authentic sample (Co-TLC, m.m.p. and superimposable IR spectra).

This is the first report of the occurrence of the above three compounds in Eucalyptus hybrid leaves. 2-α-Hydroxy ursolic acid is reported for the first time in the genus Eucalyptus and for the third time in the family Myrtaceae. It also occurs in the leaves of Psidium guajava Linn5, and Callistemon lanceolatus DC 6.

Ursolic acid is of medicinal value. It is a well-known anti-inflammatory, antibiotic, antiarthritic, antiallergic and hypolipidemic agent 6. About 4.28 lakh hectares of Eucalyptus hybrid were planted 7 in India up to 1977. Plantation programmes of various states include plantation of this species in large areas and by now the area under this species will be much more. About 1.2% of ursolic acid occur in the leaves of E. hybrid as reported above. Thus, the large quantities of the leaves available in the country may be utilized for the isolation of ursolic acid.

The author is thankful to Shri G.P. Maithani, and Dr K.S. Ayyar, for their keen interest. to Dr R.K. Buwa of K.D. Malaviya Institute of Petroleum Exploration, Dehra Dun for mass spectra, and to Prof. M.R. Parthasarathy, Department of Chemistry, University of Delhi for the authentic sample of 2-α-hydroxy ursolic acid. NMR and IR spectra.

24 December 1986; Revised 3 March 1987

NEW RECORD OF A BACULOVIRUS DISEASE IN LEUCINODES ORBONALIS GUEN

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LEUCINODES ORBONALIS Guen is a serious pest of eggplant throughout India often resulting up to 80% infestation 1. It has also been reported on potato 3 and other wild plants 5. The information on natural biotic mortality factors of the pest is limited and has been reviewed by Tewari and Krishna Moorthy 4.

During September–October, 1985, the survey studies conducted on the natural mortality factors of L. orbinalis revealed the presence of sluggish larvae with symptoms of loss of appetite on fruits of eggplant. With passage of time they became fragile with flaccid body and died inside the fruit. The disease infection varied from 1.06 to 6.38% (table 1).

As the larvae were suspected to be virus-infected, further studies were carried out on isolation, purification and identification of virus in the laboratory. The infected larvae were macerated in 0.05 M phosphate buffer (pH 7.2) at 1 ml of buffer per g of infected larvae in a pestle mortar for 5 min and filtered through double muslin cloth. The filtrate was centrifuged at 10,000 g for 15 min and the supernatant was mixed with 6% polyethylene glycol (6000 MW) and 0.1 M NaCl, shaken for 30 min and then centrifuged at 30,000 g for 1 hr. The supernatant was discarded and the pellet was dissolved in 0.05 M phosphate buffer (pH 7.2). The virus was further purified by differential ultracentrifugation at 1,50,000 g for 2 hr. The pellet was collected and dispersed in 0.05 M phosphate buffer (pH 7.2) and again centrifuged at 7,000 g for 10 min. The supernatant was collected and this constituted the purified virus preparation. The purified virus suspension was sprayed on formvar-coated copper grids stained with

<table>
<thead>
<tr>
<th>Date</th>
<th>No. of larvae observed</th>
<th>No. of larvae discarded</th>
<th>Infection (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>4-9 1985</td>
<td>188</td>
<td>2</td>
<td>1.06</td>
</tr>
<tr>
<td>10-9 1985</td>
<td>242</td>
<td>9</td>
<td>3.72</td>
</tr>
<tr>
<td>23-9 1985</td>
<td>164</td>
<td>8</td>
<td>4.88</td>
</tr>
<tr>
<td>3-10 1985</td>
<td>47</td>
<td>3</td>
<td>6.38</td>
</tr>
</tbody>
</table>
2% uranyl acetate (pH 3.5) and examined in JEM 100-S (JEOL) model transmission electron microscope.

The rod-shaped particles of baculovirus were observed in the electron microscope (figure 1). The size of the virus particles ranged from 194 to 286 x 83 to 143 nm (Av. 250 x 122.4 nm). The pathogenicity was also tested by injecting virus suspension in healthy larvae. Inoculated larvae exhibited symptoms as described earlier. The percentage mortality was 100%.

A perusal of literature revealed that the occurrence of baculovirus on L. orbonalis is the first record. The present findings have opened the new possibility of management of this pest through baculovirus. Moreover, baculoviruses are now emerging as potent vehicles for gene transfers because of their large sized genome.

19 May 1986; Revised 13 August 1986


**PROPAGATION OF ZEPHYRANTHES THROUGH TISSUE CULTURE**

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*Zephyranthes carinata* L., 'Rose Amaryllis', as it is known (Fam. Amaryllidaceae), is a herbaceous ornamental producing very showy, bright, pink flowers. Propagation is by means of bulbs and it was considered useful and interesting to develop a method by which the plant could be multiplied in larger numbers and in less time by the application of tissue culture techniques. Literature pertaining to the propagation in *vitro* of several allied plant species such as *Lilium, Amaryllis, Iris* and other ornamentals is available but *Zephyranthes* does not seem to have been subjected to experimentation of any kind. Hence this bulb plant was investigated with a view to studying its response in aseptic cultures aimed at regenerating complete bulblets.

Material was obtained from plants growing in the garden attached to this Institute. The bulbs were washed with water to which a few drops of Teepol had been added as a surfactant. They were sterilized using 5% NaOCl and then with HgCl₂ (0.25%) each for 10 min and then washed thoroughly in sterilized distilled water. The bulbs (20 x 18 mm in dia) were divided into halves and quarters by vertical incisions and planted on Murashige-Skoog (MS) medium to which cytokinins and auxins in different concentrations had been added and gelled by 0.8% agar. The cultures were exposed to 16 hr, photoperiod and

**Figure 1-3. Stages in development of leafy bulblets in culture. Note abundant rooting in figure 3.**