Table 1  *Schistosoma incognitum* worm recovery from white mice along with the egg out put in the faeces. Faecal exam was started on 30th day.

<table>
<thead>
<tr>
<th>No. of cercariae (S. incognitum)</th>
<th>Time in days before sacrifice</th>
<th>Schistosoma incognitum Recovery</th>
<th>Turned positive (in days)</th>
<th>Faecal Egg load Min. (eggs per g)</th>
<th>Faecal Egg load Max. (eggs per g)</th>
</tr>
</thead>
<tbody>
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<td>36</td>
<td>145</td>
<td>23</td>
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<td>36</td>
<td>153</td>
<td>69</td>
<td>-ve</td>
<td>-</td>
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<td>40</td>
<td>36</td>
<td>8</td>
<td>-ve</td>
<td>-</td>
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<tr>
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<td>180</td>
<td>36</td>
<td>-ve</td>
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<td>8</td>
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<tr>
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<td>73*</td>
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<td>1</td>
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<td>71*</td>
<td>2</td>
<td>1</td>
<td>49</td>
<td>300</td>
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</tbody>
</table>

Died. Worm recovery was not by perfusion technique.

2 April 1985


**ANTHER CULTURE OF CATHTHANTHUS ROSEUS L.—DEVELOPMENT OF POLLEN EMBRYOIDS**

**LEELA GEORGE**

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*Catharanthus roseus* L. is a small herbaceous subshrub growing wild and also cultivated because of its medicinal value. Vincristine and vinblastine now extracted commercially from the leaves of *C. roseus*, are used in the treatment of leukaemia and Hodgkin's disease and also other types of cancer. In recent years, the technique of anther culture is being increasingly used for rapid isolation of homozygous recombinants in several plants of economic importance. Anther culture of *C. roseus* has been initiated for rearing the haploids and isolating the superior lines with increased alkaloid content. The following is a brief account of the results obtained.

Flower buds at the uninucleate stage of pollen development (bud size 0.3–0.5 cm) collected from vigorously growing plants were surface-sterilized and the anthers were cultured aseptically on MS basal medium alone or MS medium containing various auxins, cytokinins and other growth adjuvants in varying concentrations and combinations. Solid as well as liquid media containing 8% sucrose were used for culturing the anthers. For cold treatment prior to culture the buds were stored at 10°C for 0, 3, 7 and 14 days. Elevated temperature treatment was given by incubating the cultured anthers at 30°C for 0, 3, 7, 14 and 21 days in a BOD incubator before transferring them to 22±2°C in dark. For each experiment a minimum of 240 anthers were used and over 11,000 anthers were cultured in all.

A few anthers were examined at regular intervals from each treatment. Most of the anthers turned brown in 6–8 weeks. Addition of activated charcoal into the medium did not prevent their browning. All the anthers were observed cytologically after 8 weeks
of culture. They were squashed in 1% acetocarmine and examined in a microscope.

When cultured on a medium containing 2% sucrose, profuse callusing occurred from all over the anthers. Cytological examination of the callus showed that the callus was of somatic origin (2n = 16).

The pollen were mostly uninucleate (figure 1) at the time of culture. There was no division in the pollen when anthers were cultured on MS medium alone. The first division was observed 9–10 days after culture (figure 2) when anthers were inoculated on MS medium containing an auxin (NAA, IAA or 2,4-D, 0.1–0.5 mg/l) and a cytokinin (kinetin, BAP, 2iP or Zeatin; 0.05–3 mg/l). However, further divisions could not be obtained in most of these media irrespective of the culture conditions. Cold treatment of buds (0–14 days at 10°C) and incubation of cultured anthers at 30°C (0–21 days) did not enhance pollen embryoid formation. Repeated divisions leading to the formation of pollen embryoids occurred only when anthers stored at 10°C for 3–7 days were cultured on MS liquid medium containing NAA (0.5 mg/l) and BAP (0.05 mg/l) and incubated at 30°C for 3–7 days (figures 3 and 4). About 5% of the anthers responded in culture. The pollen embryoids failed to grow beyond the early globular stage even when transferred to fresh medium of the same composition or with 3% sucrose.

Since the pollen have the potential to form embryoids in culture, isolated pollen culture may lead to the development of pollen plants in this species. This aspect is under investigation.

The author thanks Dr P. S. Rao, Head, Plant Biotechnology Section, for useful discussions and encouragement.

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SCANNING ELECTRON MICROSCOPIC STUDIES ON THE SPERMODERM OF SOME LEEA SPECIES

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CONSIDERABLE literature exists on the seed anatomy of Leea species\textsuperscript{1}, but no work on the scanning of the spermoderm of this taxon has been done hitherto. The present paper, therefore, deals with this aspect in Leea indica (Burm. f.) Merr. Philip and L. macrophylla Roxb. ex Hornom.

The seeds of L. indica were collected from Santhal Pargana (L\textsubscript{1}), Sikkim Himalayas (L\textsubscript{2}), Khasi hills (L\textsubscript{3}) and those of L. macrophylla from Santhal Pargana.

Dry mature seeds after cleaning were attached to aluminum stub by silver paint and coated with a very thin layer of gold. For uniformity, central portion of dorsal side of seeds were scanned in the Philips-5 SEM at RSIC, Bose Research Institute, Calcutta.

Spermoderm of L. macrophylla shows irregular fissures with fibrillar waxy depositions (figure 1). These fissures run along the long axis of the seed. Spermoderm of L. indica shows considerable variations, hence described separately. Spermoderm of L\textsubscript{1} shows prominent ridges and furrows with heavy depositions of wax on ridges (figure 2). Spermoderm of L\textsubscript{2} shows irregular polygonal areas having more deposition of wax on ridges and net-like deposition