Bot. Bull. Academia Sinica (1989) 30: 1-7

Developmental studies on shoot apical organization in *Zinnia elegans* Jacq.

Manju Sharma and K. C. Sharma[1]
Department of Botany, University of Rajasthan
Jaipur-302 004, India

(*Received March 25, 1988; Accepted August 15, 1988*)

Abstract. Organization of the shoot apical meristems in *Zinnia elegans* Jacq. was studied from the mature embryo to flowering stage. The embryonal shoot apex flanked by a pair of opposite leaf primordia, shows a double layered tunica covering the homogenous corpus. Zonation based on cell net analysis is established on the fifth day after seed wetting. Plastochron dependent variations in size of the apex and central mother cell zone are observed. The apex enters the reproductive phase through a transitional stage. A cambium-like zone is seen in the vegetative apex of fourth week and in the transitional apex. The inflorescence apex shows a mantle-core organization and produces involucre of bracts and acropetally arranged floret primordia. The entire apex is consumed and no residual meristem remains.

Key words: Ontogeny; Plumular apex; Reproductive apex; Shoot apical organization; Vegetative apex; *Zinnia elegans* Jacq.; Zonation.

Introduction

There are a few reports (Rogan and Smith, 1974; Mauseth, 1978 a, b) on changes in shoot apical organization studied during embryonal to reproductive phases. Sharma and Pillai (1986) reported ontogenetic studies on shoot apical organization in *Raphanus*. Data on origin, organization and behaviour of shoot apical meristems from mature embryo to flowering stage in *Zinnia elegans* Jacq. are reported here.

Materials and Methods

Plumular apex from mature embryos dissected out from the soaked seeds of *Zinnia elegans* Jacq. and shoot apices from germinating seeds at 24-h intervals for the first seven days after seed wetting and subsequently from growing plants at weekly intervals till flowering were collected and fixed in FAA. Fifteen to twenty apices from each collection were dehydrated through TBA series, sectioned at 6-7 μm and stained with tannic acid-iron chloride, safranin, light green combination as well as with PAS reaction. Diameter (D) of the apex was measured from the adaxial bases of the two youngest leaf primordia using an oculometer. Based on comparisons of the height (H) of the apex above the axil of the youngest leaf primordium and the height of the latter above the same reference point, the apices were assigned to their relative positions in the plastochron. The plastochronic index

[1] Mailing address: B-88, Surya Marg, Tilak Nagar, Jaipur-302 004, India.

(PI)=Height of the apex in μm/Height of the youngest leaf primordium in μm, was calculated following Paolillo and Gifford (1961). Depth of CMZ and mature pith were measured from the summit of dome. H/D ratio calculated at different developmental stages of the shoot apex and higher values of this ratio were considered to be indicative of shoot elongation.

Observations

Zinnia elegans Jacq., the annual herb, flowers after five weeks of vegetative growth. The plumular apex and nonzonate and zonate vegetative apices show a tunica-corpus organization and the reproductive apices a mantle-core organization.

The Plumular Apex

The mature embryo shows a flat plumular apex flanked by two opposite foliage leaf primordia (about 73.0 μm long) (Fig. 1). The apex with two-layered tunica covering a homogeneous corpus measures about 64.0 μm in diameter. Both tunica and corpus are uniformly densely stained. The corpus is about 59.0 μm deep.

The Vegetative Apex

The nonzonate apex: The one-day-old seedling shoot apex is flat and narrow (35.0 μm wide) (Figs. 2 and 3). The corpus is 62.0 μm deep. During the first three days after seed wetting the embryonal leaves grow to a height of about 258.0 μm. The apex remains flat while there is a fall in the diameter from plumular to the one-day-old apex after which a gradual increase during the first week (Fig. 12).

The second pair of leaves is initiated at four days after seed wetting. Cells at the site of leaf initiation are denser stained and divide in various planes. About 10–12 cells at the proximal end of the corpus in the 4-day-old apex indicate early stages in formation of pith meristem (PM).

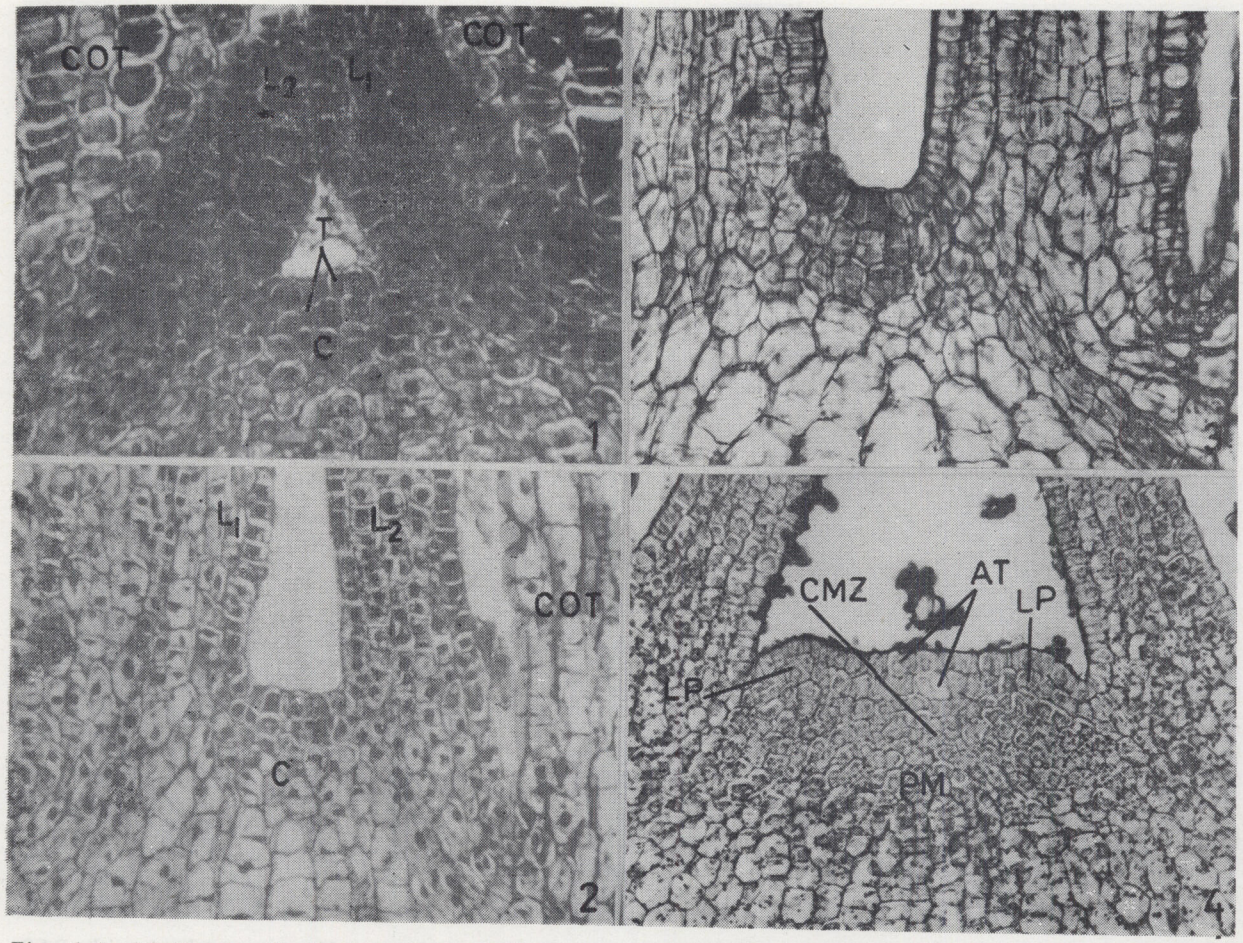
The Zonate apex: The apex during 5 days to 5 weeks is a flat to low and broad dome (H/D ratio of 0.07) when dome depending on the plastochronic stage. The tunica is 2–3 layered. Axially located tunica cells are broader, lightly stained and have larger vacuoles. Based on differences in planes of cell arrangement, the corpus may be demarcated into peripheral zone (PZ) with regularly arranged cell layers, central mother cell zone (CMZ) with irregularly arranged cells and pith meristem (PM) with cell files subjacent to the CMZ. This zonation pattern is well established at 5 days but all the zones are uniformly densely stained. A few apices also show lighter stained CMZ (Fig. 6). The apex is described under three plastochronic stages (minimal, mid, maximal) and the plastochronic index (P. I.), depth of CMZ and mature pith etc. are given in the Table 1.

Minimal stage: The apex is flat and narrow. Tunica is 2-layered. Packets of 2–3 cells enclosed in a common cell wall are observed in the CMZ. A few oblique divisions in this

Table 1. *Plastochronic Index (PI) and measurements at the three plastochronic stages*

Values are averages of 15–20 readings.

Plastochronic stage	Height of apex (μm) (a)	Height of youngest leaf primordium (μm) (b)	PI (=a/b)	Diameter of apex (μm)	Depth of CMZ (μm)	Depth at which pith occurs (μm)	H/D ratio
Minimal	—	30.8	—	65.6	54.8	80.6	—
Mid	5.0	116.5	0.05	73.4	62.2	88.5	0.07
Maximal	5.8	182.8	0.03	84.2	76.2	107.5	0.07



Figs. 1–4. Median longitudinal sections of the plumular, nonzonate and zonate apices stained with normal morphological stains (Fig. 1, 2) and with PAS reaction (Fig. 3, 4). Fig. 1. Plumular apex ×300. Figs. 2, 3. Nonzonate apices ×375. Fig. 4. Zonate apex at minimal stage ×250. (AT-Axial tunica, C-Corpus, CMZ-Central mother cell Zone, COT-Cotyledon, L_1, L_2-Embryonal foliage leaves, LP-Leaf Primordium, PM-Pith meristem, T-Tunica).

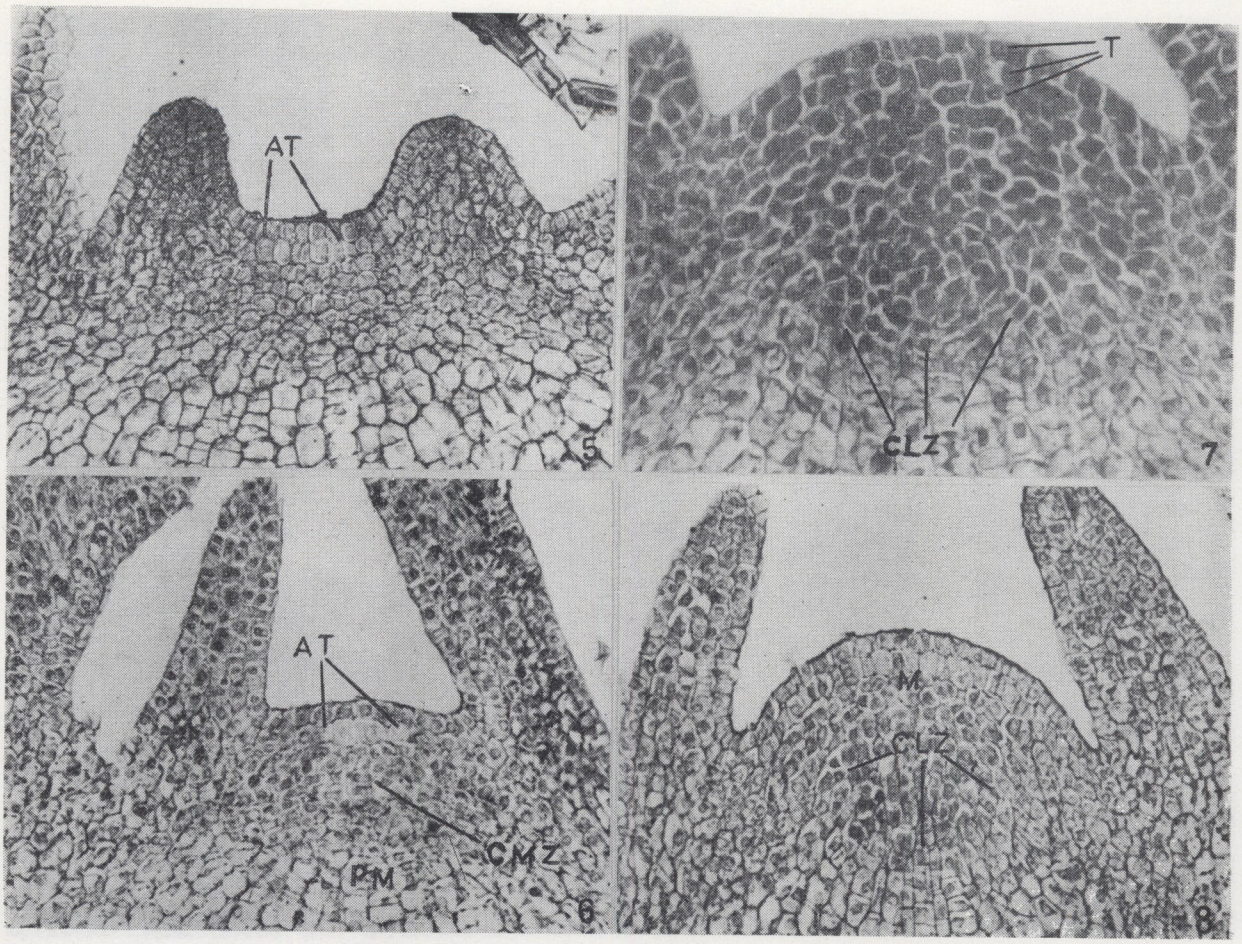
zone indicate the contribution of cells to the PZ and PM. The PZ is not clear at this stage. Cells in the PM are arranged in 3–4 tiers of 3–4 cells each (Fig. 4).

Mid stage: The apex shows an increase in size and depth of CMZ. It is a very low (almost flat) dome. Distal cells of the CMZ are stratified simulating the tunica. PZ is 2–3 layered and PM is similar to the minimal stage (Fig. 5).

Maximal stage: The apex shows a further increase in size though it is still a low dome with a greater increase in diameter than height. The CMZ remains below the attachment of youngest leaf primordia. PZ is 3–4 layered and PM is similar to the previous stages. (Fig. 6).

The Reproductive Apex

The inflorescence is an indeterminate capitulum as the apex produces involucre of bracts followed by florets in an acropetal order. The main shoot apex enters the reproductive phase through a transitional stage during the fifth week. The apex at 4 weeks shows an increase in size with an H/D ratio of 0.19 (Fig. 7). There is a cambium-like zone of about 3–4 cell layers showing parallel



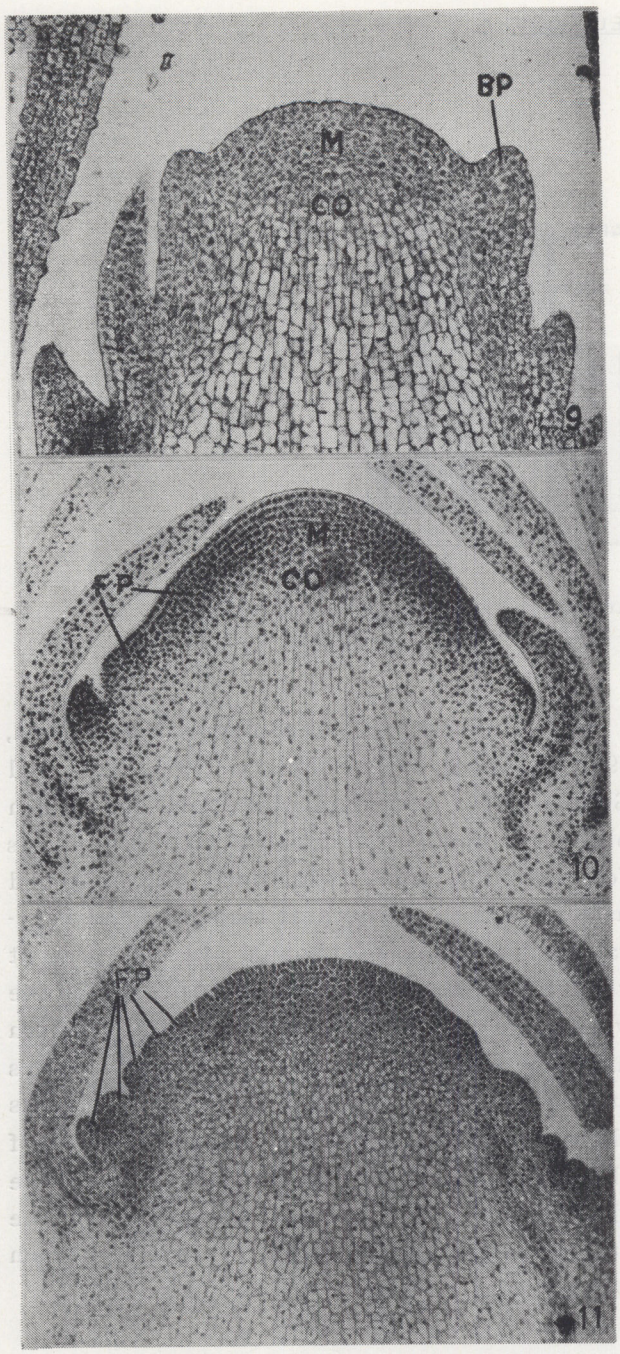
Figs. 5–8. Median longitudinal sections of the stoot apex. Fig. 5. Apex at mid stage stained with PAS reaction×250. Fig. 6. At maximal stage×250. Fig. 7. Apex during fourth week showing enlargement and cambium-like zone×325. Fig. 8. Transitional apex×250.
(AT-Axial tunica, CLZ-Cambium-like zone, CMZ-Central mother cell zone, M-Mantle, PM-Pith meristem, PZ-Peripheral zone, T-Tunica).

alignment and arranged in a cup-shaped zone immediately proximal to the CMZ (Figs. 7, 8). This zone extends across (80.8 μm) the apical dome in a concave arch, the peripheral region of which merges with the PZ. The cells in this zone are small, densely stained and divide by regular periclines, proximal derivatives of these differentiate into the pith.

The transitional apex: The high and broad dome of the fourth week evidences a further increase in size during the fifth week forming a transitional apex (52.0 μm high and 154.0 μm broad). Width of the cambium-like zone is increased to 128.0 μm. The mantle is 4–5 layered (Fig. 8).

The inflorescence apex: The apex bearing the first two bract primordia (in L. S.) measures about 51.0 μm in height and 164.0 μm in diameter. Further increase in diameter than in height results in "squarring" of the apex (H/D ratio at this stage is 0.31) (Fig. 9). The apex produces bract primordia in quick succession resulting in closely arranged bracts.

After producing an involucre of bracts the diameter of the apex increases further to 174.0 μm and the height is only 39.0 μm. Later, size



Figs. 9–11. Median longitudinal sections of the reproductive apices. Fig. 9. Reproductive apex showing mantle-core organization and producing bracts ×150. Fig. 10. Main reproductive apex with early stages in floret formation ×150. Fig. 11. With floret primordia initiation in acropetal order×100.
(BP-Bract primordium, CO-Core, FP-Floret Primordium, M-Mantle).

of the apex increases further and the apex with a mantle-core organization and producing florets, measures about 73.0 μm in height and 180.0 μm in diameter (Fig. 10). Florets are initiated from the peripheral mantle layers which are denser stained than the axial mantle (Fig. 11). The second and third mantle layers at the site of floret primordia show denser staining and divide in various planes. The outermost mantle layer cells at these sites show frequent anticlinal divisions. The apex is consumed in floret formation and no residual meristem remains.

The floret apex: The floret primordia (14.5 μm high and 50.0 μm broad) show densely stained 2–3 layered mantle and a subjacent core. The floral parts originate acropetally.

Discussion

There is an age-related gradual increase in size of the apex as reported by others (Rogan and Smith, 1974; Sharma and Pillai, 1986) and a sharp increase prior to flowering. The apex is consumed in floret formation and no residual meristem left. No correlation was seen between shape of apex and age of the plant.

The majority of reports document the presence of a cytohistological zonation superimposed on a tunica-corpus organization in the herbaceous angiosperm shoot apices. Mauseth (1978a) and Sharma and Pillai (1986) tracing the origin, development and establishment of zonation, reported that initiation of zonation in the seedling shoot apex is not directly related to shape, size or the number of plastochronic cycles. The present study supports this. But a well marked cytohistological zonation is not seen at all the developmental stages of the apex though the planes of cell division and cell arrangement indicate a zonate pattern.

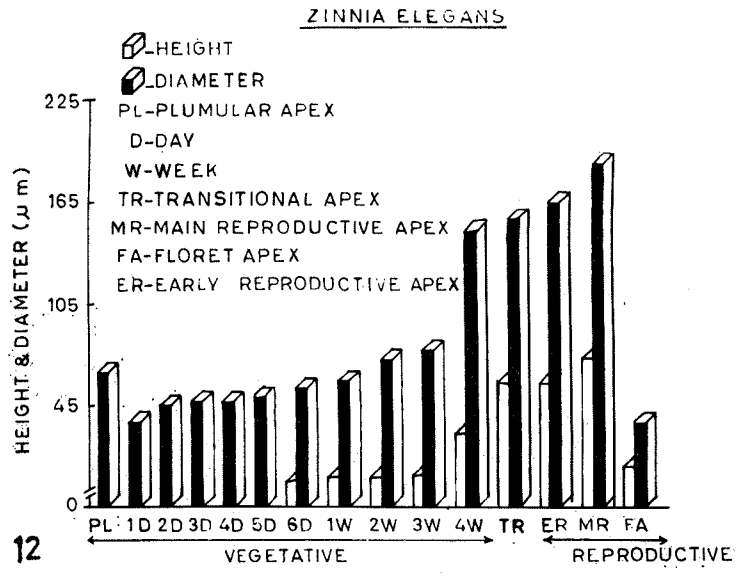


Fig. 12. Histogram depicting height and diameter of the shoot apex at different developmental stages. (Values are averages of 15-20 readings).

A few distal CMZ cells and 3-4 axially located second tunica cells are broader and have larger vacuoles. Earlier adherents of French school considered the méristème d' attente (comparable to the CMZ) to be mitotically inactive during vegetative growth. Other workers maintain that cells of the CMZ are active to varying degrees, but apparently more active than those of the quiescent centre in roots. The presence of cell divisions and blocks of cells enclosed in a common cell wall (indicating recent divisions in the CMZ) precludes the interpretation of this zone as méristème d'attente. Anatomical stains show only minor differences between cells of the CMZ and PZ and possibly this may indicate a more active CMZ in *Zinnia*. In *Delbergia*, Agarwal and Puri (1977) suggested that the centrally located cells probably act as initials for the peripheral zone exhibiting greater activity. Contribution of cells to the PZ and PM from the CMZ supports the suggestion that the CMZ be considered as the source of cellular structure of the shoot (Newman, 1965).

As reported by others (Popham and Chan, 1950; Philipson, 1954; Fahn *et al.*, 1963; Singh and Singh, 1976), a cambium-like zone has been observed. Fahn *et al.* (1963) termed this zone as "cambium-like transitional zone" and observed as a permanent structural feature of the vegetative shoot apex of banana. In the present report, the cambium-like zone is present in the vegetative and transitional apices during fourth and fifth weeks and agrees with the Philipson's suggestion that at least in some of the plants the cambium-like zone is a specific variation of the central meristem. Its development in the shoot apex of *Zinnia* may be linked with the stem enlargement, particularly with the stem elongation.

Literature Cited

Agarwal, R. M. and V. Puri. 1977. Ontogenetic studies in some important timber trees of India. I. Shoot apex organization and leaf development in *Delbergia sissoo*. Phytomorphology 27: 296-302.

Fahn, A., S. Stoler, and T. First. 1963. Vegetative shoot apex of banana and zonal changes as it becomes reproductive. Bot. Gaz. 124: 246-250.

Mauseth, J. D. 1978a. An investigation of the morphogenetic mechanisms which control the development of zonation in seedling shoot apical meristems. Am. J. Bot. **65**: 158–167.

Mauseth, J. D. 1978b. An investigation of the phylogenetic and ontogenetic variability of shoot apical meristems in Cactaceae. Am. J. Bot. **65**: 326–333.

Newman, I. V. 1965. Pattern in the meristems of the vascular plants. III. Persuing the patterns in the apical meristem where no cell is a permanent cell. J. Linn. Soc. (Bot.) **59**: 185–214.

Paolillo, D. J., Jr. and E. M. Jr. Gifford. 1961. Plastochronic changes and the concept of apical initials in *Ephedra altissima*. Am. J. Bot. **48**: 8–16.

Philipson, W. R. 1954. Organization of the shoot apex in dicotyledons. Phytomorphology **4**: 70–75.

Popham, R. A. and A. P. Chan. 1950. Zonation in the vegetative stem tip of *Chrysenthemum morifolium* Bailey. Am. J. Bot. **37**: 476–484.

Rogan, P. G. and D. L. Smith. 1974. The development of the shoot apex of *Agropyron repens* (L.) Beauv. Ann. Bot. **38**: 967–976.

Singh, A. and V. Singh. 1976. Shoot apex organization and leaf development in *Ricinus communis* L. Proc. Indian Acad. Sci. **83**: 166–176.

Sharma, M. and A. Pillai. 1986. Ontogenetic studies of shoot apical organization in *Raphanus sativus* L. J. Indian Bot. Soc. **65**: 212–218.

百日菊的莖頂組織化的發育

M. Sharma and K. C. Sharma

Department of Botany, University of Rajasthan
Jaipur-302 004, India

本研究探討百日菊（*Zinnia elegans* Jacq.）莖頂分生組織由成熟胚至開花期的組織化過程。胚的莖頂兩側爲一對葉的原生組織，且整個頂端細胞被兩層外囊（tunica）所包圍。根據分析，帶的形成（Zonation）建立於浸種後第五天。間隔期則依頂端細胞和中心母細胞帶大小而異。莖頂細胞經由過渡期進入生殖期。在第四週，無性頂端細胞及過渡期的頂端細胞可看見一類形成層帶（cambium-like zone）。花芽頂端細胞則呈一有核心的組織化，產生了總苞及向上排列的小花原生組織，於是整個頂端細胞分化完成。