## A New Ouaternary Ammonium Compound with Plant Growth Regulatory Activity

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Summary. Of several quaternary ammonium derivatives tested, 1-allyl-1-(3,7-dimethyloctyl)piperidinium bromide showed the best growth-retarding activity. On Chrysanthemum morifolium the growth retardation obtained with a single application of 100–200 ppm compared favourably with that obtained from 2–3 applications of 3400 ppm of N-dimethylaminosuccinamic acid (daminozide).

Growth-retarding activity of quaternary ammonium compounds have been reported by Tolbert<sup>2</sup>, Cathey<sup>3</sup> and Buchenauer and Erwin<sup>4</sup>. To elucidate the plant growth regulatory activity of quaternary ammonium compounds containing a terpenoid moiety, a number of compounds were synthesized by conventional methods and tested in our laboratories.

The growth retardant activities of 7 compounds, selected out of a large number of quaternary ammonium deriv-

atives, were investigated on *Chrysanthemum morifolium* Ramat cv. Princess Ann. The plants which were propagated and grown in the greenhouse were sprayed to run-off when the sideshoots were 2–4 cm long. The

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- <sup>2</sup> N. E. Tolbert, J. biol. Chem. 235, 457 (1960).
- <sup>3</sup> H. M. CATHEY, A. Rev. Plant Physiol. 15, 273 (1964).
- <sup>4</sup> H. Buchenauer and D. C. Erwin, Z. PflKrankh. 9, 567 (1973).

Table I. Growth retardation activity of quaternary ammonium compounds on Chrysanthenum morifolium cv. Princess Ann



Substituent					Length (cm)		Growth reduction relative to control (%)		
R <sub>1</sub>	$R_2$	R <sub>3</sub>	X	4000	ppm 1000	4000	ppm	1000	
Untreated				20.2		0			
Dimethyl-amino succinamic acid (daminozide)				15.6		22.8			
Allyl	3,7-dimethyloctyl	CH,	Br	11.7	13.8	42.1		31.8	
Allyl	3,7-dimethyloctyl	CH,	Cl	12.3	14	39.2		31.7	
Propyl	3,7-dimethyloctyl	$CH_2$	J	12.3	16.2	39.2		19.8	
Allyl	3,7-dimethylnonyl	$CH_2$	$_{ m Br}$	13.6	16.2	32.8		19.8	
Ethyl	3,7-dimethyloctyl	$CH_2$	$\operatorname{Br}$	13.8	15.9	31.8		21.2	
Allyl	3,7-dimethyloctyl	0	$\operatorname{Br}$	15.7	20.2	22.3		0	
Allyl	3,7-dimethyl-2,6-octadienyl	$CH_2$	$_{ m Br}$	20.2	20.1	0		0.5	

p 0.05; SE ± 3.63.

Table II. Growth regulatory activity of 1-allyl-1-(3,7-dimethyloctyl)piperidinium bromide on 6 species in the greenhouse

Testplants	Treatment + ppm	Length (cm)	Per plant	Standard Error (±		
			Number of ripe fruits	Latex (mg)		
Vitis vinițera	Untreated	44			2.07	
Lycopersicon esculentum	1000 Untreated 4000	19.5	117 178.5		13.92	
Euphorbia pulcherrima	Untreated 6000	39.6 28	178.5		1.94	
Pachistachys lutea	Untreated 1000	20 15			0.96	
Brassica napus	Untreated 1500	82.6 54.8			4.55	
Ficus elastica	Untreated 5000			64.6 127.2	23.33	

water-soluble compounds were dissolved in water, insoluble compounds were formulated as wettable prowders. The concentrations used were 4000 and 1000 ppm of the active ingredients.

Of the 7 compounds tested, 4-allyl-4-(3, 7-dimethyloctyl) morpholinium bromide and 1-allyl-1-(3,7-dimethyl-2,6octadienyl)piperidinium bromide demonstrated little or no growth retardant effect (Table I), whereas 1-allyl-1-(3, 7-dimethyloctyl)piperidinium bromide, the corresponding chloride, and 1-propyl-1-(3,7-dimethyloctyl)piperidinium iodide showed excellent growth retardant activities. The 1-allyl-1-(3,7-dimethylnonyl)piperidinium bromide and the 1-ethyl-1-(3,7-dimethyloctyl)piperidinium bromide were slightly less active. The growth retardation effect could be antagonized by indole-3-acetic acid and gibberellic acid.

As the 1-allyl-1-(3,7-dimethyloctyl)piperidinium bromide (ISO approved common name: piproctanylium bromide) was the most active of the 7 compounds, its activity was further investigated in the greenhouse on 6 additional species: Vitis vinifera L. cv. Riesling × Sylvaner, Euphorbia pulcherrima Wild. cv. Paul Mikkelsen, Pachistachys lutea Nes. and Brassica napus L. cv. Rapol. Heights were recorded 4 weeks after treatment, except with Euphorbia pulcherrima where the assessment was carried out 10 weeks after application. The effect on fruit ripening was tested on Lycopersicon esculentum cv. Tiny Tim. The plants were sprayed when the first fruits turned red. The number of ripe fruits was counted 3 weeks later. The latex-flow stimulation activity was investigated on Ficus elastica Roxb.

The wide range of plant growth regulatory activity of piproctanyliumbromide is demonstrated in Table II. Growth retardant effects could be observed on Vitis vinifera, Euphorbia pulcherrima, Pachistachys lutea and Brassica napus. The compound accelerated fruit ripening on Lycopersicon esculentum and stimulated latex-flow on Ficus elastica. Both these effects were reported as being associated with the plant hormone ethylene by Wang et al.5.

Of the quaternary ammonium derivatives which have been synthesized and tested in our laboratories, a number of compounds have shown interesting plant growth responses, but so far 1-allyl-1-(3,7-dimethyloctyl)piperidinium bromide is the most promising. Therefore large scale greenhouse trials were initiated in commercial nurseries, especially with pot varieties of Chrysanthemum morifolium. The results obtained with a single application of 100-200 ppm compared favourably with those obtained from 2-3 applications of 3400 ppm of daminozide. There fore piproctanylium bromide will shortly be made available, as ALDEN, for this use. Other uses, based on the results presented above, are currently under investigation.

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## Adenovirus Type 12 Infection of Defined Mouse-Human Hybrid Cell Clones

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Summary. Human adenovirus type 12 does not multiply in mouse cells; only viral T-antigen is detected. Mousehuman cell hybrid clones containing human chromosomes A3, B5, C7, C11, C12, D14, E17, F19 and F20, support synthesis of adenovirus DNA and capsid antigens.

Recent advances in mammalian somatic cell hybridization have allowed the analysis of the mechanisms of host cell restriction to viral infections. The infection of mousehuman cell hybrids with poliovirus has demonstrated that the permissiveness for virus infection can be associated with the human chromosome F19 coding for a cell surface receptor1.

Oncogenic adenovirus type 12 multiplies in human cells but not in hamster or mouse cells. In hamster cells, an abortive cycle is induced. The infected cells synthesize T-antigen<sup>2</sup> and adenovirus-specific mRNA is transcribed, but synthesis of viral DNA, late mRNA, or viral capsid proteins cannot be detected<sup>3</sup>. When heterokaryocytes of hamster and human cells were infected with Ad12, the synthesis of Ad12 DNA and late viral capsid proteins was demonstrated in nuclei of hamster origin that had been nonpermissive prior to cell fusion<sup>4</sup>. Mouse cells have been reported to be totally nonpermissive to adenovirus type 12 infection and unable to support synthesis of either T-antigen or viral capsid proteins 5.

We decided to study the infection of specific clones of mouse-human cell hybrids to determine whether the presence of certain human chromosomes in hybrid cells would make them permissive for adenovirus replication. Contrary to previous reports we observed that when mouse cells (3T3 or L) are infected with adenovirus type 12, the adenovirus-specific T-antigen can be demonstrated in the nuclei of infected cells (Figure 1). No virus-specific DNA and late viral capsid proteins were detected, but the infected cells were killed by the infection (Figure 2). The fraction of cells killed was in good correlation with the frequency of T-antigen positive cells. As in the hamster cell system2, when stationary 3T3 cells were infected, adenovirus type 12 induced a round of cellular DNA synthesis (Table I).

Hybrid cells were made by hybridization of C1-1D mouse cells deficient in thymidine kinase to either KOP-2 human fibroblasts<sup>6</sup> or a line of SV40-transformed human cells derived from patients with the Lesch-Nyhan syndrome7. Mass cultures of hybrid cells were cloned on

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