Botany. — *The effect of phenoxycompounds on waterplants.* By G. L. FUNKE. (From the Botanical Institute, Government University, Ghent.) (Communicated by Prof. W. H. ARISZ.)

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It is only a short time since phenoxycompounds have become known as plant growth regulators, but their remarkable and manifold effects have created quite a sensation among plant physiologists. They are applied as weed killers (7, 8, 11, 13), for inducing parthenocarpy (5, 6, 10, 15), different correlative and formative effects (1, 12, 14, 15, 16), inhibiting of seed germination (4), etc.

ZIMMERMAN and HITCHCOCK (15*b*) describe very peculiar modifications of leaves due to a treatment with several phenoxycompounds. They reminded me of the effect of α-naphtylacetic acid and indol compounds on the leaves of waterlilies which I observed in former experiments (2 *e*). In Nymphaeaceae we see a succession of different leaf forms, a more or less gradual transition from the primary sagittate leaves, which remain submerged under all circumstances, towards the secondary, round, floating leaves. In my experiments the secundary floating leaves became sagittate under influence of the hormones above mentioned (in a concentration of 3 mg/L) and therefore I called it a reversal to the primary form. I was anxious to see whether phenoxycompounds have a similar effect and therefore I made some experiments with 4-chlorophenoxyacetic acid and 2,4-dichlorophenoxyacetic acid. The plant material was grown in aquaria of 40 cm deep which were kept in hothouses. The apparatuses of my laboratory having been neglected in my absence from Ghent during the war and nobody being available to put them in working order, I was dependent on the temperature of these hothouses (20°—24°). The summer, unfortunately, was very cool, and consequently I could not keep the water at optimal temperatures. The results, therefore, cannot be accurately compared with those obtained with other growth regulators, but nevertheless may be considered sufficiently interesting to justify this report.

Nymphaea odorata. Young specimens were transplanted from a shallow pond into aquariums, each specimen separately. After 8 or 10 days the plants had recovered and had formed new leaves; then the substances were added. In the solutions of 2,4-dichlorophenoxyacetic acid the reaction became visible after 1, 2 or 3 days; the petioles showed a renewal of growth with the consequence that they soon meandered on the surface. The reaction was far from being as explosive as with α-naphtylacetic acid (2 *f, g, h, i, j, k, l, m*), but a constant elongation was maintained during a more or less large number of days. The maximum growth per day did not exceed 25 cm, whereas in solutions of α-naphtylacetic acid it can be 50, 60, 70 and even 90 cm per day. But here especially we should not forget that my plants had not the occasion to grow at a temperature of 30°. I could confirm the observations in my former experiments, viz. that only those leaves reacted strongly which were neither very young nor quite adult; leaves which at the beginning of the experiment had reached about half the height of the aquarium, therefore with petioles about 20 cm long and with blades still folded, showed the best reaction. Three experiments gave identical results; in table 1 a selection is given of the ultimate lengths reached in the different concentrations.

TABLE 1.

Ultimate lengths of the petioles of Nymphaea odorata in solutions of phenoxycompounds.

Subst.	Conc. in mg/L	Ultimate lengths in cm	Reached after-days	Condition of plants at end of exp.
Control		45— 52— 55	36	good
2,4-dichl.	$\frac{1}{4}$	178—185—209	29	good
	$\frac{1}{2}$	223—238—262	29	some decaying
	1	198—221—225	20	decaying
	2	185—196—258	17	decaying
4-chlor.	$\frac{1}{2}$	65— 75— 90	36	good
	1	65— 80—100	36	good
	2	111—113—165	36	good

The response in the solutions of 2,4-dichlorophenoxyacetic acid 1 and 2 mg/L was rapid, but came soon to an end because blades and petioles began to decay; not over their whole surface or length, but in definite places. The optimal concentration for longitudinal growth must be somewhat below $\frac{1}{2}$ mg/L, which is higher than for α-naphtylacetic acid where it is between $\frac{1}{8}$ and $\frac{1}{4}$ mg/L for most Nymphaeaceae. The response to 4-chlorophenoxyacetic acid is much slower and its effect is much less; up till now I have not had the occasion to determine its optimal concentration.

The young leaves which at the beginning of the experiment had not yet unfolded their blades, kept them in the rolled-up position; the youngest leaves with outspread blades showed a hyponastic rolling-up; the older ones did not react any more. Otherwise no formative effects of the substances could be observed. Higher concentrations are perhaps needed for phenomena of reversal to the primitive leaf form, but they would probably inhibit any development owing to their noxious effect.

Nymphaea terminalis — *N. Devoniensis.* The specimens were taken from material in the Victoria basin; the experiments were made twice; the results are summarized in table 2.

We see again that, at least as far as N. terminalis is concerned, 4-chlorophenoxyacetic acid has hardly any influence in a concentration in which 2,4-dichlorophenoxyacetic acid is very effective. The specimen of N. Devoniensis in 2,4-dichlorophenoxyacetic acid 1 mg/L of 293 cm in-

TABLE 2.

Ultimate lengths of the petioles of Nymphaea Devoniensis and N. terminalis in solutions of phenoxycompounds.

Subst.	Conc. in mg/L	Ultimate lengths in cm	
		N. Devoniensis	N. terminalis
Control		45— 50— 65	45— 60 —70
2,4-dichl.	$\frac{1}{4}$	65— 70— 71	81— 87— 90
	$\frac{1}{2}$	80— 82— 90	140—168—186
	1	125—150—192—293	166—195—203
4-chlor.	$\frac{1}{2}$	70— 72— 75	78— 82— 90

dicates that a very strong reaction is possible, but during these experiments it was shown in this case only.

The reaction of the leafblades was very feeble; only those which had not yet reached the surface at the beginning of the experiment kept their blades in rolled-up position.

Limnanthemum nymphaeoides. The material was taken from a very shallow pond outdoors. The petioles did not measure more than 10—12 cm; when planted in the aquaria of 40 cm, these petioles started a new growth immediately and after 24—36 hours the blades were floating again. This proves that the plants were in excellent condition (comp. 2*d*, *g*; 3). The experiments were made twice, the results can be seen in table 3.

TABLE 3.

Ultimate lengths of the petioles of Limnanthemum nymphaeoides in solutions of phenoxycompounds.

Subst.	Conc. in mg/L	Ultimate lengths in cm	Reached after-days	Condition of plants at end of exper.
Control		50— 57— 60	16	good
2,4-dichl.	$\frac{1}{2}$	83— 83—132	10	poor
	1	149—152—180	11	poor
	2	85— 90—100	5	decaying
4-chlor.	1	85—100—119	16	poor

The concentration 2 mg/L of 2,4-dichlorophenoxyacetic acid is distinctly noxious; after no more than 5 days the experiment had to be discontinued because the petioles were hardly measurable owing to their strong coiling; they broke easily and were partly decaying. 1 mg/L and even $\frac{1}{2}$ mg/L are not favorable either. The same phenomena were to be seen in the solution of 4-chlorophenoxyacetic acid, although to a minor degree. The effect of the latter substance is again less than that of 2,4-dichlorophenoxyacetic acid, but the difference of effect between these two acids is not so great in this case as for the three species of Nymphaea.

Although both phenoxycompounds appear to be noxious to Limnanthemum, its leafblades showed no abnormalities save the usual hyponastic bending.

Some dozens of petioles of every species were examined as to their cell-lengths. These measurements confirm completely the outcome of the thousands of measurements made in foregoing years, viz. that in the three species of Nymphaea the definite length is reached partly by cell elongation and partly by the formation of new cells, and that in Limnanthemum the elongation is mainly the result of cell division only. In the noxious concentrations of the acids cells of the Nymphaeae were considerably longer than in the optimal concentrations; this is also according to what I have found earlier.

Discussion. ZIMMERMAN and HITCHCOCK have stated the interesting fact that the different phenoxycompounds vary in their effect on different plant species. For example, 4-chlorophenoxyacetic acid modifies the leaves of tobacco (15 *b*, pag. 330); 2,4-dichlorophenoxyacetic acid does not modify tobacco leaves, but greatly modifies the leaves of tomato (l.c., pag. 338). As far as my experiments go we see that 2,4-dichlorophenoxyacetic acid has a much stronger effect than 4-chlorophenoxyacetic acid on all species examined. In another paper (9) HITCHCOCK and ZIMMERMAN report that of several phenoxy- and other compounds tested, only 4-chlorophenoxyacetic acid caused distinct responses in Kalanchoe daigremontiana. I have treated plants of Kalanchoe thyrsiflora with lanoline preparations of both my substances around the stem (8 mg/g). The leaves above the treated region responded by a strong epinasty, and afterwards the stems decayed at their bases. And again these phenomena were much more distinct in the plants treated with 2,4-dichlorophenoxyacetic acid than in those treated with 4-chlorophenoxyacetic acid.

The noxious effect of the phenoxycompounds on Limnanthemum and in higher concentrations on Nymphaea goes parallel to what they do to weeds. But this effect is by no means particular to these substances. In my former experiments I have invariably stated that even the concentrations which may be called optimal as far as the elongation goes, are noxious to the plant because they shorten its life cycle when the treatment is not discontinued in time. It remains to be seen why substances such as α-naphtylacetic acid have a similar and probably much stronger effect on waterplants than 2,4-dichlorophenoxyacetic acid, whilst on higher land plants they are inactive, at least when they are applied in the way in which phenoxycompounds are used.

Summary. 2,4-dichlorophenoxyacetic acid, in concentrations of $\frac{1}{4}$, $\frac{1}{2}$, 1 and 2 mg/L, causes a very strong excess growth in the petioles of Nymphaea odorata, N. terminalis, N. Devoniensis and Limnanthemum nymphaeoides. When immersed in solutions of this substance the petioles can

attain a manifold of their original length within one or a few weeks. The optimal concentration seems to be about ½ mg/L. The daily growth can attain 25 cm, and probably much more at optimal temperatures. 4-chlorophenoxyacetic acid is much less efficient and it makes its influence felt only after a considerable lapse of time.

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Botanical Institute.

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