Control of shoot growth in *Agrostemma githago* L. by 4,4-dimethylmorpholinium chloride and related compounds

J. S. KNYPL

Plant Growth Substances Laboratory, University of Łódź
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### ABSTRACT

4,4-Dimethylmorpholinium chloride and (2-chloroethyl)-trimethylammonium chloride retarded shoot growth in *Agrostemma githago* L. This effect was nullified by gibberellin. 4,4-Dimethyl-2-oxomorpholinium chloride and 4,4-diethyl-2-oxomorpholinium chloride slightly stimulated shoot growth.

# INTRODUCTION

Of a number of dialkylmorpholinium derivatives (Witek et al., 1967) the most active as plant growth retardants were found DMMC <sup>1</sup> (Jung, 1970; Krawiec 1973; Borkowski, 1976), DMOMC and DEOMC (Krawiec et al., 1973). DMMC affects growth of a wide range of plant species, and the symptoms of its action resemble those produced by CCC (Knypl et al., 1976). It has been suggested that mechanisms of action of both compounds might be similar, *i.e.* they might block gibberellin biosynthesis and reduce the level of active auxin in the treated plants (Knypl, 1977a). DMOMC and DEOMC are more selective in respect to the range of sensitive plants. The compounds possibly block biosynthesis of some gibberellin fractions, indirectly affecting the auxin-controlled phenomena (Knypl, 1977b).

Comparative analysis of the effects of the compounds in question on shoot growth in Agrostemma was the aim of this study.

<sup>&</sup>lt;sup>1</sup> Abbreviations: DMMC, 4,4-dimethylmorpholinium chloride; DMOMC, 4,4-dimethyl-2-oxomorpholinium chloride; DEOMC, 4,4-diethyl-2-oxomorpholinium chloride; CCC, (2-chloroethyl)trimethylammonium chloride; GA<sub>3</sub>, gibberellic acid.

### MATERIAL and METHODS

After-ripened seeds of *Agrostemma githago* L. cv. Greifswald were germinated on wet filter paper for 2 days in darkness at 25°C. Pregerminated seeds were sown at a depth of 2 cm in a mixture of quartz and and peat moss (1:1) in 8-cm pots. Peat moss was a commercial product ST-K-2 (Zakłady Przemysłu Torfowego, Szczecinek) supplemented with macro- and microelements, pH 5—6. The plants were cultivated in a growth chamber at 22—25°C under a natural photoperiod (June-September).

6-day-old seedlings (hypocotyl length 24—28 mm) were irrigated with 20 ml of 1 mM or 10 mM solution of DMMC, DMOMC, DEOMC or CCC. GA $_3$  (10  $\mu g$  in a 5  $\mu l$  volume) was applied to the apex at the same time. The treatments were repeated after 7 days. The length of hypocotyl and epicotyl internodes was measured each week for 5 weeks. Epicotyl internodes were numbered consecutively starting from the cotyledonary node.

#### RESULTS

DMMC and CCC reduced epicotyl growth as measured a week after the 2nd treatment. The growth of the epicotyl was not affected

Table 1

Comparative effects of DMMC, DMOMC, DEOMC, CCC and GA<sub>3</sub> on growth of Agrostemma githago L.

1) 6-day-old seedlings (mean hypocotyl length 26 mm) were irrigated twice with 20 ml of each growth retardant, or treated with 10 µg of GA<sub>3</sub> at a week interval. 2) Hypocotyl length at the time of 1st treatment was subtracted from the shoot length. Shoot=hypocotyl and epicotyl. 3) Data followed by different postscripts within each column differ significantly at a 5% probability level. 4) Control = untreated plants.

Treatment <sup>1</sup> )	Net elongation <sup>2,3)</sup> (mm)				
	20-day-old plants		41-day-old plants		
	Shoot	Epicotyl	Shoot	Epicotyl internode	
				II	Ш
Control <sup>4</sup>	40a	12a	152a	20a	45a
DMMC (1 mM)	43a	9b	132b	14b	38b
DMMC (10 mM)	38a	8b	117c	9c	30c
DMOMC (1 mM)	39a	15c	163e	27d	50a
DMOMC (10 mM)	38a	12a	156a,e	22a	44a
DEOMC (1 mM)	39a	11a	160e	26d	49a
DEOMC (10 mM)	37a	11a	156a,e	21a	47a
CCC (1 mM)	39a	8b	112c	9c	30c
CCC (10 mM)	38a	8b	96d	8c	26c
GA $_3(2\times10~\mu g)$	40a	26d	200f	29d	44a

by DEOMC and significantly stimulated by a lower dosage of DMOMC. After 4 weeks the retarding effect of DMMC and CCC on the epicotyl growth was noticeable (Table 1). Shoots of the treated plants were thicker, and the leaves had a darker green pigmentation as compared with the control plants. The stimulatory effect of DMOMC on growth still persisted. Growth of the 2nd internode was stimulated by DEOMC.

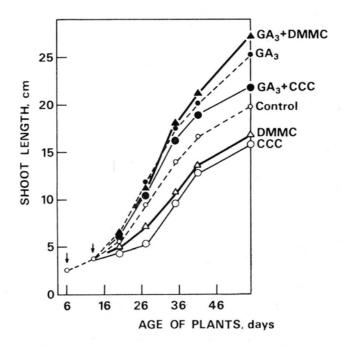


Fig. 1. Reversal by gibberellin of retarding effect of DMMC and CCC on shoot growth in Agrostemma githago.

6-day-old seedlings were twice irrigated with 20 ml of 10 mM solution of DMMC or CCC, or treated with 10  $\mu g$  of GA<sub>3</sub> (cf. Table 1). Application of growth regulators is indicated by arrows.

DMMC was selected for further experiments on the possible physiological interaction with gibberellin (cf. Knypl and Janas, 1977).  $GA_3$  completely reversed the growth-retarding effect of DMMC, and markedly reduced that of CCC (Fig. 1).  $GA_3$  nullified the growth-retarding effect of CCC when the latter compound was applied in a lower dosage of  $2 \times 20$  ml of 1 mM solution (data not shown).

The general appearance of young Agrostemma plants treated with the growth retardants alone and in combination with  $GA_3$  is presented in Fig. 2. The compounds in question neither affected flowering of the plants nor the yield of seeds (data not shown).



Fig. 2. Retarding effect of DMMC and CCC on growth of A. githago is reversed by  $GA_3$ .

41-day-old plants are presented in the photo; other details as in Fig. 1. a — Control (untreated plants); b — GA3; c — DMMC; d — DMMC+GA3; e — CCC; f — CCC+GA3

## DISCUSSION

The degree of the retarding effect of DMMC on shoot growth in Agrostemma was dosage dependent, whereas that of CCC was rather dosage-independent. It seems that  $2\times 20$  ml of 1 mM solution of CCC represented an almost saturating dosage of this retardant in the experiments reported here. Per a molar concentration basis DMMC was found to be around 5 times less efficient growth retardant than CCC in Spirodela oligorrhiza (Knypl et al., 1976). The same seems true for Agrostemma. Since GA<sub>3</sub> nullified the effect of DMMC on growth, it can be suggested that the compound acts as an inhibitor of gibberellin biosynthesis (cf. Lang, 1970; Knypl, 1977; Knypl and Janas, 1977).

Although DMOMC and DEOMC are structurally related to DMMC, they did not retard growth in Agrostemma. Marked differences in physiological activity between DMMC on the one hand, and DMOMC and DEOMC, on the other, seem to be due to the fact that the heterocyclic ring of the latter compounds undergoes spontaneous hydrolysis to form betaine derivatives (Knypl, 1979). Spontaneous formation of betains explains the phenomenon of a greater biological selectivity of DMOMC and DEOMC as compared with DMMC (Knypl, 1977b). The moderate stimulation of epicotyl growth by DMOMC and DEOMC in Agrostemma (Table 1) is in agreement with reports that the compounds enhanced bulb enlargement in onion (Knypl, 1979) and increased yield of green matter in alfalfa (Michałowski et al., 1977), grasses (Zebrowski et al., 1973) and other crops (Witek et al., 1973). Pre-sowing soaking of seeds in solutions of both DEOMC and DMOMC stimulated subsequent growth of seedlings in many leguminous plant species (Krawiec, 1973). The growth stimulatory effects of dialkylmorpholinia are secondary as it seems; nevertheless, they are worth a more detailed study.

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Author's address: Prof. dr hab. J. S. Knypl Plant Growth Substances Laboratory, University of Łódź, Banacha 12/16; 90-237 Łódź; Poland

Kontrola wzrostu łodygi Agrostemma githago L. przez chlorek 4,4-dwumetylomorfoliniowy i substancje pokrewne

### Streszczenie

Chlorek 4,4-dwumetylomorfoliniowy i chlorek (2-chloroetylo)-trójmetyloamoniowy hamują wzrost łodygi kąkolu, przy czym efekt ten niweluje giberelina. Chlorek 4,4-dwumetylo-2-oksomorfoliniowy i chlorek 4,4-dwumetylo-2-oksomorfoliniowy nieznacznie pobudzają wzrost łodygi.