

oxygen consumption of 0.32 mg/l in a period of 5 days. From this, it can be calculated that about 10% of the organic carbon had been converted to CO<sub>2</sub> (tertiary biodegradation). However, chemical analyses of the incubated samples indicated that 51% of the product had disappeared and thus undergone primary biodegradation.

In the aerobic MM-test, the pesticide is dissolved in a mineral medium inoculated with soil and incubated at room temperature on a rotary shaker (cf. the screening test of the OCDE-method for biodegradability testing of detergents). A rapid disappearance of Tioctilate was observed (see table). It should be added that the sterile control A was incubated in the dark, while the sterile control B was incubated in the light. Hence, Tioctilate is not very susceptible to photolysis. In the anaerobic MM-test, the samples are incubated under strictly anaerobic conditions. Under these conditions, only about 40% of the product disappeared in a period of 20 days.

The aerobic OM-test corresponds to the standard activated sludge test for detergents<sup>4,6</sup>. A rapid and complete (99.8%) biodegradation of Tioctilate was observed, even without a preliminary adaptation of the sludge system. No apparent disturbance of the microbial community could be detected upon introduction of the product to the activated sludge bassins. For the anaerobic OM-test, a disappearance of up to 80% was noted after 5 days, while 90% of the product was biodegraded after 20 days.

**Discussion and conclusions.** The various experiments indicate that Tioctilate is quite susceptible to microbial metabolism provided aerobic conditions prevail. Hence, this product should rapidly disappear from oxygen-rich envi-

ronments such as aerobic waste treatment systems and surface waters and soils. However, it must be pointed out that the results of the die-away tests reflect the primary biodegradation of the compound. The BOD-test, which indicates the complete conversion of the compound to carbon dioxide and water, suggests that the tertiary biodegradation of Tioctilate proceeds rather slowly. Nevertheless, in view of the nonxenobiotic character of the immediate metabolites of Tioctilate (i.e. benzoic acid and octanethiol), one can expect a normal albeit slower mineralization of these metabolites in aerobic environments.

The results of the anaerobic tests reveal that, provided organic matter is present, Tioctilate is metabolized fairly rapidly by the bacteria. In oligotrophic anaerobic environments, however, the biodegradation proceeds rather poorly. These results indicate that in the most important anaerobic habitats such as septic tanks, methane digesters, sewers and sludge-waters interphases of rivers, a fairly rapid disappearance of Tioctilate will occur.

- 1 Seresci, 46, av. Jean-Jaurès, 1030 Bruxelles.
- 2 W. Verstraete and E. Van Vaerenbergh, *Tijdschr. Becewa* 4, 50 (1977).
- 3 J.P. Voets, P. Pipyn, P. Van Lancker and W. Verstraete, *J. appl. Bact.* 40, 67 (1976).
- 4 OCDE, Pollution par les détergents, Publications de l'OCDE, 2, rue André-Pascal, Paris 16<sup>e</sup>, France.
- 5 American Public Health Association, Standard Methods for the examination of water and wastewater, 13th ed. New York.
- 6 W.K. Fisher, P. Gerike and W. Holtmann, *Water Res.* 9, 1131 (1975).

## Conjugated terpenoid ketones: A new group of plant growth regulators<sup>1</sup>

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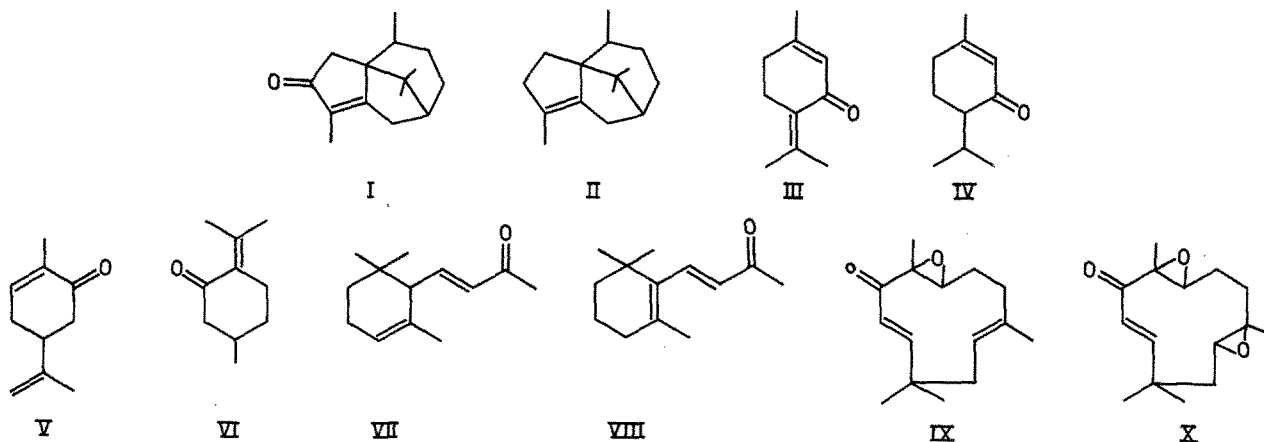
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**Summary.**  $\alpha,\beta$ -unsaturated terpenoid ketones have a root-inducing property on the hypocotyl cuttings of *Phaseolus aureus*. Significantly isopatchoulone (I) is distinctly more active in causing rooting over IAA.

Terpenoid lactones are emerging as a new group of plant growth regulators. The biological activity of these natural products is associated with the exomethylene group in conjugation with the lactone carbonyl, and this structural feature is almost indispensable<sup>2</sup> for this action. An earlier communication<sup>3</sup> from our laboratory showed that some

terpenoid  $\gamma$ -lactones, in which a cyclopropane or a trisubstituted double bond was in conjugation with the lactone carbonyl, are more active than  $\alpha$ -methylene- $\gamma$ -lactones.

Recently<sup>4</sup> terpenoids with a cross conjugated ketone moiety have been shown to cause adventitious root formation in the hypocotyl of mung bean cuttings. In order to obtain



Effect of terpenoids on the root formation of *Phaseolus aureus*

		Concentration (ppm)				
		5 ppm	10 ppm	15 ppm	20 ppm	25 ppm
		Number of roots				
Isopatchoulene	(I)	6.0±1.2	26.8±1.4	33.7±1.3	43.0±1.8	54.6±1.4
Cyperene	(II)	5.6±1.4	6.4±1.2	7.2±1.2	6.8±1.4	7.3±1.1
Piperitenone	(III)	21.0±1.7	34.6±1.1	17.0±1.4	23.5±1.5	28.5±1.8
Piperitone	(IV)	31.2±1.3	28.7±1.4	27.5±1.5	20.5±1.3	18.2±1.3
Carvone	(V)	5.6±1.7	6.6±1.3	7.8±1.3	5.6±1.5	6.4±1.2
Pulegone	(VI)	15.6±1.2	17.0±1.3	23.8±1.1	17.0±1.3	14.6±1.6
$\alpha$ -Ionone	(VII)	11.0±1.3	9.7±1.6	10.0±1.7	11.2±1.3	13.5±1.2
$\beta$ -Ionone	(VIII)	6.6±1.2	25.2±1.3	24.0±1.4	21.6±1.3	15.5±1.5

Mung bean rooting tests were performed in the laboratory using the basic methodology of Hess<sup>6</sup>. IAA was dissolved in minimum quantity of ethyl alcohol and the dilutions were made with distilled water to give a standard ppm solution for control reference. Control experiments: H<sub>2</sub>O: 7.6±1.4; IAA (10 ppm): 16.7±1.8.

structure-activity data a large scale screening of essential oils and their component terpenoids was undertaken in this laboratory for their evaluation as plant growth regulators.

The essential oil from the tubers of *Cyperus scariousus* significantly induced the formation of roots on the stem cuttings of *Phaseolus aureus*. A major identified terpenoid constituent of this oil is the crystalline  $\alpha,\beta$ -unsaturated ketone isopatchoulene<sup>5</sup> (I). Our earlier findings<sup>4</sup>, that structurally related cross-conjugated terpenoid ketones stimulate root formation, suggested that the biological activity of the essential oil may be due to I.

These data prompted us to evaluate  $\alpha,\beta$ -unsaturated terpenoid ketones for regulating growth in plants. The work reported in this paper shows that, with the glaring exception of carvone (V),  $\alpha,\beta$ -unsaturated ketones cause adventitious root formation in the hypocotyl cuttings of *P. aureus*. At suitable concentrations (table), several conjugated ketones were found to be more potent to cause root formation over IAA. Significantly isopatchoulene (I) in concentrations above 10 ppm was more prominent in displaying this effect.

This is to our knowledge the first demonstration of root-forming ability of  $\alpha,\beta$ -unsaturated terpenoid ketones. To confirm that the plant growth activity in I is due to  $\alpha,\beta$ -unsaturated ketone moiety was revealed when the root promoting activity fell off in cyperene (II), which is the

parent hydrocarbon of I, and is present in the same essential oil as another major component.

A comparison (table) of stimulation of rooting by I with IAA and other conjugated ketones suggests that the extent and position of substitution of enone chromophore, together with the endo- or exo-position of the double bond in the ring, may be the deciding factor for this action. Interestingly like<sup>4</sup> (IX and X)  $\alpha$ -ionone (VII) with a disubstituted double bond is only slightly active over control experiments. With extended conjugation its isomer  $\beta$ -ionone (VIII) regains the root-inducing potential which is more than IAA at 10 ppm.

- 1 Acknowledgment. The work was financially supported by the Punjab State Government, India, under the scheme 'Chemistry of some natural products and their significance in agriculture'. The authors thank 'Dragoco', Holzminden, Federal Republic of Germany, for supplying the samples of monoterpenes.
- 2 H. Shibaoka, M. Shimokoriyama, S. Iriuchijima and S. Tamura, *Pl. Cell Physiol.* 8, 297 (1967).
- 3 P.S. Kalsi, V.K. Vij, O.S. Singh and M.S. Wadia, *Phytochemistry* 16, 784 (1977).
- 4 P.S. Kalsi, O.S. Singh and B.R. Chhabra, *Phytochemistry* 17, 576 (1978).
- 5 S.B. Nerali, P.S. Kalsi, K.K. Chakravarti and S.C. Bhattacharyya, *Tetrahedron Lett.* 45, 4053 (1965).
- 6 C.E. Hess, *Hormolog* 33 (1961).

Absorption and translocation of tetrachlorodibenzo-p-dioxine by plants from polluted soil<sup>1</sup>

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**Summary.** Measurements were made of contamination of plants grown in soil polluted with tetrachlorodibenzo-p-dioxine. Findings show that the pollutant is absorbed and translocated by the plants studied and suggest that the pollutant may be eliminated in the course of time.

After the explosion of the trichlorophenol reactor at Seveso in northern Italy<sup>2</sup> which caused an escape of tetrachlorodibenzo-p-dioxine (hereafter referred to as TCDD dioxine), which is a toxic (teratogenic and acnegenic<sup>3,4</sup>) substance, a knowledge of intake, accumulation and metabolism of TCDD by higher plant life came to be of vital importance. The possible ability of plants to take in and accumulate dioxine might represent a danger in spreading pollution; furthermore, their ability, if any, to take in, accumulate and eliminate dioxine might contribute to resolving the problems of decontamination; it might, for example, be the only solution to the problem of reclaiming extremely extensive areas of land with a low level of pollution.

Isensee and Jones<sup>5</sup> have experimented with oat and soya plants, showing that dioxine does not appear to interfere with growth of the plant to maturity, and that it is found in limited quantities in the tissues and does not appear to accumulate. Their studies also reveal that TCDD concentration in plants appears to reach a maximum level and then to decrease with time, particularly in the aerial part of the plant. Isensee and Jones suggest a removal by metabolism, volatilization from the tissues or backward transport to the roots.

The possibility that plant life may in some way degrade dioxine has been suggested by US Air Force research workers, who have shown that, when plants are treated with