

n-caproic acid at pH 6.3. After washing carefully the fibrin was resolved in alkaline urea and measured photometrically. As a next step a mixture of 10 uniformly labeled amino acids C^{14} was given to control, SNL or phenobarbital treated rats. In each animal 5 $\mu C/100$ g body wt. was given i.p. Blood was taken 2, 3, 6 and 8 h later and the radioactivity of fibrin was measured by using a Packard Tri-Carb liquid scintillation system. The isolation of fibrin has been done as mentioned earlier, but the solubilization of fibrin clot for counting was performed according to ATENCIO and LORAND⁷.

Results and discussion. As can be seen in Table I, it is obvious that there is a significant increase of liver weight as well as plasma fibrinogen both in the SNL and phenobarbital treated groups on the 4th day.

The increase is more pronounced in the phenobarbital treated group than in the animals treated with SNL. The incorporation of the labeled amino acids into fibrin isolated from the SNL treated rats is about the double, compared with the control groups and even higher after phenobarbital treatment. The peaks of incorporation in time are also changed both in the SNL and phenobarbital treated groups. In these cases the maximums of radioactivity are at 3 h after giving the labeled amino acids while in controls the peak is at 6 h.

On the basis of our findings we can say that both SNL and phenobarbital are capable of inducing an enhanced synthesis of fibrinogen and this results in an elevated plasma fibrinogen level after treatment for 3 days.

ATENCIO and LORAND⁷ have found that a single dose of ACTH given to rabbits caused a similar increase of plasma fibrinogen 24 h later but a far more enhanced incorporation (more than 10-fold increase was noted) even 2 h after giving ACTH. This effect of ACTH seems not be mediated through corticosterone since this steroid did not have any effect either on the plasma fibrinogen or on the rate of incorporation. It is worth mentioning that corticosterone was given in this experiment in a single dose.

Our investigations showed that SNL as a steroidal compound given for 3 consecutive days increased fibrinogen synthesis. This might represent an interesting way of influencing fibrinogen metabolism by steroids. Phenobarbital has the same but more pronounced effect in rats. This fact means that fibrinogen metabolism is able to meet adaptation in large scale and can be influenced by several structurally unrelated substances. Eventually – these drugs known as microsomal enzyme inducers – influenced the synthesis of an enzymatically inactive protein of the liver.

Recently it has been reported by REMMER and CASALS⁸ that phenobarbital provokes an enhanced synthesis of the albumin in the liver and this results in an elevated plasma albumin level of rats. The functional and pathological aspects of the elevated fibrinogen level in blood under the effect of inducers are still questioned and unsolved.

Résumé. Par un dosage de 20 $\mu M/100$ g poids de spironolactone ou de phénobarbital pendant 3 jours, un niveau de fibrine élevé peut être produit chez des rats. En même temps, l'incorporation des aminoacides C^{14} au fibrinogène est aussi intense. Le phénobarbital est dans tous les cas plus efficace que le spironolactone. Dans notre cas, ces médicaments, connus comme inducteurs des enzymes microsomiques, augmentaient la synthèse protéique, enzymatiquement inactive du foie.

S. TÓTH, T. SZILÁGYI and J. CSONGOR

Pathophysiological Institute of University Medical School, Debrecen 12 (Hungary), 2 May 1972.

⁷ A. C. ATENCIO and L. LORAND, *Am. J. Physiol.* 219, 1161 (1970).

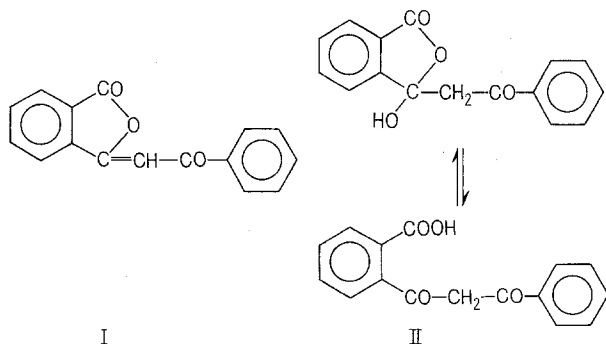
⁸ H. REMMER and J. CASALS, *Naunyn-Schmiedeberg's Arch. Pharmacol.* 269, 455 (1971).

New Inhibitors of Auxin Transport

While investigating the effects of certain lactones on the development of the crucifer *Arabidopsis thaliana* grown under controlled conditions¹, we noted that 3-phenacylidene phthalide I and its hydrate II caused a complete loss in root geotropism when incorporated into the growth nutrient at $2 \times 10^{-5} M$. The synthetic plant growth regulators 2, 3, 5-triiodobenzoic acid (TIBA) and 1-naphthylphthalamic acid (NPA) are both highly effective inhibitors of auxin transport^{2,3} and at low

concentrations both abolish the normal positive geotropic response of seedling roots^{4,5}. In the present work we have found that I and II are inhibitors of basipetal auxin transport of activity comparable to that of TIBA and NPA.

A quantitative assessment of the effect of I and II on root curvature was made using cress (*Lepidium sativum*) and ryegrass (*Lolium rigidum*) based on the procedure of JONES et al.⁵. Germinated seeds of both species with straight roots 1–2 cm long were transferred to the surface of agar plates containing the compounds under test at various concentrations. The plates were then placed in the dark in a vertical plane such that the initial direction of root growth was horizontal. In untreated plates



¹ B. T. BROWN, *Pestic. Sci.*, 3, 161 (1972).

² G. W. KEITT and R. A. BAKER, *Pl. Physiol.* 41, 1561 (1966).

³ C. C. MCCREADY, in *Biochemistry and Physiology of Plant Growth Substances* (Ed. F. WIGHTMAN; Runge Press, Ottawa 1968), p. 1005.

⁴ P. K. FUENTE and A. C. LEOPOLD, in *Biochemistry and Physiology of Plant Growth Substances* (Ed. F. WIGHTMAN; Runge Press, Ottawa 1968), p. 1129.

⁵ R. L. JONES, T. P. METCALF and Q. A. SEXTON, *J. Sci. Fd Agric.* 5, 32 (1954).

Table I. Effects on root geotropism

Molar concentration	Cress				Ryegrass			
	4×10^{-6}	4×10^{-7}	4×10^{-8}	4×10^{-9}	4×10^{-6}	4×10^{-7}	4×10^{-8}	4×10^{-9}
Compound I	++	++	++	+	++	+	+	—
Compound II	++	++	++	+	++	+	+	—
TIBA	+	+	—	—	+	—	—	—
NPA	++	+	—	—	++	+	—	—
					(++) ^a	(+) ^a		

Ratings: ++, complete loss of root geotropism; +, partial loss; —, no loss. ^a Ratings recorded in reference⁵.

subsequent root growth produced a change in direction of 90°, whereas with a complete loss in root geotropism subsequent growth continued horizontally with no change in direction. The effects on root growth after 24 h are recorded in Table I, and indicate that in this assay both I and II are more effective than TIBA or NPA in interfering with root geotropism.

Stem application of auxin transport inhibitors can lead to a loss in apical dominance and accelerated growth of lateral buds⁶. Accordingly a comparison was made between the effects of stem applications of II and TIBA on

apical dominance in *A. thaliana*. A fine dispersion of each compound in lanolin (0.1%) was applied to 14-day-old plants as a complete circle around the stem 3 cm above the rosette. Six days later some reduction in growth at the terminal bud was observed in each treatment group. With both compounds apical dominance was lost resulting in considerable secondary growth particularly from the rosette leaf axils (Figure). This suggests that at this concentration II was as effective as TIBA in blocking apical dominance.

The activity of I and II in preventing the basipetal movement of auxin was compared to that of TIBA using isotopically labelled indol-3-ylacetic acid and isolated petiole segments from *Phaseolus vulgaris*. Donor blocks of agar gel were prepared containing 1.0 µg per ml IAA-2-C14 (2 cm diameter, 0.5 ml volume, 1.5% agar, IAA activity 30 mCi per mM). These were then placed on the distal ends of a group of 20 1 cm segments cut from the petioles of *P. vulgaris*. Similar receiver blocks of plain agar gel (control) or blocks containing various concentrations of I, II or TIBA were then placed at the proximal ends of the segments. The segments were maintained in a humid environment as described² and the radioactivity transported into the receiver blocks during a 4 h period was determined. The results in Table II indicate that both I and II inhibit IAA transport under these conditions at concentrations comparable to those at which TIBA and NPA are active.

It is of interest that other derivatives of benzoic acid substituted in the *ortho* position with hydroxyl or carbonyl groups^{5,7,8} also show effects on the response of seedlings to gravity. The facile synthesis⁹ of analogous of I and II provides an opportunity to determine structure-activity requirements in these compounds.

Zusammenfassung. 3-Phenacylidinphthalid und dessen Hydrat verursachen einen konzentrationsabhängigen Verlust der geotropischen Reaktion der Wurzel und die Aufhebung der Apikaldominanz des Sprosses. Es wird nachgewiesen, dass die beiden Substanzen primär den basipetalen Transport des endogenen Auxins hemmen.

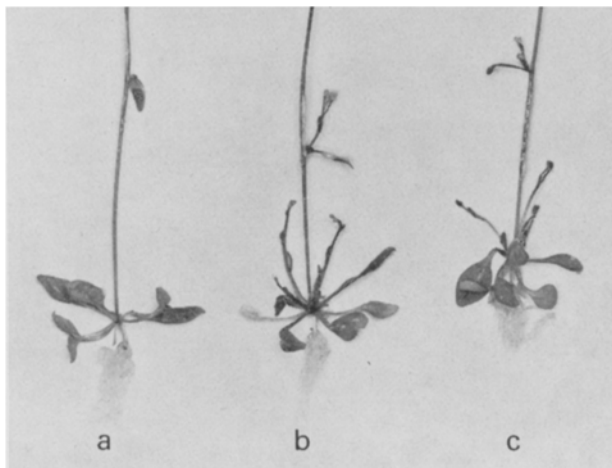
B. T. BROWN, O. JOHANSEN and W. H. F. SASSE

CSIRO, Division of Plant Industry, P. O. Box 1600, Canberra 2601, and Division of Applied Chemistry, Sydney 2070 (Australia), 17 March 1972.

Table II. Percentage radioactivity transported through isolated segments from *Phaseolus vulgaris*^{a, b}

Compound	Molar concentration in receiver block					
	10^{-4}	10^{-5}	10^{-6}	10^{-7}	10^{-8}	10^{-9}
I	5	16	44	61	98	92
II	2	4	20	32	52	62
TIBA	5	11	41	75	112	97
TIBA ^c	5	5	30	70	100	96
NPA ^c	<5	<5	<5	30	75	82

^a Expressed as percentage of the radioactivity transported through control segments. ^b Each value is the mean of 2 experiments. ^c Calculated from Figure 4 of reference².



Secondary growth on *Arabidopsis thaliana* following stem application of a) lanoline; b) compound II (0.1%) in lanoline; c) TIBA (0.1%) in lanoline.

⁶ V. W. MITCHELL, P. C. MARTIN and G. D. FREEMAN, J. Agr. Food Chem. 13, 326 (1965).

⁷ T. KOZUMI and H. HARADA, Nature, Lond. 226, 559 (1970).

⁸ A. BERRIE, Nature, Lond. 233, 347 (1971).

⁹ To be described elsewhere.