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ONTOGENETIC STUDIES OF SHOOT APICAL ORGANISATION IN *SOLANUM MELONGENA*

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Shoot apical organisation in plumular, vegetative, transitional and reproductive apices has been studied. The plumular and vegetative apices showed Tunica-Corpus organisation with a 2- layered tunica covering homogeneous parenchymatous corpus. A clear demarcation between the lighter stained axial and deeply stained peripheral tunica layers was seen. The vegetative apex exhibited cytohistological zonation which was supported by histochemical data and N/C ratios as well. Transitional apex was seen before the vegetative apex was converted into the reproductive apex. Transitional and reproductive apices showed Mantle-core organisation with a 4-5 layered densely stained mantle covering the subjacent lighter core. The entire vegetative apex is consumed in the formation of the reproductie apex.

Keywords : Mantle-core; Plumular; Reproductive; Transitional; Vegetative.

Introduction

Over the years, the structural organisation and development of the shoot apical meristem attracted the attention of many distinguished botanists.[1-3] The present report deals with the apical organisation and behaviour of the shoot apex of *Solanum melongena* Linn. belonging to family Solanaceae, from mature embryo to flowering stage. Localisation of several metabolites at all developmental stages has also been studied.

Materials and Methods

Plumular apex from mature embryos dissected out from the soaked seeds of *Solanum melongena* Linn. and shoot apices from germinating seeds at 24 hour intervals for the first seven days after seed wetting and subsequently from well established plants at one week intervals till flowering were collected and fixed in FAA and Carnoy's solution. These were then washed thoroughly in 70% alcohol followed by dehydration through Tertiary Butyl Alcohol (TBA) series and embedded in paraffin. Sections cut at 5- 7 µm were stained by Northan's variation of Foster's technique using tannic acid-iron chloride, safranine and light green. Some apices were stained with Pyronin-Y for RNA[4], Mercuric bromophenol blue for proteins[5], periodic acid Schiff's (PAS) reaction for insoluble polysaccharide[6] and Feulgen Method for DNA[7].

In median longitudinal sections, width of the apex was taken at the level of youngest visible leaf primordia from the adaxial side and height was measured from the tip of the apical dome to this basal reference point.

At various developmental stages, average values for nuclear and cell areas were calculated from cells of each zone of the apex and the ratios of the nuclear area/cell area (N/C ratio) estimated.

Observations

The plumular apex is a broad dome having an average height of width of 4.8 µm and 27.8 µm respectively. A single layered tunica covers the subjacent corpus which constitutes a homogeneous mass of parenchymatous cells which are irregularly arranged, larger and more vacuolated than tunica cells. At this stage the cells are more or less uniform in size and staining behaviours. No periclinal divisions were observed in the tunica. Tunica-corpus organisation with uniformly stained nuclei in both the zones were seen when stained for DNA. Uniform distribution for RNA and total proteins was also seen in both the zones (Fig. 1 A,B). Nuclear as well as cytoplasmic RNA was observed. The cotyledons and other parts of embryos show considerable quantities of stored proteins (Fig. 1B).

There is a gradual increase in height and width of the shoot apex during the vegetative period. The apex was seen to range from a low to high dome depending on the plastochronic stage. The average height and width measured were 6.8 µm and 27.8 µm respectively.

The actively growing vegetative apex has a 2-layered tunica. The tunica cells at the summit of the dome are more vacuolated as compared to those on the flanks and the term "axial tunica" as used by Molder and Owens[8] is applicable. Cytohistological zonation was established in the first week and characters like planes of cell division, cell size, vacuolation and intensity of staining, the corpus can be demarcated into central mother cell zone (CMZ), peripheral zone (PZ) and the pith meristem (PM).

The CMZ is proximal to the tunica cells at the summit of the apical dome. It is made up of a group of large lightly stained and irregularly arranged cells. Cell packets of 2-3 cells enclosed in a common wall are seen clearly (Fig. 2A). After establishment of zonation the depth of CMZ gradually increases with seedling age and size of apical dome. Variations in the depth of CMZ are noticed in apices at different stages of a plastochron. It is 37.2 µm at the minimal stage and becomes 44.7 µm during maximal stage of plastochron. Cell layers around the CMZ and PM constitute the PZ. The cells are arranged in 4-6 files and are slightly elongated along the long axis of the shoot and densely stained. Predominantly anticlinal divisions have been observed with a few periclinal divisions which result in regularly arranged cell files that broaden proximally and form the site for initiation of lateral primordia. PM is formed as a result of transverse divisions at the base of the CMZ. This is a group of more or less regularly arranged cells subjacent to the CMZ. The pith is formed by further divisions of these cells and differentiation of the products. Vacuolation of the cells increases progressively proximally.

Plastochronic index (PI) has been calculated following Erickson and Michelini[9] formula, PI = Height of apex

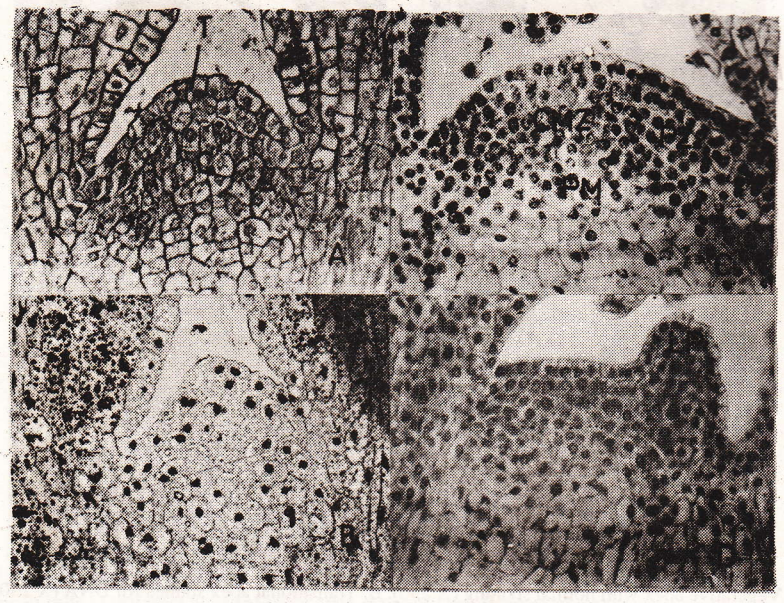

Fig. 1 : A. Plumular apex stained for RNA (X 250); B. Plumular apex stained for total proteins (X 250); C. Vegetative apex stained for DNA (X 250); D. Vegetative apex stained for total proteins (X 250).

in µm/Height of youngest leaf primordium in µm

Minimal phase showed a flat apex with a PI of 0.67, 0.54, at the mid and 0.26 at the maximal phase of the plastochron. The depth of CMZ measured as 37.2 µm, 43.0 µm and 44.7 µm at minimal, mid and maximal phases respectively. No change in number of tunica layers was recorded with change in plastochronic phase.

The staining for DNA, RNA and total proteins brought out a distribution pattern resembling the cytohistological zonation (Figs. 1C, 1D, 2A). The intensity of staining of cell walls for insoluble polysaccharides in various zones of the vegetative apex is uniform. The outer walls of the first tunica layer and epidermis of the developing leaves take darker staining.

Transitional Apex - During the change over from vegetative to the reproductive phase, the dome of the shoot apex broadens and the number of covering layers increases. No periclines were observed in the tunica. So the increase in the covering layers must have resulted from a more orderly and layered arrangement of the distal cells of the CMZ. These alongwith the outer few cell layers of the PZ and the original 2-layered tunica constitute a 4-5 layered 'mantle'. The height and width is maximum during this stage reaching an average of 12.2 µm and 30.8 µm respectively (Fig. 3A). Proximal to the mantle lies the 'core' which is a homogeneous mass of lighter stained, irregularly arranged cells.

The nuclei in the mantle layers stain deeper than those in the core when

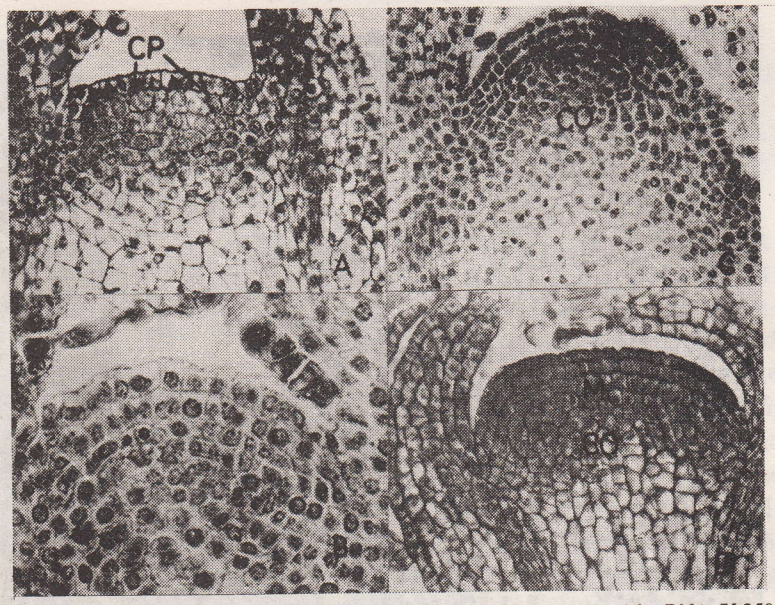

Fig. 2 : A. Vegetative apex with normal staining (X 250). B. Transitional apex stained for DNA (X 250); C. Transitional apex stained for total proteins (X 250); D. Reproductive apex stained for insoluble polysaccharides (X 250).

stained for DNA. Although the nuclei in the axially located cells of the surface layers are comparatively lighter stained than in the peripheral cells (Fig. 2B). A similar lighter axial mantle was observed for RNA. When stained for total proteins and insoluble polysaccharides, uniformly deeply stained mantle layers were seen (Fig. 2C).

Reproductive Apex - Reproductive apex was seen at the 6th week with height and width of 8.6 µm 30.0 µm respectively. A mantle-core organisation is seen clearly with a 4-5 layered dark mantle covering a lighter stained core. Uniformly deeply stained mantle was seen in preparations of DNA, RNA and total proteins. Insoluble polysaccharides, showed deeply stained mantle and lighter core, although the difference in staining is attributed more to the cytoplasmic staining than to difference in wall staining (Fig. 2D). Average Nuclear area/cytoplasmic area (N/C) ratios have been calculated in cells of various zones at different developmental stages of the apex and presented as histogram (Fig. 3B).

Discussion

A gradual age related increase in size of the apical dome is evinced in *Solanum melongena*. The attainment of maximum size is followed by initiation of the reproductive phase.

The N/C ratios in the plumular apex point to a higher meristematic potential in tunica cells. The marked cytohistological zonation in the vegetative apex at the maximal phase of the plastochron can be attributed to the increase in the extent of the CMZ and PZ prior to leaf initiation.

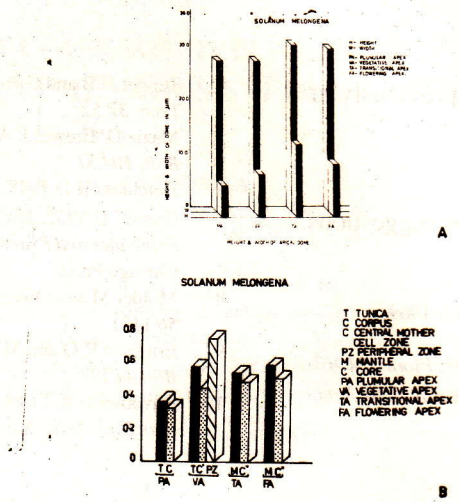

FIG. 3

Fig. 3 : A. Histogram showing height and width of shoot apex at various developmental stages; B. Histogram showing N/C ratios in cells of different zones of shoot apex at various developmental stages.

CP - Cell Packets; LP - Leaf Primordium; PM - Pith Meristem.

The anatomical and histochemical data presented here with reference to cytohistological zonation in the vegetative apex are in general agreement with those by other workers[1,10]. The distribution patterns of nucleic acids and total proteins follows the cytohistological zonation with PZ showing the maximum. The same trend is indicated by the N/C ratios. Regular cyclic fluctuations in the size of CMZ and PZ observed during every plastochron indicate that the CMZ contributes cells to the PZ and is indirectly involved in leaf initiation and the entire vegetative apex is involved in the formation of the reproductive apex.

A transitional apex was observed in the plants grown in season and hence exposed to photoperiods which in nature induce normal flowering under inductive conditions and can be considered as a preparatory step for flowering with the CMZ as the focal point of changes. There seems to be no regular or predictable pattern in the presence or absence of a lighter axial mantle in either the transitional or the reproductive apex regarding the staining pattern of RNA and DNA. It might be related to the siting and development of lateral primordia and consequent deeper staining of the peripheral mantle cells. The significance of the transitional apex seems to be that a time gap appears between the establishment of a mantle-core organisation and the initiation of lateral primordia.

In the reproductive apex, a mantle region is formed over the central core involving the reorganisation of the entire apex and all the zones rather than

the stimulation of a single previously inactive zone.

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