

by 32H1 was recovered as NH_4^+ from the cell supernatant¹⁵. Preliminary results indicate that, in our system also, NH_4^+ was excreted into the medium, transported to the plant callus and used for the synthesis of nitrogen-containing compounds.

One of the prerequisites for a quasi-symbiotic situation, the transfer of fixed N from rhizobia to plant cells in an in-

vitro association, is suggested by our results. Further investigations concerning the physical nature of the transferred fixed N and the utilization of this compound by the plant cells can now be made. We consider that these approaches can provide information about the possible practical use and economic importance of in-vitro associations between non-leguminous plants and rhizobia.

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Insecticidal effect of trans-2-nonenal, a constituent of carrot root

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Summary. Vapour of trans-2-nonenal killed the carrot fly larva, *Psila rosae*, with an LC_{50} of 2.17 mg/24 h. The aldehyde was identified in the essential oil of carrot in concentrations of up to 40 μg /root.

Plants use chemicals in their defence against insects with effects that may be non-lethal such as repelling insects or inhibiting their growth³ or lethal as caused, for example, by the pyrethrins⁴. A systematic investigation of the effects of compounds associated with the carrot root (*Daucus carota* L.) on the larva of its parasite, the carrot fly (*Psila rosae* (F.)) (Diptera: Psilidae), has identified a naturally-occurring toxin, trans-2-nonenal $\text{CH}_3 \cdot (\text{CH}_2)_7\text{CH}:\text{CH} \cdot \text{CHO}$.

The following amounts of trans-2-nonenal (purity 99% GC; Bush Boake and Allen Ltd, London) dissolved in chloroform (Merck analytical grade) were each pipetted onto filter paper discs (Whatman No. 4, 5 cm diameter): 0.05, 0.1, 0.5, 1.0, 2.5, 4.0, 5.0, 7.5, 10.0 and 15.0 mg (4 replicates of each); control discs received only solvent. After a 3-min interval to evaporate solvent, each disc was placed in a 400 cm^3 Kilner jar which was then sealed by a paraffin-coated glass plate. After 5 min a batch of 5 3rd-instar larvae, still active after dissection from carrot roots and subsequent starvation for 24 h at 19 °C, was introduced into each jar in a stainless steel tray containing moist vermiculite (70 ml distilled water/60 g vermiculite); each tray was placed on glass vials to provide a clearance of 1.5 cm above the paper disc. These conditions formed a rather stringent test of the compound as direct contact with the insects was not allowed. Tests lasted 24 h and the time taken to immobilize 50% of larvae per treatment was noted; dead larvae were recorded following a 4-h exposure to fresh air. These were shrivelled and had a browned cuticle.

2-nonenal has been previously identified in both steam-distilled⁵ and cold-solvent⁶ extracts of carrot root and to supplement these observations, steam-distilled extracts were made from 4 carrot cultivars, Chantenay-Red-Cored Elite, Regulus Imperial, Danro and Regol, for analysis by GLC.

Vapour from 15 mg of trans-2-nonenal induced 100% larval mortality in 24 h, 0.05 mg induced none and the LC_{50} was 2.17 (figure 1). 50% of larvae were immobilized in 15 min by 15 mg, in 30 min by 2.5 and in more than 4 h by 1 mg; larvae were neither immobilized nor killed by controls. Trans-2-nonenal was identified in the oils of the 4 carrot cultivars, representing 0.2 ppm Chantenay-Red-Cored Elite, 0.2 ppm Regulus Imperial, 0.5 ppm Danro and 0.6 ppm Regol (figure 2); the latter value is equivalent to 40 μg of trans-2-nonenal per average carrot root. Such GC identification is not definitive but there was insufficient of

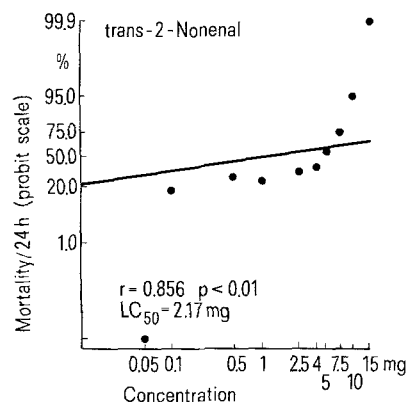


Fig. 1. Dosage/mortality relationship for 24 h exposure of the 3rd-instar carrot fly larva to trans-2-nonenal. As cis-2-nonenal is not commercially available the stereospecificity of this effect could not be ascertained.

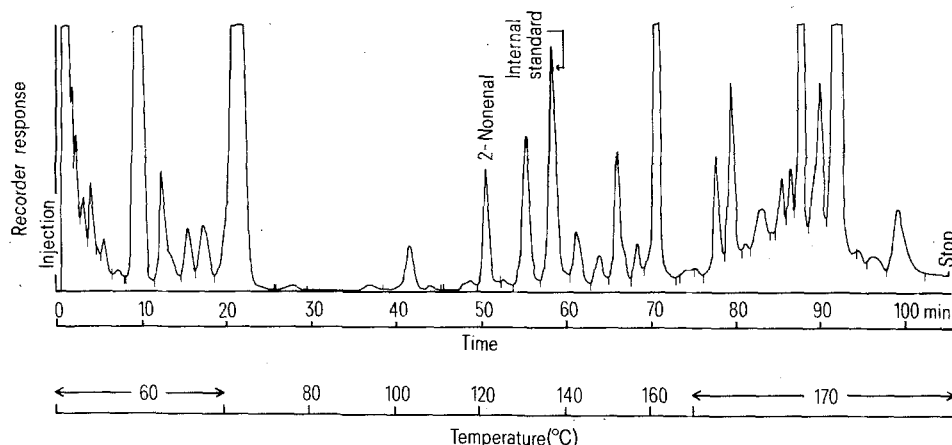


Fig. 2. Identification of 2-nonenal (retention time 50.57 min) by GLC of the atmospheric steam distilled oil of the carrot cultivar Regol. Operating conditions: instrument, Hewlett Packard 5750G equipped with automatic computing integrator 3380A; column, glass 3 m×4 mm inner diameter; packing, 7% Carbowax 20 M; injection port temperature, isothermal at 60°C for 20 min followed by a programmed 2°C/min, rise to 170°C. Identification is by comparison with the retention time of trans-2-nonenal and quantification is by reference to known amounts of menthol using the internal standard method.

the compound in the oil for collection and confirmation by other methods of its isomeric form. Nevertheless, taken in conjunction with the earlier reports^{5,6} it very strongly suggests that 2-nonenal is present in the oil and the trans form is most likely due to its greater stability.

This is not the only indication of a protective role for 2-nonenal, as it has been identified in the defensive secretion

of the tenebrionid beetle *Eleodes beameri* (Blais)⁷, but it seems the first demonstration of it as an insecticide.

The existence of a plant-associated compound with such a profound effect on its parasite is consistent with current views on the co-evolution of plants and insects³ and investigation of its field efficacy and mode of action against *P. rosae* would seem appropriate.

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Physico-chemical properties of South American iguanid albumins

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Summary. Except for a markedly reduced anodal electrophoretic mobility, the serum albumin of the Galapagos marine iguana was physico-chemically identical to that of terrestrial iguanids. Reduction in albumin net charge may have facilitated the adaptation of this species to a semi-aquatic environment.

Among the Reptilia, plasma osmotic pressures directly relate to albumin concentration¹. Both the charge and plasma levels of this protein vary throughout the range of reptilian species, apparently to accommodate specific environmental conditions¹. This adaptability appears to have been a major factor in the shaping of reptilian evolution.

Serological differences exist among the serum albumins of several closely related genera of Galapagos land and marine iguanas². Since these reptiles occupy distinct habitats within the Galapagos region, such variations may reflect particular biochemical adaptations to their respective environments. In order to further probe the relationship between environment and serum protein modifications, the physical properties of several South American iguanid albumins were analyzed; the results form the basis of this report. These observations include the first compara-

tive biophysical study of Galapagos iguana serum albumins.

Materials and methods. Adult iguanas, *Amblyrhynchus cristatus* (Galapagos marine iguana), *Conolophus pallidus* and *C. subcristatus* (Galapagos land iguanas), and *Iguana iguana* (mainland South American iguana), were bled by cardiac puncture and the sera separated from clotted blood by centrifugation³. Serum albumins were isolated by precipitation in 10% trichloroacetic acid followed by solubilization with 100% ethanol². Electrophoresis of whole sera and purified albumins utilized cellulose acetate/Tris-barbital pH 8.8⁴ and 7% acrylamide/Tris-glycine pH 9.2² systems. Molecular sieve chromatography of iguana serum proteins was done on Sephadex G-200 (Pharmacia) with 0.15 M NaCl/10⁻³ M Tris buffer, pH 6.8; absorbancy of 2-ml effluent fractions was recorded at 280 nm. Albumin molec-