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Floral differentiation and its modification

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Abstract. Flower initiation is an important morphogenetic event. In this brief review the formative, ultrastructural, cytological and biochemical changes that occur in the transitional meristems in a few selected species have been discussed. In the evoked meristems, the number of plastids, mitochondria and ribosomes are usually higher. Further, an early shift of 4C nuclei to the 2C value, an increase in respiration and enhanced activities of dehydrogenase and phosphatase have been observed. The molecular events that ensue immediately after induction need further study.

The factors that regulate flower morphogenesis in vitro and reversal of excised flower buds to vegetative growth have been discussed. Reports on the modification of inflorescence development through the application of growth regulators have been analysed.

Keywords. Barren capitula; *Calendula*; *Chenopodium*; chlorflurenol; *Chrysanthemum*; floral differentiation; floral meristem; floral morphogenesis; floral organogenesis; gibberellic acid; inflorescence; long-day plants; reversal to vegetative growth; short-day plants; *Sinapis*; *Spinacea*; *Tagetes*; ultrastructure; virescent inflorescence; *Xanthium*.

1. Introduction

The transformation of a vegetative shoot apex into a reproductive one is a fundamental morphogenetic event. The causal factors of floral evocation have been studied in several angiosperms (Chailakhyan 1958; Lang 1965; Evans 1969), yet "how the reproductive apex, during its metabolism and growth gives rise to organs of characteristic size, position and symmetry and to the tissue pattern within" (Wardlaw 1965) is far from clear and has remained a subject of deep interest.

Flowering plants display wide variation in flowering response. The vegetative shoot apical meristem itself may be consumed in forming a flower or an inflorescence, or it may continue to form both vegetative structures and flowers.

The study of formative and histological changes in the vegetative and reproductive apices has been pursued by several investigators (Popham 1963; Cutter 1965; Lyndon 1978; Halperin 1978; Wareing 1978; Havelange 1980; Mohan Ram 1980; Bernier *et al* 1981). Precisely-timed measurements of morphological changes at the shoot apex during floral transition indicate that flower formation is usually accompanied or even preceded by several macroscopic changes which are often regarded as "symptoms" of flowering (see Bernier *et al* 1981) such as increased elongation of young internodes; precocious initiation of axillary buds, increased leaf growth rate, change in leaf shape, increased rate of initiation of leaves and primordia, enlargement and doming of the meristem, and increased phyllotaxis. Our knowledge of the cellular and sub-cellular events occurring in the meristems has been greatly enriched using plants such as *Sinapis alba* and *Spinacea oleracea* (long day plants) and *Xanthium strumarium* (short day plant) in which flowering can be induced by providing a single inductive cycle.

2. The floral meristem

2.1 *Ultrastructural changes*

Qualitative and quantitative ultrastructural changes in meristems of *Sinapis* and *Spinacea* have been studied by Havelange and co-workers (1974, 1980) and by Auderset and Greppin (1977). In *Sinapis*, the number of mitochondria per cell increases as early as 18 hr after start of the long day, and at the time of the initiation of the first flower primordia at 62 hr, this number is 2 to 3 times higher than at the start of induction. In *Spinacia* and *Xanthium* both plastids and mitochondria show an increase in their number. In the former the splitting of existing vacuoles has been noted 18 hr after induction. In *Spinacea* dumbbell-shaped or lobed plastids and mitochondria are frequently seen, suggesting that these are dividing. Frequent divisions of proplastids were also observed by Gifford and Stewart (1965) in the transforming meristems of *Chenopodium album*. Furthermore, *de novo* formation of flowers from explants containing multiple cell layers of tobacco is also characterized by an increase in the number of mitochondria (Tran Thanh Van and Trinh 1978).

The concentration of ribosomes is usually greater in the pre-floral meristems than in the vegetative meristems (Lance-Nougarède and Bronchart 1965; Lance-Nougarède 1967; Lin and Gifford 1976) and the complete structural organization of the nuclei is indicative of increased synthesis and export of ribosomal subunits. An increase in endoplasmic reticulum and dictyosome number and activity have been reported in *Sinapis*, *Spinacia*, *Xanthium* and *Chenopodium album* (Gifford and Stewart 1965). The nucleoli of evoked meristems are larger, less compact and more vacuolate, and within the nucleus an increased volume of dispersed chromatin is seen in *Sinapis*. Examination of Feulgen-stained preparations of transforming meristems in *Lolium* and cauliflower show chromatin decondensation (Knox and Evans 1976; Sadik and Ozbun 1967).

2.2 *Cytological changes in the floral meristem*

The most conspicuous event in floral transformation is the appearance of numerous mitotic figures. Using Feulgen cytophotometry to establish the distribution of the 2C and 4C cell populations Jacqmard and Miksche (1971) showed that in the vegetative (intermediate) meristems prior to LD induction, nuclei with 4C amount of DNA (G_2 nuclei) are more numerous than those with 2C DNA content (G_1 nuclei). In the transitional meristems, there is an early shift of 4C nuclei to the 2C value, and this leads to near-synchronization of the cell population at the 2C condition at the 30th hr, *i.e.*, at the completion of the first mitotic wave when 65 to 70 % of the total cell population is in G_1. The mean cell-doubling time in Petkus winter rye and lupin was estimated by Sunderland (1961) from counts of total cell numbers in apices and numbers of cells produced by them during a given time interval. Direct measurements of the cell-doubling time in each zone of transforming meristems can also be achieved by counting metaphase cell accumulation after colchicine treatment. Use of this technique by Corson (1969) in the day-neutral plant (DNP) *Datura* and by Bodson (1975) in the long-day plant (LPD) *Sinapis* reveals that cell-doubling times decrease in both the central and the peripheral zones of the meristem during transition to flowering. During floral transition a cell population in the meristem becomes transiently synchronized. In *Sinapis* there are two mitotic waves during the transition: the first one is clearly

associated with cell synchronization while the second, which occurs concurrently with the onset of flower initiation, is related to the increased rate of cell division (Bernier *et al* 1981). A single application of a low dose of a cytokinin made at midnight to the apical bud of a vegetative plant of *Sinapis alba* produces a mitotic wave similar in every respect to that found in the meristem of plants induced to flower by a single long day (Bernier *et al* 1974). It was, therefore, proposed that cytokinin may be one of the components of the complex floral stimulus in this species (Bernier *et al* 1977).

2.3 *Biochemical changes*

Transforming apices of *Iris* have higher rates of transpiration than their vegetative counterparts. Microrespirometric and histochemical studies by Thein (1957), Krekule and Teltscherova (1966) and Opatrná (1970) in the evoked meristems of both long- and short-day plants show that respiration and dehydrogenase activity increase during passage from the vegetative to the reproductive stage. A histochemical investigation by Jacqmard (1978) has revealed that succinic dehydrogenase activity increases significantly by the 22nd hr after the start of the inductive LD in the meristem of *Sinapis* and coincides with the increase in the size of the condriome found in the evoked meristem (Havelange *et al* 1974). These two events reflect an increase in cellular respiratory activity for the release of energy (Bodson 1977; Bodson *et al* 1977). A marked increase in the number of mitochondria accompanies these changes. Similar observations have been reported in the SDP *Xanthium* (Thein 1957; Havelange 1980). Localisation of glucose-6-phosphate dehydrogenase and 6-phosphogluconic dehydrogenase by Gahan *et al* (1979) in the meristem sections of *Spinacia* indicates an augmentation in the activity of pentose phosphate pathway as a result of floral induction. There is a marked accumulation of starch in all parts of the apex during transition to flowering both in LDP's (Bernier 1971; Bodson 1977), SDP's (Gifford and Tepper 1962) and cold-requiring plants (Sadik and Ozbun 1967). The increase in RNA and protein content can be measured histochemically (Jacqmard *et al* 1972). The concentration of RNA in the apex of *Silene* increases by about 30% during induction as it does in many other plants (Miller and Lyndon 1976). New kinds of proteins specific to floral condition have not been detected (Stiles and Davies 1976). Just one or two new bands have been observed in electrophoretic analysis (Sawhney *et al* 1976). However, Barber and Steward (1968) had noted that the induction of flowering in *Tulipa* led to detectable qualitative protein changes "before the floral organs had developed".

An increase in acid phosphatase activity in evoked meristems of *Sinapis* was detected as early as 14 hr after the start of the inductive long day (Bernier *et al* 1981). This intensity was also found correlated with regions of rapid cell division in other systems (Wilson and Cutter 1955; Fosket and Miksche 1966; Shaykh and Roberts 1974). The increase in ribonuclease activity begins at 18 hr, a few hours after a rise in RNA synthesis in the 1·5 mm apical bud of the same species (Pryke and Bernier 1978). The results concerning acid phosphatase and ribonuclease show that catabolic reactions are also activated during evocation of flowering along with synthetic activities (Jacqmard 1978).

3. Flower morphogenesis in vitro

The technique of culturing young excised flower primordia introduced by LaRue (1942) affords a potentially useful tool for (a) controlling flower morphogenesis

(b) assessing the influence of growth regulators and nutrients on buds in the absence of intervening vegetative tissues; (c) testing the autonomy of the floral apex; and (d) performing a variety of surgical manipulations.

Johri and Ganapathy (1967), Wadhi (1967), Mohan Ram and Jaiswal (1975) and Konar and Kitchlue (1982) have reviewed the literature on flower morphogenesis in vitro. In general when excised immature flower buds are cultured they are able to produce viable pollen grains and ovules in very few instances. The work done by Tepfer *et al* (1963, 1966) on *Aquileqia* has demonstrated that the younger the flower bud at culture, the more elaborate are its growth requirements. Whereas Tepfer *et al* (1966) believed that IAA was necessary for carpel development, Bilderback (1972) failed to demonstrate its requirement. The role of gibberellic acid (GA_3) in the development of calyx, corolla and ovary in the excised immature flower buds in *Viscaria candida* was demonstrated by Blake (1966). In the cultured immature inflorescences of *Cyperus rotundus*, Mohan Ram and Batra (1970) noted that cytokinins promoted the origin and development of new flowers. Hicks and Sussex (1970) observed that initiation and early growth of organ primordia are independent of exogenously applied hormones, but that kinetin plays a key role in enhancing the eventual development of all the organ primordia.

The number of examples in which altered morphogenetic responses have been noted in the cultured immature flower buds are numerous. Callusing, production of roots, shoots or entire plantlets through differentiation of embryoids have been reported by Konar and Nataraja (1964), Konar and Konar (1966), Mohan Ram and Wadhi (1966), and Ganapathy (1969). Greyson and Raman (1975) studied *Nigella damascena* using genotypically dissimilar 'single' and 'double' flowers, the 'double' being inherited as a single gene recessive. Floral apices when cultured on Murashige and Skoog's (MS) medium supplemented with kinetin (Kn) supported stamen and carpel initiation in both the genotypes, whereas 'single' could do so on MS medium alone. In the medium minus (Kn), GA_3 was essential for organ initiation in the 'double', but it totally inhibited stamen initiation in the 'singles'. Raman and Greyson (1978) further observed that GA_3 inhibited the initiation and growth of stamens and nectaries in 'singles'. The use of IAA or Kn in any combination failed to overcome the inhibitory action of GA_3 on 'singles'. These authors have also succeeded in obtaining graft unions between bisected floral meristems of the two forms (Raman and Greyson 1977).

A rather difficult problem to tackle is the spatial relationship in regard to the origin of organ primordia in the floral apex. At what point of ontogeny does the specific pattern of organ primordia become blocked in the nascent meristem? If it is conceived that the floral apex is a mosaic of irrevocably determined sites of different organs, the removal of a particular part of the flower should cause an irreparable loss of the part (Heslop-Harrison 1972). It could also be presumed alternatively, that the sequences of organ initiation is not predetermined and the terminal tissue of a floral meristem is capable of replacing the lost parts (Mohan Ram and Jaiswal 1975). Experiments dealing with bisected and cultured immature flower buds have shown that floral organs are able to regenerate even at the cut surfaces (Jensen 1971). Hicks (1972) studied the fate of bisected stamen primordia at different developing stages while they were still attached to the floral apex. Remarkably enough, he observed that two stamens, each containing four anther lobes developed from a bisected young stamen primordium. However, such regeneration was not possible with older stamens. Whereas GA_3 causes the production of stamen in the stamenless-2

prefloral tomato mutants (Sawhney and Greyson 1973), the work of Hicks and Sand (1977) has established that undifferentiated stamen primordia of a male sterile tobacco hybrid in vitro are capable of relatively autonomous development and are not influenced by the hormone.

3.1 *Stability or instability of the floral state in vitro*

Inflorescence segments of day-neutral tobacco plants (DNP's) regenerate flower primordia in vitro, whereas inflorescence segments of daylength-sensitive tobacco varieties produce only vegetative buds. This differential behaviour was explained by Chailakhyan *et al* (1975) in the following manner: in DNP's all components of the floral hormone complex are synthesized in all plant parts independently of daylength and are present in explants of inflorescence axes which can thus initiate flowers in vitro. On the contrary, in daylength-sensitive plants the hormonal complex is believed to be formed only in leaves that are exposed to the appropriate photoperiod. In tissue explants excised from the inflorescence of these plants, the hormonal complex is rapidly exhausted due to absence of leaf tissues and only vegetative buds are regenerated. As predicted by Chailakhyan, inflorescence fragments of the SDP *Streptocarpus nobilis* produce only vegetative buds in vitro even when cultivated in inductive SD, whereas discs collected from leaves regenerate flower buds in SD, but not in LD (Rossini 1970).

4. Reversion of reproductive apices to vegetative growth

In nature vegetative structures may arise spontaneously within positions of flowers in many species. These malformations were termed "teratisms" by morphologists. With the discovery that the passage of plants to the reproductive condition is frequently governed by certain environmental parameters, physiologists succeeded in producing such "chimeric" flowers (or inflorescences) (Von Witsch 1965). Usually such structures appear in some photoperiodic and cold-requiring species given marginal, interrupted, or otherwise perturbed induction. Occurrence of a graded series or vegetative-reproductive interphases suggests that apices that have reached the realization (reproduction) stage can revert to a more or less typical vegetative condition (Von Witsch 1965).

When a transitional meristem of *Petasites* presumably at the early prefloral stage is bisected, punctured at the centre, or isolated from adjacent subapical tissues by vertical incisions, the small, simple bracts develop into large, petiolate and laminate leaves, and no florets are initiated. At later stages of capitulum development similar surgical treatments no longer impede flower formation (Wardlaw 1963).

4.1 *Reversal of floral explants to vegetative condition*

The development of leafy shoots from excised flower buds is extremely rare. LaRue (1942) observed the occurrence of leafy shoots in cultured floral buds in two species (*Kalanchoe globulifera* and *Nemesis strumosa*) out of 92 spp of angiosperms. According to him the shoot buds had been formed prior to culture. Mohan Ram and Wadhi (1966, 1968) reported reversion from floral to vegetative condition in *Kalanchoe pinnata*. These authors cultured flower buds of two early stages of development on modified

White's medium with supplements. The buds failed to attain full development and the primordia of floral organs lost their ability for normal morphogenesis under culture conditions. The authors attributed the phenomenon of reversion to vegetative growth in *Kalanchoe* to early excision, which apparently hinders the normal sequence of floral morphogenesis.

4.2 *Inflorescence reversions*

Inflorescence reversions, *i.e.*, inflorescences that have acquired in their last-formed portions several or all characters of vegetative shoots, are known to occur spontaneously in plants such as *Ananas* and *Lavandula stoechas*. Experimental reversion of the inflorescence can be brought about in the Labiatae such as *Perilla* and *Salvia*, by transfer of these plants to unfavourable daylength regimes, but only if induction is suboptimal (see Bernier *et al* 1981). Reversions also commonly occur in *Nicotiana glutinosa* and *Hyoscyamus* (Diomaiuto-Bonnand 1969; Seidlova and Stichova 1965), brussel sprouts (Stokes and Verkerk 1951), *Sinapis* (Bagnard *et al* 1972; Bagnard 1978) and *Cheiranthus cheirii* (Diomaiuto-Bonnand 1972). Inflorescence proliferation may also occur in some grasses as a result of incomplete induction (Evans 1964). These morphological anomalies can be traced to spikelet proliferation, *i.e.* spikelets with an apical meristem continuing growth and producing a leafy shoot, and are consequently due to failure of determination in spikelet primordia which are uncommitted at initiation (Evans 1964).

5. Special cases

An ontogenetic study of reproductive structures in cauliflower (*Brassica oleracea* var. *botrytis*) by Sadik and Ozbun (1967) has revealed that the apical vegetative meristem is first transformed into a typical prefloral meristem, similar to the prefloral meristem of *Sinapis* or other crucifers, with precocious initiation of axillary meristems. In cauliflower, however, the prefloral stage is long-lasting, and the process of branching is enormously amplified. With time, second-order axillary meristems are initiated around meristems of first order branches, and later, third- and higher-order axillary meristems are initiated. This process continues until a very large number of meristems and branches accumulate and form the "curd". Although these meristems do not differentiate into flower primordia, the curd is obviously no longer a vegetative structure; rather it is an amplified prefloral inflorescence axis (Sadik and Ozbun 1967).

In some species such as *Phytolacca decandra* and tomato with sympodial growth, inflorescences occupy neither terminal nor strictly axillary positions. In these plants, the shoot apical meristem is normally transformed into a reproductive meristem after a certain period of vegetative functioning. Concomitantly with this transformation, vegetative growth is resumed by the development of a nearby vegetative meristem which assumes terminal position: now the initial reproductive meristem appears in a lateral position. After formation of a number of leaves, the new terminal meristem flowers and is in turn displaced laterally by a new vegetative meristem soon (Lance-Nougaréde and Rondet 1957).

In grapevine (*Vitis vinifera*) inflorescence primordia also arise in specialized axillary buds, the so-called "bourgeons latens" (Srinivasan and Mullins 1979). The first step in

the formation of inflorescences in these buds is the initiation of an extra-lateral meristem called "anlage" by the apical meristem. The anlage rapidly produces a bract on its abaxial side and then another lateral meristem on the opposite side. This adaxial meristem or "inner arm" becomes the main body of the inflorescence primordium if conditions are conducive to flowering, and the "outer arm" (adjacent to the bract) develops into the lower branch of the inflorescence primordium. These specialized buds of grapevine are apparently irreversibly evoked and committed to determinate growth early in their ontogeny. If conditions are not favourable to flowering, the two arms do not undergo branching, and the anlage then develops into tendrils which, like thorns are typically determinate structures (Srinivasan and Mullins 1979).

6. Floral organogenesis

Various contradictory conceptual models have been proposed for interpreting the flower (see Sattler 1965; Meeuse 1966; Eyde 1975). One such model explains the flower as a modified monaxial shoot (Eames 1961; Von Bartalanffy 1965; Cronquist 1968; Takhtajan 1969). According to another model, the basic unit of flower is the gonophyll (Melville 1962–1963). Meeuse (1966) uses the model of an anthocorm and gonoclad for the interpretation of flowers while Croizat (1960, 1962, 1964) thinks of a flower in a less rigid way as an axis with scales, some or all of which are sexualized. He distinguishes two types of flowers—the panstrobilar and the circumnucellar.

Present day developmental morphology lays emphasis on causal interrelations. This aspect of enquiry necessarily leads to the study of lower levels of organization such as cells, organelles, and macromolecules. As a result, cell biology and molecular biology have become dominant fields of enquiry, relegating comparative studies of organs, whole organisms, and their development to the background. Sattler (1973) emphasizes how much is yet to be learnt from comparative organogenesis of an almost ubiquitous organ-system such as the flower.

Two techniques have been used to study floral organogenesis. One is the familiar microtechnique (Johansen 1940; Feder and O'Brien 1968) which involves serial sectioning of floral buds and their subsequent reconstruction. The second is the dissection of floral buds and their photography under alcohol, using close-up lenses (Sattler 1968). Whereas sections reveal cytological and histological details, their utility in obtaining a geometrical picture is limited. The second technique offers a direct three-dimensional view of the floral buds, their primordial appendages and protodermal cells, but no internal tissue details. The limits of observation have been extended through the use of SEM, in which the minutest details of formative changes can be traced. The scanning pictures permit exact measurements of phyllotactic parameters in the young flower (Lyndon 1978).

7. Development of the capitulum and its modification

The capitulum of the Compositae has been an object of biological curiosity because of the compaction of florets on the axis with a centripetal diversification. There is an enormous increase in the area that the shoot apex undergoes in forming a capitulum [nearly 400-fold in *Chrysanthemum* (Schwabe 1959)] and in the differentiation of a multitude of floret growth centres in the place of a single vegetative meristem. The

capitulum of sunflower, which simulates a single flower, is probably the largest inflorescence (Heiser in 1976 recorded a 'two and one-half feet' sunflower) in the family and also the most complicated in phyllotaxis, with innumerable intersecting spirals of florets (Leppik 1977). Details of the topographical and floral organogenetic changes occurring during the transformation of a vegetative meristem into a capitulum have been studied by Popham and Chan (1952) in *Chrysanthemum.* In *Calendula officinalis*, chlorflurenol—a morphactin—suppresses the development of involucral bracts to various degrees, prevents full expansion of the thalamus and either inhibits the initiation of the florets or interferes with the completion of development of those florets which are already differentiated. Total elimination of only disc florets, or ray florets or a reduction in the number and size of the florets, leaving a vacant surface of the thalamus, either in the centre or along the periphery or between the florets have also been observed in *Calendula* and *Tagetes* (Mohan Ram and Mehta 1973; Mehta 1976; Chandra 1982). Various degrees of fusion of florets have been observed.

Chlorflurenol drastically retards the size of the capitula, by inhibiting the mitotic activity of the tunica, pith rib meristem and intercalary regions, and poor differentiation of the procambium in *Calendula officinalis.* Meristematic activity is also absent in barren capitula of *Tagetes.*

Bennink (1973) showed that under long days, all the initiated floral buds develop into "crown buds" each consisting of a capitulum and bracts without flower primordia, reminiscent of the barren capitula induced by chlorflurenol in *Calendula.* When a solution of benzylandenine (BA) was injected into the plants under long days, it induced the formation of normal flowers (Bennink 1973).

The barren capitula in both *Calendula* and *Tagetes* are rich in starch which may be a consequence of growth inhibition. However, cell division, cell expansion and morphogenetic activity inherent in a meristem are not irreversibly inhibited by chlorflurenol. When the barren capitula are cultured on a medium supplemented with IAA + GA_3 + Kn, mitotic activity resumed in the tunica, pith rib and intercalary regions. Structures resembling floret primordia and vegetative shoot buds were also initiated on the surface of the barren capitula in *Calendula* (Mehta and Mohan Ram 1978). In *Tagetes*, the barren capitula cultured on B_5 medium + BAP (10^{-5} M) callused from the cut end as well as from the surface and subsequently gave rise to shoot buds which could be excised and rooted to obtain flowering plants. Occasionally structures intermediate between shoot and florets appeared directly from the surface of the cultured capitulum and via a callus in *Tagetes* (Mohan Ram and Mehta 1982).

Regeneration of plantlets from fully developed capitula of *Gerbera jamesonii* has been reported by Pierik *et al* (1973, 1975). In these cultures, shoot buds developed from the base of the involucral bracts or from the receptacle. Development of adventitious shoots from the receptacle, remnants of disc florets, axils of disc florets or involucral bracts was reported in *Chrysanthemum cinerariaefolium* (Roest and Bokelmann 1973).

According to the classical concept, the flower is a metamorphosed vegetative shoot of limited growth in which the floral leaves arise in close succession either in spirals or cycles, the meristem being generally used up in their formation. Even botanists of the previous century had observed that in some varieties of roses and pears a secondary flower grew out from the centre of the first flower. This indicated that under certain conditions the floral meristem was not consumed but continued to grow. Occasional occurrence of secondary inflorescences has also been noted in certain members of the Compositae and the Dipsacaceae. Schwabe (1951) reported several teratological

features in *Chrysanthemum* such as formation of bracts on the receptacle as well as secondary inflorescences and petaloid stamens in the inflorescences of plants exposed to extended periods in long days. Bose and Nitsch (1970) reported that gibberellic acid causes this phenomenon in *Calendula officinalis*. Mohan Ram and Mehta (1978) also showed that GA_3 stimulated the formation of secondary and even tertiary inflorescences. Thus gibberellins are involved in the transformation of a floret primordium with a committed and limited developmental plan into a primordium of a secondary inflorescence with a greater morphogenetic potentiality. The peripheral florets, particularly the ray florets, appear to be labile in this respect.

In *Calendula officinalis*, a curious phenomenon observed under Delhi conditions late in the flowering season (April–May onwards, marked by lengthening of the photoperiod from 10–12 hr and a sudden rise in the mean day temperature (from about 20°C to 35 ± 5°C) was the replacement of the colourful capitula by green witches' broom-like structures termed "virescent inflorescences". This is manifested by the development of vegetative shoots from the base of the ovary in place of the ovule in the florets. These penetrate the ovary wall and emerge as leafy shoots (Rao and Mohan Ram 1984). It is also believed that mycoplasma-like organisms (MLO's) are able to cause virescent capitula in the Compositae (S Misra and A Varma, personal communication).

There are numerous instances in the literature of the development of bulbils from flower buds (*Agave*, *Yucca*, *Nymphaea*, *Allium*, etc.) although the causal factors are not established. Dúpuy (1963) and Guédés and Dúpuy (1979) have studied in detail the teratological modifications in the flowers of several genera.

8. Conclusions

Although earlier studies relating to reproductive development were mainly restricted to observational changes at the morphological or histological level, application of recent techniques of histochemistry, biochemistry, electron microscopy, and tissue and organ culture have enabled a better understanding of the causal aspects of floral development. The molecular events of floral evocation are still not fully known. The role of new messenger RNA's, and the changes in protein composition during floral transition of the meristem and whether the control of these changes is at the transcriptional level, the translational level, or beyond are not yet understood. Experimental modification of differentiation has demonstrated the role of environmental factors and growth regulators in determining or reversing the events of floral developments.

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