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INTERACTIVE EFFECTS OF (2-CHLOROETHYL)-TRIMETHYLAMMONIUM CHLORIDE AND PHYTOHORMONES ON GROWTH OF WHEAT SEEDLINGS

F. BEGUM AND M. I. KHAN

Department of Botany, University of Karachi, Karachi-32, Pakistan.

Abstract

The effect of (2-chloroethyl)-trimethylammonium chloride (CCC) in combination with either GA or IAA on the growth of wheat seedlings were studied. Increasing concentration of CCC decreased the shoot length but increased the root length with the exception of 10 ppm CCC where no significant difference as compared to control was observed. The inhibitory effect of CCC on shoot growth was reversed by the addition of either 1.0 ppm. GA or 0.1 ppm IAA solution.

The present study revealed that CCC acted like an "antigibberellin " and an "antiauxin".

Introduction

In the past several years, increasing attention has been paid to the mechanism of (2-chloroethyl)-trimethylammonium chloride (CCC) in dwarfing plants without inducing much injury to the plants (Cathy & Stuart, 1961; Lockhart, 1962; Kuraishi & Muir, 1963; Cleland, 1965). Sachs *et al* (1960) have demonstrated that plant growth retardant acts by inhibiting cell division in the sub-apical meristem zone of the stem, reducing the length of internodes whereas the apical meristem remains unaffected. It has been suggested that CCC inhibits the synthesis of Gibbrellic acid (GA) (Kende *et al*, 1963) and is antagonistic to GA and IAA (Indoleacetic acid) action (Kuraishi & Muir, 1963).

The purpose of the investigation reported here was to examine the interaction of GA and IAA with CCC in altering the growth of wheat seedlings.

Materials and Methods

Triticum aestivum cv. C-591 seeds were surface sterilized with 0.2 per cent $HgCl_2$ solution for 3 min and subsequently washed with sterile distilled water. Seeds were germinated on a wet filter paper in a dark room at 25 ± 2 °C. After 3 days of germination, uniform seedlings were transferred to the plastic pots containing different concentrations of GA, IAA or CCC prepared in 0.2 strength Arnon solution. 200 ml of nutrient solution (with or without treatment) was added in the plastic pots of 300 ml capacity and the nylon net was so adjusted that it just touched the surface of the solution. The pH of the solution was adjusted to 6.0 and the pots were kept at 25 ± 2 °C. Ten hours of 200 ft cdls light was provided with fluorescent white lamps supplemented with incandescent bulbs. Solutions were changed on alternate days.

The increase in shoot and root length was recorded after a period of 6 days. Fresh and dry weight of shoots and roots was also recorded.

Results and Discussion

The first morphological visible response of wheat seedlings treated with different concentrations of CCC was the appearance of dark green leaves broader in diameter than the control.

The interaction of GA and CCC concentrations on the growth of wheat shoots is presented in Fig. 1. A progressive decrease in shoot length of CCC-treated plants was observed. However, CCC-induced inhibition of shoot growth was reversed in the presence of 1 ppm GA solution. Maximum shoot promotion of 130 per cent over control was obtained when the plants were treated with 0.1 ppm CCC and 1 ppm GA. This clearly indicates that the addition of GA not only reversed the growth inhibition but also resulted in promoting the shoot growth. Root growth, as measured by root length, under the influence CCC alone indicated growth promotion, except at 10 ppm where no significant difference over control was observed (Fig. 2). However, when 1 ppm GA was mixed with CCC concentrations (0.01 to 10.0 ppm) further growth promotion resulted. An increase of 140 per cent over control was found at 10 ppm CCC and 1 ppm GA concentrations.

Effect of CCC concentration in the presence or absence of 1 ppm GA on the water content and dry matter of shoots and roots is presented in Table 1. It was noted that CCC when present alone did not disturb the water balance of shoots whereas it did in the presence of 1 ppm GA. When roots were exposed to different concentrations of CCC, they absorbed more water than the un-treated control. Addition of 1 ppm GA further increased the absorption of water. Dry matter of shoots and roots of CCC-treated seedlings, indicated a small increase at 1 and 10 ppm CCC. However, when the seedlings were treated with 1 ppm GA mixed with various levels of CCC (ranging from 0.01 to 10.0 ppm), no significant difference was found at 0.01 and 0.1 ppm CCC, whereas dry weights increased at concentrations higher than these.

Effect of different concentrations of GA (0.01 to 10 ppm) in the presence or absence of 10 ppm CCC on the growth and water content of shoots and roots is presented in Table 2. Fresh and dry weights of shoots and roots increased when the plants were treated with different concentrations of GA. Addition of 10 ppm CCC in GA solutions decreased the water content, fresh and dry weights of shoots and roots as compared to untreated control. In the absence of CCC, a maximum promotion of shoot growth was obtained at 10 ppm GA. In the presence of 10 ppm CCC, GA has comparatively smaller effect on growth with a maximum being reached at 1 ppm. It is apparent from these results that the inhibitory effect of the retardant cannot be

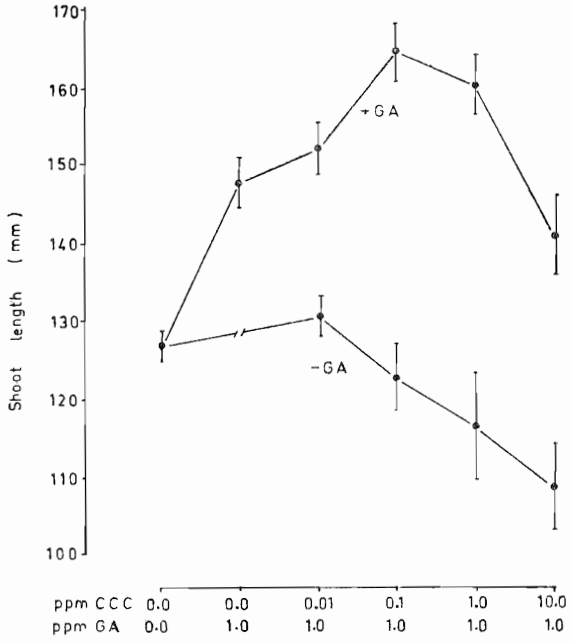

Fig. 1. Effect of CCC and 1 ppm GA on the growth of wheat shoots after 6 days.

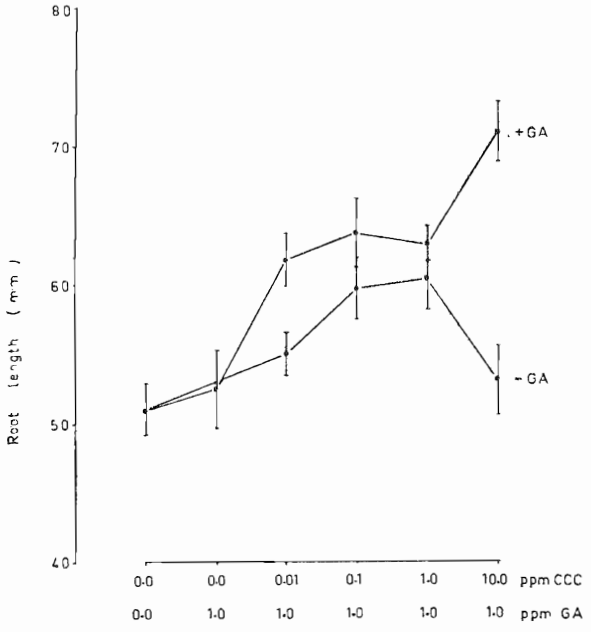

Fig. 2. Effect of CCC and 1 ppm GA on the growth of wheat roots after 6 days.

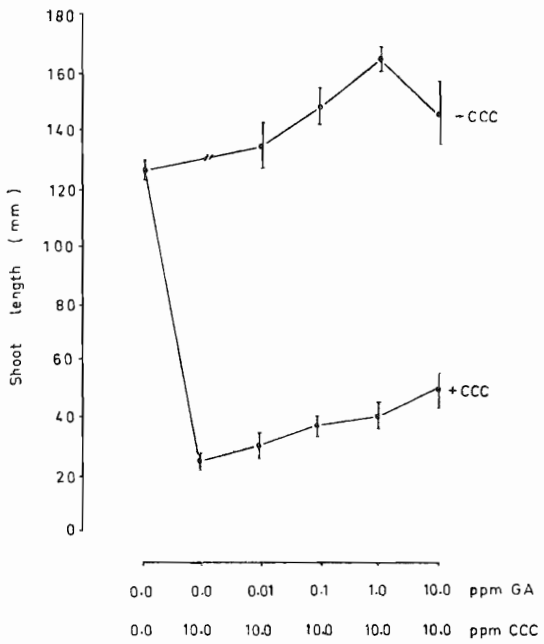

Fig. 3. Effect of GA and 10 ppm CCC on the growth of wheat shoots after 8 days.

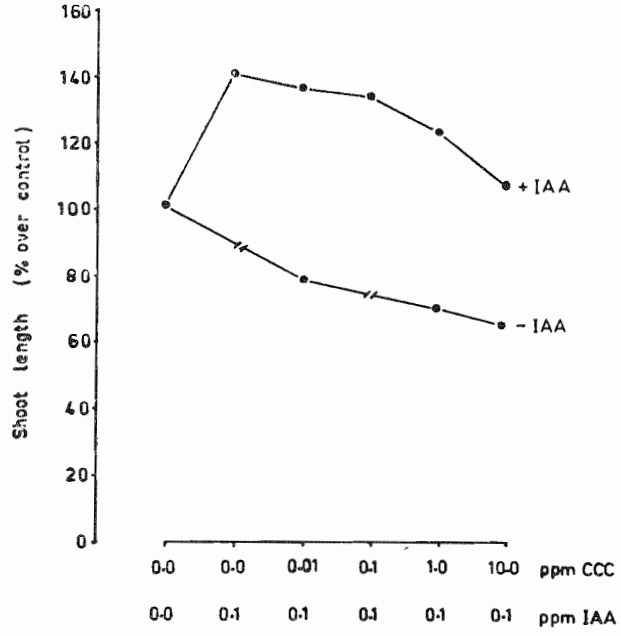

Fig. 4. Effect of CCC and 0.1 ppm IAA on the growth of wheat shoots after 6 days.

Table 1. Interaction of CCC and 1 ppm GA on water content, fresh weight and dry weight of wheat seedlings after 6 days of growth.

Concentration of CC in ppm		— GA		+ GA	
		Shoots	Roots	Shoots	Roots
	Fresh Weight	95.03	17.72	96.33	26.86
	(mg/plant)	+3.9	±2.60	±2.54	±0.72
	Dry weight	7.61	1.15	10.53	1.02
	(mg/plant)	±0.47	±0.35	±1.26	±0.29
0.00	Water content	87.42	16.57	85.80	25.84
(Control)	(mg/plant)				
	Water content	1148.75	1140.86	814.81	2533.33
	(% of D. Wt)				
	μg D. Wt./mg	79.9	—	96.5	—
	F. Wt.	±4.00		±0.35	
	Fresh weight	94.50	30.93	101.43	32.65
	(mg/plant)	±2.00	±2.31	±4.93	±0.84
	Dry weight	8.70	1.27	9.73	1.68
0.01	(mg/plant)	±0.88	±0.47	±0.71	±0.12
	Water content	85.80	29.66	91.70	30.97
	(mg/plant)				
	Water content	986.20	2335.43	942.41	1843.45
	(% of D. Wt.)				
	μg D. Wt./mg F. Wt.	84.40	—	95.70	—
		±3.00		±1.10	
	Fresh weight	97.03	32.86	113.60	43.77
	(mg/plant	±6.0	±2.32	±3.16	±0.65
	Dry weight	8.36	0.95	8.50	1.65
0.1	mg/plant	±0.70	±0.18	±0.58	±0.01
	Water content	88.67	31.91	105.91	42.12
	(mg/plant)				
	Water content	1060.64	3358.94	1236.47	2552.72
	(% of D. Wt.)				
	μg D. Wt./mg F. Wt.	85.80	—	93.30	—
		±1.90		±2.29	

Concentration of CC in ppm		— GA		+ GA	
		Shoots	Roots	Shoots	Roots
	Fresh Weight	103.70	34.01	120.21	49.40
	(mg/plant)	±3.2	±3.22	±4.19	±2.86
	Dry weight	9.06	1.15	11.21	2.20
1.0	(mg/plant)	±0.36	±0.19	±0.50	±0.12
	Water content (mg/plant)	94.64	32.86	109.00	47.20
	Water content (% of D. Wt)	1044.59	2857.39	972.34	2145.45
	µg D. Wt./mg F. Wt	87.30 ±1.10	—	93.10 ±1.20	—
	Fresh Weight	104.91	36.76	119.18	34.36
	(mg/plant)	±0.64	±1.98	±2.55	±3.69
	Dry weight	10.01	1.96	11.06	2.18
	(mg/plant)	±0.19	±0.19	±0.15	±0.01
10.0	Water content (mg/plant)	94.90	34.80	108.12	32.18
	Water content (% of D. Wt)	948.06	1775.51	977.57	1476.14
	µg D. Wt./mg F. Wt.	95.3 2.70	—	92.9 2.1	—

Table 2: Interaction of GA and 10 ppm CCC on water content, fresh weight and dry weight of wheat seedlings after 8 days of growth.

Concentration of GA in ppm		— CCC		+CCC	
		Shoots	Roots	Shoots	Roots
	Fresh weight	72.42	55.00	23.47	8.91
	(mg/plant)	±4.22		±1.68	
	Dry weight	8.05	4.81	3.33	1.13
	(mg/plant)	±0.41		±0.71	
0.00 (Control)	Water content (mg/plant)	64.37	50.19	20.14	7.78
	Water content (% of D. Wt.)	799.62	1043.46	604.80	688.49
	µg D. Wt./mg F. Wt.	111.30	—	140.50	—

Concentration of GA in ppm		— CCC		+ CCC	
		Shoots	Roots	Shoots	Roots
0.01	Fresh Weight (mg/plant)	75.34 ±3.43	55.00	29.54 ±2.09	9.73
	Dry weight (mg/plant)	8.39 ±0.45	4.84	3.75 ±0.47	1.30
	Water content (mg/plant)	66.95	50.16	25.79	8.43
	Water content (% of D. Wt)	797.82	797.97	687.73	648.46
	µg D. Wt./mg F. Wt.	111.20	—	143.90	—
0.1	Fresh Weight (mg/plant)	79.10 ±1.24	60.05	34.48 ±2.09	10.87
	Dry weight (mg/plant)	8.82 ±0.18	4.64	5.50 ±0.43	1.62
	Water content (mg/plan)	70.28	55.41	28.98	9.25
	Water content (% of D. Wt.)	796.82	1194.18	526.90	570.98
	µg D. Wt./mg F. Wt.	111.50	—	160.50	—
1.0	Fresh weight (mg/plant)	82.49 ±4.82	49.47	40.52 ±2.87	8.98
	Dry weight (mg/plant)	8.52 ±0.18	4.19	5.49 ±0.32	1.48
	Water content (mg/plant	73.97	45.28	35.03	7.50
	Water content (% of D. Wt.)	868.19	1080.66	638.06	506.75
	µg D. Wt./mg F. Wt.	103.30	—	163.70	—
10.0	Fresh weight (mg/plant)	88.20 ±3.35	33.66	39.27 ±138	7.60
	Dry weight (mg/plant)	8.72 ±0.19	2.92	4.96 ±0.28	1.29
	Water cnntent (mg/plant)	879.48	31.37	34.31	6.31
	Water content (% of D, Wt.)	911.46	1074.37	691.73	489.53
	µg D. Wt./mg F. Wt.	110.30	—	146.70	—

overcome completely but can be reduced. Similar results were obtained where GA has been found to reduce the inhibotory effect of CCC on the stem growth of tomato (Wittwer & Tolbert, 1960-a), lettuce (Wittwer & Tolbert, 1960-b), leaf sheath growth of wheat (Tolbert, 1960, 1961), mutant maize and the germination of lettuce seeds (Wittwer & Tolbert, 1960-b). Kende *et al* (1963) found CCC and Amo-1618 to supress the biosynthesis of gibberellin in *Fusarium moniliforme* without having any affect on the growth of the mycelium. Lockhart (1961) regarded the growth retardants as antigibberellins in a biological sense. This was later confirmed on the basis of kinetic studies (Lockhart, 1962). From these studies he concluded that the growth retardants exert their influence by decreasing the activity of gibberellins in the growth system.

Halvey (1963) and Kuraishi & Muir (1963) have suggested that the retardation of growth by CCC was due to the lowering of the auxin level and was independent of gibberellin. If this is true, then the CCC-induced inhibition of growth should be partly reversed by the addition of auxin. The result presented in Fig. 3 indicates that CCC-induced inhibition can be reduced by increasing the concentration of GA. CCC-induced growth inhibition of shoot was also reversed by the addition of 0.1 ppm IAA in the growth medium (Fig. 4). Thus GA and IAA can reduce the inhibitory effect of CCC on growth.

From the results reported in this paper it may be concluded that CCC acts like an "antigibberellin" and "antiauxin" as its toxic effect can be reversed by increasing the level of either GA or IAA.

References

Cathy, H.M. and N.W. Stuart. 1961. Comparative plant growth retarding activity of Amo-1618, Phosphon and CCC. Bot. Gaz., **123:** 51—57.

Cleland, R. 1965. Evidence on the side action of growth retardants. Cell Physiol., **6:** 7—15.

Halevy, A. H. 1963. Interaction of growth retarding compounds and gibberellin on indoleacetic acid oxidase of Cucumber seedlings. Plant Physiol., **38:** 731—737.

Kende, H., H. Ninnemann and A. Lang. 1963. Inhibition of gibberellic acid biosynthesis in *Fusarium moniliforme* by Amo-1618 and CCC. Naturwiss, **50:** 599—600.

Kuraishi, S. and R.H. Muir. 1963. Mode of action of growth retarding chemicals. Plant Physiol., **38:** 19—24.

Lockhart, J.A. 1961. Physiological mechanism of action of stem growth inhibitors. Plant Physiol., **36:** (Supt.) 38.

———. 1962. Kinetic studies of certain antigibberellins. Plant Physiol., **37:** 759—764.

Sachs, R.M., A. Lang, C.F. Brilz and J. Roach. 1960. Shoot histogenesis: subapical meristematic activity in a caulescent plant and the action of gibberellic acid and Amo-1618. Amer. J. Bot., **47:** 260—266.

Tolbert, N.E. 1960. (2-Chloroethyl)-trimethylammonium chloride and related compounds as plant growth substances. II. Effects on growth of wheat. Plant Physiol., **35:** 380—385.

———. 1961. Structural relationship among chemicals which act like antigibberellin. In Gibberellin, Ad. in chemical Ser. pp. 145-151. Amer. Chem. Society Press, Washington, D.C.

Wittwer, S.H. and N.E. Tolbert. 1960-a. (2-chloroethyl)-trimethylammonium chloride and related compounds as plant growth substances. III. Effect on growth and flowering of the tomato. Amer. J. Bot., **47:** 560—565.

———. 1960-b. (2-Chloroethyl)-trimethylammonium chloride and related compounds as plant growth substances. V. Growth, flowering and fruiting response as related to those induced by auxin and gibberellin. Plant Physiol., **35:** 871—877.