.
[13] BUTA, J. G. and STEFFENS, G. L. (1970) *J. Agric. Food Chem.* **18,** 536.
[14] STEFFENS, G. L. and BUTA, J. G. (1970) *Am. J. Botany* **57,** 1055.