

scopoletin to reduced photosynthesis. The distribution of proanthocyanidins in the leaves and their role in photosynthesis need a detailed investigation.

15 May 1985

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A DOUBLE-EMBEDDING TECHNIQUE FOR SECTIONING EMBRYOIDS

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INVESTIGATIONS on plant tissue cultures, particularly those involving elucidation of the process of embryoidogenesis (a term for the development of embryoid *in vitro*) and the morphology of the embryoids, the histological characteristics of tissue organization, the mode of vascular differentiation, etc, would involve the sectioning of embryoids at different stages of growth. The minuteness of some of these materials, particularly the embryoids, renders them extremely difficult to manipulate if processed by the customary methods. The celloidin-spray method of Bhandari¹ is quite acceptable but for the high cost of celloidin and the stringent conditions for material preparation. The present communication describes a much simpler and less expensive procedure. In fact, this method, designated the double-embedding method gave extremely satisfactory results in the androgenetic investigations on *Nicotiana tabacum*^{2,3}. This method involves the embedding of materials twice; first in agar sol and then the solidified agar blocks with the materials are re-embedded in paraffin by the customary procedure.

The procedure for double-embedding is given below:

1. Dissolve about 1g of good quality agar-agar powder (Difco or other good make) in 100 ml of hot water.
2. Get the materials to be embedded ready: fixed as well as fresh materials can be chosen. Prior staining of material is optional for fixed materials.
3. Prepare a paper boat of convenient size and apply a thin coat of glycerine on its inner surface.
4. Pour a required quantity of agar sol into the paper boat. Transfer the materials into the boat and arrange them carefully leaving enough space between materials for easy partitioning of individual blocks later.
5. Place the boat in a cold chamber to facilitate rapid and uniform solidification.
6. Cut out each agar block containing the material, taking care to retain a uniform sheath of agar around the material.
7. Process the agar blocks for microtomy by customary methods⁴. If fresh materials are agar-embedded, proper fixation is essential before pro-

cessing for microtomy. Infiltrate the agar blocks and re-embed them in paraffin.

8. Section the material, process the slides by customary method and stain them in a suitable schedule. Haupt's adhesive and 3% formalin are effective for (floating and affixing) the paraffin-agar ribbons.

A thin film of agar persists on the slide around the material even after mounting and staining. Being transparent this does not seriously hinder observation or photography. This method is also useful for histochemical investigations.

The author thanks Dr D. A. Govindappa for encouragement.

1 May 1985

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PROMOTION OF FEMALE FLOWER FORMATION IN CASTOR BEAN (*RICINUS COMMUNIS* L) BY PHTHALIMIDES

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EXOGENOUSLY applied plant growth regulators are known to affect sex expression in several monoecious and dioecious species^{1,2}. Studies have been made on the possible role of gibberellin^{3,4}, amino acids⁵, silver and cobalt ions⁶, ethrel and chloroflurenol⁷ and peroxidase and its isozymes⁸ in sex expression of *Ricinus communis*. Gibberellin, when applied to the plants exogenously, promotes the formation of female flowers in this species⁷. Recently, some substituted phthalimides are reported to possess several growth regulating properties^{9,10} and have been found to regulate the sex expression in monoecious and gynoeious cucumber^{11,12}. The present work was undertaken to investigate the effect of phthalimides on sex expression in *R. communis* L, a monoecious plant.

Plants of *R. communis* L were raised from seeds in

earthen pots filled with properly manured soil. Foliar applications of two derivatives of phthalimide were made upto run-off level to the plants having 12–15 nodes before inflorescence initiation. The plants were sprayed with 125, 250 and 500 mg l⁻¹ of 1-(3-chlorophthalimido)-cyclohexanecarboxamide (AC-94377) or 1-(1-cyclohexene-1,2-dicarboximido)-cyclohexanecarboxamide (AC-99524) supplemented with 0.1% triton X-114 as a wetting agent. Plants sprayed with distilled water containing only 0.1% Triton X-114 were considered as control. Second and third applications were made after 7 and 14 days of the first spraying respectively. Each treatment consisted of 6 plants replicated 2 times. The data are expressed as mean value \pm S.D.

Phthalimides increased the percentage of female flowers with simultaneous decrease in the percentage of male flowers on the treated plants. The percentage of female flowers increased from 21.2 (control) to 26.4, 34.6 and 28.5 at 125, 250 and 500 mg l⁻¹ of AC-94377. The plants treated with AC-99524 also exhibited increased percentage of female flowers which were 24.2, 29.6 and 32.3 at 125, 250 and 500 mg l⁻¹ respectively as compared to the control. Male flowers constituted the remaining percentage at each concentration of both the phthalimides. AC-94377 at 250 mg l⁻¹ and AC-99524 at 500 mg l⁻¹ were most effective for promoting the formation of female flowers (table 1). Overall, AC-94377 was more effective than AC-99524 in inducing femininity in this species. The total number of male and female flowers per plant increased with the increase in the concentration of each phthalimide but the size of male flower buds got reduced in both the cases. At 500 mg l⁻¹ of each phthalimide, some male flower buds failed to open, dried and abscised. Female flowers produced on the treated plants were somewhat thin and elon-

Table 1 Effect of phthalimides (AC-94377 and AC-99524) on sex expression in *Ricinus communis* L

Treatment (mg l ⁻¹)	Percentage of female flowers \pm S.D.	Total No. of flowers per plant mean \pm S.D.
Control	21.2 \pm 2.4	42.6 \pm 10.2
AC-94377		
125	26.4 \pm 2.9	53.2 \pm 8.3
250	34.6 \pm 3.8	59.0 \pm 11.4
500	28.5 \pm 3.3	64.3 \pm 9.0
AC-99524		
125	24.2 \pm 2.5	50.8 \pm 8.0
250	29.6 \pm 3.5	58.3 \pm 6.7
500	32.3 \pm 4.1	64.0 \pm 12.3