

Figures 1 and 2. 1. *Christella multiauriculata* Punetha, Holotype; 2. 72 bivalents during meiosis in a spore mother cell of *C. multiauriculata*.

Jermy is fairly widespread all over the region and is extremely variable in its leaf morphology. Its variable leaf morphology lead many taxonomists to establish several varieties within it¹. Recently Punetha³ described a new species, *C. multiauriculata* from Kumaon, which in general morphology is identical to *C. dentata* but differs distinctly from the latter in having slender capitate hairs between veins on the lower surface of the leaflets, several pairs of

pinnae above the base having well-developed auricles on the acroscopic bases (hence the name) and presence of a few pairs of basal pinnae reduced and not more widely spaced (figure 1). Cytologically, *C. dentata* (Forssk.) Brownsey and Jermy have been represented by diploid and tetraploid races⁴. No hybrids have so far been found in nature, however, intraspecific crossing between diploid and tetraploid resulted triploids⁵.

The cytology of the fern was studied to determine whether *C. multiauriculata* is a distinct species or whether they are mere cytological variations. Acetocarmine smears of developing sori showed unequivocally 72 bivalents suggesting that *C. multiauriculata* is a sexual tetraploid species (figure 2).

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GENOTYPIC DIFFERENCES OF *IN VITRO* LATERAL BUD ESTABLISHMENT AND SHOOT PROLIFERATION IN PAPAYA

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PAPAYA (*Carica papaya*) is a principal horticultural crop of tropics and is a good source of provitamin 'A' and ascorbic acid. It also yields an alkaloid, carpaine, which is used as a heart depressant, amoebacide and as diuretic¹. Some papaya production is devoted to the recovery of papain, a proteolytic enzyme with several commercial applications².

Papaya, like other higher plants, has an indeterminate mode of growth. The leaf axes contain subsidiary meristems each of which is capable of growing into a shoot that is identical to the main axis. These axillary buds, when cultured in hormone

containing nutrient medium, result in secondary, tertiary and later order shoots in a proliferating cluster. When such a cluster attains a diameter of 3 cm, it can be subdivided into smaller clumps of shoots or into individual shoots. These, in turn, form larger clusters and multiplication process can be continued indefinitely. This approach of micro-propagation has two immediate advantages: (i) it is useful in maintenance and multiplication of selected genotypes, and (ii) the enhanced multiplication rate saves considerable time and effort during initial stages of introducing a new cultivar for commercial production. In the present communication, we compare the genotypic differences of improved papaya varieties in responding to multiplication in cultures.

Lateral buds, 1 cm² in size, were isolated from 3 to 4-month-old juvenile plants of four genotypes of papaya namely Co₆, Co₃, Co₂ and delicious. The buds were washed with detergent (teepol) solution and surface-sterilized for six min in 0.1% mercuric chloride solution. The mercuric chloride was removed by repeated washings with sterile water. This was followed by a 3 h incubation in water in a rotary motion orbital environmental shaker gyrating with 60 rpm to facilitate removal of latex from the explant.

The buds were rinsed again in sterile water and trimmed to include 2–3 mm size axillary bud with < 1 mm base by dissecting out the unwanted tissue. The buds were cultured initially in an establishment medium and were later transferred to proliferation medium^{3,4}. Murashige and Skoog's⁵ basal medium with 3% sucrose as a source of carbohydrate and solidified with 0.8% agar was used. The pH was adjusted to 5.8 before adding agar. The establishment medium, different from that used by earlier workers^{3,4}, consisted of growth regulator IPA (2-isopentenyl adenine) and NAA (α naphthalene acetic acid) at 10 and 2 mg/l respectively. This combination of growth regulators was found to show superior response in the establishment of aseptic cultures compared to BAP and NAA at similar concentrations. The cultures were incubated in light and dark cycles of 16 h and 8 h respectively at 25 \pm 1°C.

Following establishment for 3 to 4 weeks, the buds were transferred to MS medium containing BAP (6-benzylamino purine) and NAA at 0.56 mg/l and 0.08 mg/l respectively for proliferation. The experiment was replicated thrice with sample size of a minimum of 25 explants per genotype per replication. The mean per cent response was computed

Table 1 Genotypic variation for establishment and proliferation responses of lateral buds of papaya

Genotype	Establishment %	Proliferation %		
		Rapid	Low	Total
Co ₆	42.0	65.5	6.8	72.3
Co ₃	75.7	12.1	30.3	42.4
Co ₂	60.3	30.3	12.1	42.4
Delicious	80.3	27.9	18.6	46.5

both for the establishment and the proliferation phase (table 1).

The data presented in table 1 show considerable variation in the establishment responses in the four genotypes investigated. The cultivar delicious, shows a high rate of establishment while Co₆ responds the least for aseptic explant establishment. Proliferation, which is the critical phase in the propagation procedure, shows little genetic variation, for three of the four genotypes investigated. The variety Co₆ which has the lowest establishment rate responds the best to proliferation. Partitioning of proliferation responses on the basis of rate of growth, characterized by faster growth in a limited period of 2 weeks, however, indicates a wider variation. Again, the cultivar Co₆ shows a high frequency of explants responding to rapid proliferation. The varieties Co₂ and delicious did not differ much in their responses to rapidity of proliferation.

The genotypic differences reflect genetic differences in the capacity of explanted tissues in metabolizing the organic and inorganic components of the nutrient medium including phytohormones. Such genetic differences in tissue culture response have been demonstrated in several species⁶.

The present study has important implications in the choice of genotype of papaya taken up for micropropagation and the efficiency with which cost-effective tissue culture propagation can be expected.

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GOSSYPOL AFFECTS PLANT SPERMS ALSO

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Gossypol, a predominant polyphenolic triterpenoid yellow pigment of cotton seed, has been acclaimed to be an effective oral contraceptive in man, monkey, boar, dog, bull, hamster, guinea pig and rat. It decreases sperm count and renders the sperms malformed, immotile and inviable¹⁻⁵. To

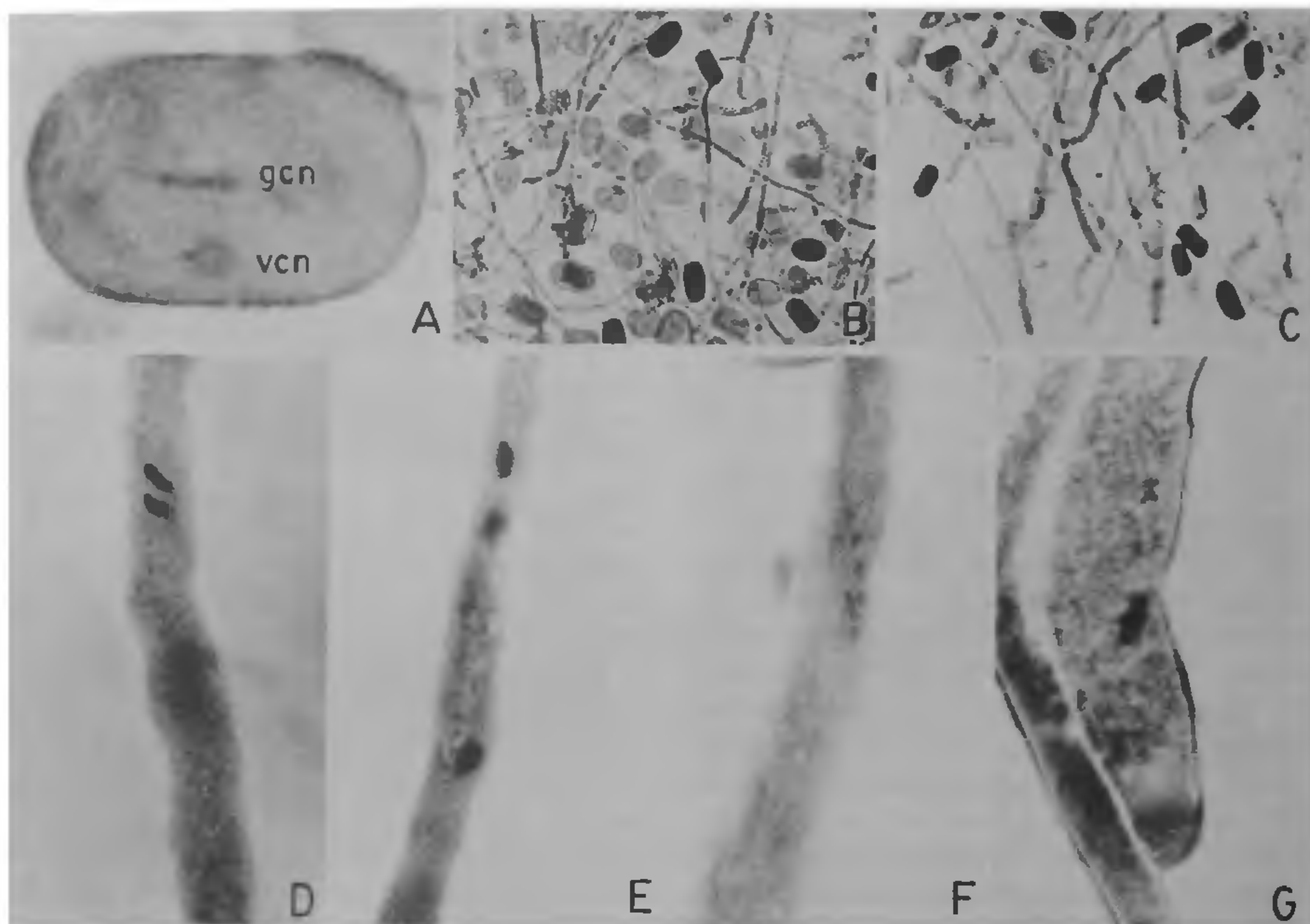


Figure 1A-G. Effects of gossypol on *in vitro* pollen germination, pollen tube growth and male gametogenesis in *Impatiens balsamina*. All but A are from sitting drop cultures stained with 1% aceto-orcein. In D-G the pollen tube tip is orientated toward the bottom of the page. A. Pollen grain showing 2-celled condition; note the globose vegetative cell nucleus (vcn) and the elongate generative cell nucleus (gcn) which is in prometaphase ($\times 1143$); B, C. 240-min-old cultures in BM (control) and in BM + 10^{-3} M gossypol. The density of pollen tubes in B is an expression of higher per cent pollen germination and greater growth of pollen tubes in the control medium. Several nonopaque grains which have issued pollen tubes were initially opaque. Owing to matting of pollen tubes, only a few tubes can be traced their entire length. Contrast between B and C is obvious. Several opaque grains have failed to germinate. Also, pollen tubes are shorter than those in B, and most pollen tube tips are club-shaped ($\times 132$); D. Anaphase of gametogenesis in pollen tube from 90-min-old culture in BM (control) ($\times 964$); E. Pollen tube from 240-min-old culture in BM showing telophase of gametogenesis and vegetative cell nucleus close to tube tip ($\times 964$); F, G. Pollen tubes from 90-min-old cultures in BM + 10^{-3} M gossypol showing characteristic arrest of male gametogenesis ($\times 964$).