Chapter 25

HORMONAL INFLUENCES ON FIBER DEVELOPMENT

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INTRODUCTION

Cotton fiber differentiation and development involves numerous biochemical, physiological and morphological changes. Many of these changes are discussed in detail elsewhere in this book (Chapters 20, 22, 23, 26 and others) and need not be repeated here. The phenotypic expression is determined by genotype, but the detailed development of fiber is determined by a combination of internal stimuli, hormonal inputs and external influences. Hormonal work with fibers has been concerned primarily with the two major phases of development, elongation and secondary wall deposition.

Some of the roles of hormones in the mechanisms of cell differentiation and growth have been documented and major progress has been made in characterizing their biosynthesis and metabolism; however, the exact mechanisms of action and interactions still are not very clear (Jones, 1973; Cleland, 1977; Osborne, 1978; Chailakhyan, 1979; and others). Numerous researchers have contributed to our knowledge of the hormonal influences on cotton fiber development (Beasley, 1973; Beasley and Ting, 1973, 1974; Baert *et al.*, 1975; Singh and Singh, 1975; Dhindsa *et al.*, 1976; Kosmidou, 1976; Jasdanwala *et al.*, 1977; Dhindsa 1978a,b; DeLanghe *et al.*, 1978). Also, much is owed to the laboratories which first successfully cultured *in vitro* cotton ovules with associated fibers (Beasley, 1971; DeLanghe and Eid, 1971).

METHODS USED IN THE HORMONAL RESEARCH

The work done so far in cotton regarding hormonal effects has taken two approaches: (1) isolation, identification and measurement of hormones from bolls and ovules; and (2) external application of hormones *in situ* and *in vitro*.

A number of research workers have isolated gibberellins (GA), auxins (IAA, NAA), cytokinins (CK), abscisic acid (ABA) and ethylene (ETH) from cotton bolls and/or ovules with the associated fibers at different boll growth stages

(Carns, 1958; Addicott *et al.*, 1964; Dale and Milford, 1965; Sandstedt, 1971; Bhardwaj and Dua, 1972; Davis and Addicott, 1972; DeLanghe and Verneulen, 1972; Lipe and Morgan, 1973; Shindy and Smith, 1975; Guinn *et al.*, 1978; Rodgers, 1980, 1981a,b,c; Guinn, 1976, 1977, 1982). In most cases, if not all, the research was aimed at purposes other than correlation with fiber development, and provided little information about hormonal evolution in fiber alone. This work was valuable in establishing correlations with plant/boll growth stages, environmental and physiological relations, etc. Also, it helped to a certain degree with the investigations of hormonal influences on fiber development by giving a picture of the fluctuations of hormones occurring in the ovule, the developing embryo and the fibers.

As far as external application of hormones is concerned, two approaches have been followed: (1) treatment of intact plant with hormones; and (2) hormones administered to organ cultures *in vitro*.

Numerous investigations have been conducted to influence boll growth and consequently fiber quality by applying hormones and growth regulators exogenously to intact plants using a variety of techniques, e.g. spraying the whole plant, soaking the ovaries, or injecting hormones into the bolls (Bazanova, 1966; Bhardwaj and Sharma, 1971; Bhatt and Ramanujam, 1971; Bhatt *et al.*, 1972; Zur *et al.*, 1972; Yue-Quing *et al.*, 1980a,b; Kosmidou, 1981). Although plants often respond differently to growth regulators in field tests than to trials in greenhouses or growth rooms (Morgan 1980), in most cases the final goal is the beneficial and practical use of plant growth regulators in cotton production. Extending the duration of fiber elongation or secondary wall thickening and promoting the translocation of photosynthates to the fruits might significantly improve either the total yield or quality of the fiber. There are certain limitations, though, in applicability of this kind of approach because of the complexity of the whole plant system and the difficulty to eliminate or distinguish the effect of other factors which influence fiber growth. Processes like pollination, pollen tube growth, ovule fertilization, embryo and endosperm growth affect, to some extent, the internal hormonal inputs as well as fiber initiation and/or growth. The external application of hormones changes the internal hormonal level and balance, which, combined with the other internal and external parameters, may lead to artifacts. Failure to obtain yield increases from field application of plant growth substances may also be due in part to the fact that the indeterminacy of cotton's flowering habit almost precludes that the hormone(s) would be available at the required site at the time the fiber might have been favorably responsive (Beasley *et al.*, 1974). Consequently, information obtained from this kind of approach about the hormonal action on fiber growth is rather limited. Still, the mechanical prevention of flower pollination, and therefore ovule fertilization, which otherwise would cause boll shedding without externally applied hormones, does provide a suitable system for certain hormonal studies on fiber growth *in situ*. The appropriate

application of growth regulators (0.1 ml GA_3 10^{-2}M with or without 0.1 ml 2,4-D 10^{-3}M per flower) after emasculation of the flower prevents shedding, and nearly normal boll and fiber growth occurs (Kosmidou, 1976).

The complexity due to plant participation in the *in situ* system is eliminated when isolated ovules and fibers are cultured *in vitro*. Recently a new technique has been developed in which polyethylene glycol could protect cellulose synthesis in detached cotton fibers (Carpita and Delmer, 1980). This technique may offer opportunities for hormonal studies even in detached fibers *in vitro*. The *in vitro* environment can be controlled completely, the exogenous influences can be eliminated, the nutrient as well as other requirements can be fulfilled, and the influencing factors can be identified and manipulated. Moreover, the *in vitro* cultures are fairly homogenous and readily obtainable in quantity. The contribution of certain laboratories in establishing the basic procedures for plant material production—aseptic transfer, appropriate nutrient media, environmental factors, etc.—was significant (Joshi, 1960; Mauney, 1961; Beasley, 1971; DeLanghe and Eid, 1971). Especially important in fiber development were the contributions from Beasley's (1971) laboratory. Most of the information available today about the hormonal influences on fiber growth are due to the research procedures which used his system. Fertilized and unfertilized ovules isolated from the mother plant at different stages—preanthesis, at anthesis, and later—have been cultured (and precultured for sequential hormone application) in media supplemented with several hormone combinations (Beasley, 1973; Beasley and Ting, 1973, 1974; Beasley *et al.*, 1974; Baert *et al.*, 1975; Kosmidou, 1976; Dhindsa *et al.*, 1976; Dhindsa 1978a,b; DeLanghe *et al.*, 1978).

EFFECT OF HORMONES ON FIBER INITIATION

HORMONAL INFLUENCES ON FIBER DIFFERENTIATION

There is evidence that gibberellins and auxins are important factors for fiber differentiation (Beasley and Ting, 1973, 1974; Kosmidou, 1976; DeLanghe *et al.*, 1978). It is well established that numerous ovule epidermal cells are differentiated into fiber initials on the morning of anthesis or later. The fiber primordial cells are first recognizable at about 16 hours preanthesis by the accumulation of phenolic substances ("dark" cells) and the higher cytoplasmic density attributed to a higher number of ribosomes (Ramsey and Berlin, 1976a,b; see also Chapter 26). It has been suggested that these phenolic substances are related to auxin metabolism and consequently may be involved in the initiation of cotton fiber differentiation. This is consistent with another report (Popova *et al.*, 1979) according to which larger amounts of phenolic substances are synthesized in ovules showing intensive fiber formation and elongation (early varieties) but none in ovules of a naked-seed type. Increase of o-diphenol oxidase activity occurred during fiber initiation (Naithani *et al.*, 1981), and the authors suggested that a shift in redox balance towards oxidation may play an important role in fiber

initiation. This hypothesis follows the general redox model of the mechanism of auxin action proposed by Arnison (1980), in which emphasis is given on the importance of phenolics as "auxin protector" substances and peroxidases as "auxin destructors." Stewart (1975), in explaining the directional growth of the fibers on a single ovule and the delay of initiation in the micropylar region, proposed either a hormonal stimulus to initiation originating at the crest of the funiculus and migrating towards the micropylar end along the polar alignment of the epidermal cells, or an inhibitor at the micropyle.

Unfertilized ovules isolated early on the day of anthesis before flower pollination and cultured *in vitro* did not produce fibers unless grown on medium supplemented with GA_3 and/or auxin (Kosmidou, 1976). Differences among three hormonal treatments were found in number of initials per ovule and in the elongation following initiation. Gibberellins (GA_3 at $3x10^{-6}M$) could induce the initiation of a normal number of initials (compared to *in situ*), but it could not stimulate as much elongation as when an auxin was also included in the medium. Auxin alone (2,4-D $3x10^{-6}M$ or IAA $10^{-6}M$) could induce the initiation of fewer fiber initials per ovule than GA_3 could, but the subsequent elongation of the initials was greater (Kosmidou, 1976). The necessity of GA_3 during initiation was also shown in another study (Beasley, 1976) in which exposure to GA_3 for only a relatively short time (24 hours) before transfer to IAA was necessary for production of equal or nearly equal amounts of fiber compared to cultures with continual presence of both hormones. However, the amount of fiber produced by the end of the culture period decreased as the length of GA_3 preculture increased from 24 to 48 and 72 hours, indicating the need of IAA in addition to GA_3 during the very first days of fiber initiation and growth. Coinciding with and strengthening the *in vitro* results were the appreciable amounts of gibberellin-like substances found at anthesis in cotton fruit extracts (Rodgers, 1981c) and an auxin activity at anthesis which increased to a peak at day 3, decreased at day 5, and thereafter increased through 7-15 days (Rodgers, 1981a). Temperature and ammonium influence the response of unfertilized ovules to IAA as well as to GA_3 and IAA in combination in producing fibers *in vitro* (Beasley, 1977a). The influence of these three factors (NH_4^+, temperature, hormones) was independent of the availability of reduced nitrogen as a general substrate for growth (Beasley *et al.*, 1979). Despite the current information on the importance of gibberellin and auxin in initiation and growth, more research needs to be done. Perhaps the difference between time of initiation for fuzz and lint fibers, the relatively distinct differences in length of the two fiber types and the much smaller nucleoli of fuzz fibers (see below), are due to sequential "perception" and relative amounts of effective endogenous auxins and gibberellins (Beasley, 1977b).

FIBER NUCLEOLAR EVOLUTION AND HORMONES

During fiber differentiation and subsequent expansion the nucleus enlarges and the usually single nucleolus (Nu) undergoes spectacular changes in size and

structure (Figure 1) indicative of increased capacity for ribosome synthesis (Kosmidou, 1976; Ramsey and Berlin, 1976a,b; Waterkeyn, 1978). Under natural conditions the general pattern of the nucleolar evolution in terms of size and appearance of light inclusions—the so-called vacuoles—in the differentiated fibers is rather unique. First, there is an exponential expansion in size starting at initiation and lasting for 4-8 days following anthesis. This is accompanied by a moderate to high vacuolation which may occur early but usually later in this period. A maximum nucleolar size of 8-11 μm is obtained. After a short stage of maintenance of the maximum size, a rapid decline of Nu-material follows which slows afterwards so that the nucleolus reaches a minimum size at the end of fiber growth.

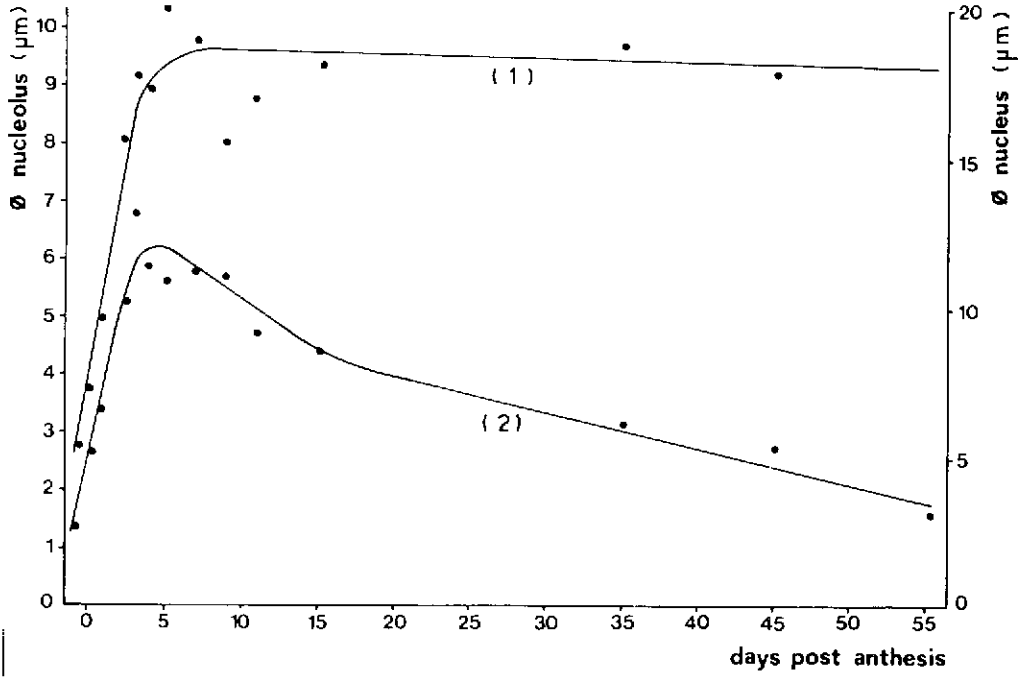

Figure 1. Evolution of nuclear (1) and nucleolar (2) sizes (diam. in μm on y axis) during fiber cell initiation, elongation, and wall thickening (days postanthesis on x axis). *Gossypium hirsutum* cv B49.

During the first stage of nucleolar expansion there is a correlation between Nu-size and length of initials (DeLanghe *et al.*, 1978) indicating that the development of the nucleolus may be related to the manner of elongation and the resulting fiber length. The nucleolar vacuolation observed during this stage (Vla-

sova, 1971; Ramsey and Berlin, 1976a; Kosmidou, 1976; DeLanghe *et al.*, 1978) and its causatives are poorly understood. The hypothesis that a rapid transport of nucleolar material caused by the high demand for protein synthesis during this period gained support by some investigators (De Barsy *et al.*, 1974; DeLanghe *et al.*, 1978), although there could be other reasons.

Vacuolated nucleoli have been described in other tissues and species (Barlow, 1970; Deltour and Bronchant, 1971; Rose *et al.*, 1972; Galan-Cano *et al.*, 1975; Moreno-Diaz *et al.*, 1980), but there has been no agreement on their function. Some authors suggest a correlation between nucleolar vacuolation and nucleolar activity. Others extend this suggestion and conclude that the vacuolated nucleolus provides a storing and/or transporting mechanism for nucleolar products (Moreno-Diaz *et al.*, 1980), but there are also opposing opinions (Rose *et al.*, 1972). However, the appearance of vacuolated nucleoli during periods of high metabolic activity of the cell and the modification of nucleolar vacuolation and size by hormones (Kosmidou, 1976; DeLanghe *et al.*, 1978) support the possibility that some processes under hormonal control in the developing fiber are determined by this early nucleolar activity. Late initials on the ovule (4-12 days postanthesis), which give rise to fuzz fibers (about 2 mm in length), always have small nucleoli without vacuoles (Kosmidou, 1976). Fuzz fibers also have small nuclei (Vlasova, 1971).

Differences among cotton varieties occur in the time required to reach maximum nucleolar size, the absolute size, and the rate and time of vacuolation. Differences also occur among ovules in the same boll according to their location within the boll (top to bottom of the ovary) and among locations on the ovule (chalazal to middle part). Fiber nucleoli of ovules at the top of the ovary increased their material earlier and faster than those at the base of the ovary. The same happens to nucleoli in fibers located at the chalazal end of the ovule compared with those at the middle. However, in all cases the distribution of Nu-size around the mean value of the sample was always normally distributed (*G. barbadense* cv Menufi; unpublished data by Hilde Vincke and E.A.L. DeLanghe, Kath. University of Leuven, personnal communication). This is probably a reflection of polar transport within the boll. Nevertheless the general pattern of fiber nucleolar development remains typical as described before. This pattern also suggests that most of the ribosome synthesis necessary for cell elongation and development occurs early.

Pollination seems to be a stimulus for Nu-material increase and fiber initiation, since its prevention *in situ* causes delay in fiber initiation, smaller nucleoli and shorter fiber initials. A quick decline of Nu-material and cessation of growth occurs, followed by boll abscission (Kosmidou, 1976). According to Beasley (1973) an extract of germinating pollen can mimic quantitatively the effect of exogenous hormones, including slight fiber elongation in unfertilized ovules *in vitro*. Unfertilized ovules isolated before flower pollination and cultured *in vitro*

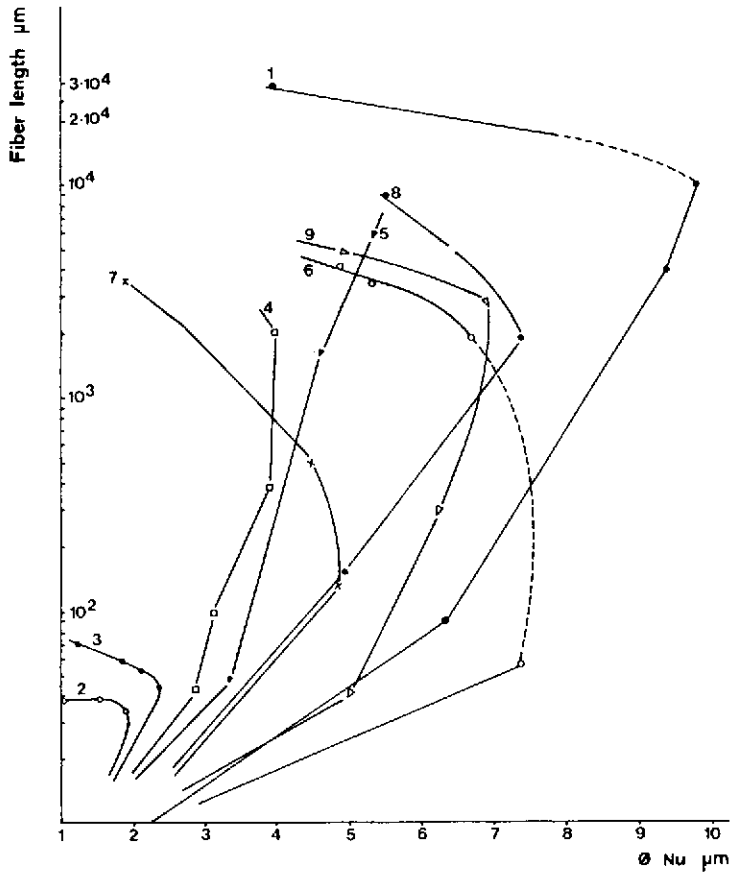

Figure 2. Combined evolution of fiber nucleolar size (diam. in μm on x axis) and fiber length (in μm on y axis—logarithmic) *in situ* and *in vitro*. *Gossypium hirsutum* cv B49.

1 in natural conditions
2 *in vitro*, unfertilized ovules, no hormone
3 *in vitro*, unfertilized ovules, $+GA_3$
4 *in vitro*, unfertilized ovules, + 2,4-D
5 *in vitro*, unfertilized ovules, $+GA_3$ + 2,4-D
6 *in situ* unfertilized ovules, + GA_3
7 *in situ* unfertilized ovules, +2,4-D
8 *in situ* unfertilized ovules, + GA_3 + 2,4-D
9 *in situ* unfertilized ovules, + GA_3 + 24 hrs later 2,4-D
(measurements at 2, 5, 8 and days postanthesis)

without hormones did not initiate fiber except for a few initials near the chalaza. These had very small nucleoli (maximum diam. 1.9 μm) which did not increase in size (Kosmidou, 1976).

Gibberellin (GA_3), auxin and ABA each, separately or in combinations, could significantly affect the nucleolar evolution and the corresponding fiber length (Figure 2), especially in unfertilized ovules *in situ* and *in vitro* during fiber differentiation, first initiation and early elongation (Kosmidou, 1976). Gibberellin (GA_3) stimulated the synthesis of dense Nu-material and prevented nucleolar vacuolation. Auxins (IAA, 2,4-D, NAA) also stimulated the increase of nucleolar size, though not when their level was high, and not *in situ*. High auxin level and ABA caused an early high Nu-vacuolation and "ring-shaped" nucleoli. In these nucleoli a large vacuole occupied the center of the nucleolus while the surrounding dense material formed a cortex. Ovules with high percentage of early "ring-shaped" fiber nucleoli, such as those treated with a high concentration of auxin or ABA, ceased their fiber growth after a very short period of rapid elongation. High concentrations of auxin are known to induce the synthesis of ethylene (Rappaport, 1980; Zurfluh and Guilfoyle, 1982), so interference of this hormone cannot be excluded. Under natural conditions (fertilized ovules *in situ*), nucleolar vacuolation was correlated with intense ribosome production and increased fiber elongation rates (Kosmidou, 1976). The effect of moderate auxin treatment on cotton fiber nucleoli resembles that found in other tissues (Guilfoyle *et al.*, 1975; Grierson *et al.*, 1980) in which auxin induced a large increase in RNA synthesis by nucleoli without affecting the properties of the RNA produced. The mechanism of this auxin action is not clear.

A ratio of GA_3:auxin in favor of auxin also stimulated early nucleolar vacuolation, higher rate of early fiber elongation and smaller nucleoli. The ratio of GA_3:ABA seemed important too, as ABA could counteract the GA_3 effect. ABA may inhibit or even block the synthesis of Nu-material and may eventually participate, like auxin, in ribosome transport and utilization (DeLanghe *et al.*, 1978). The hormonal balance of auxin, GA and ABA seems to be important for the regulation of synthesis and release of nucleolar material. Fiber elongation is related to these phenomena; however, as long as we do not have complete information about the way in which rRNA production is regulated, many questions will remain.

EFFECT OF HORMONES ON FIBER ELONGATION

Lint fibers begin their elongation phase at anthesis. Depending on genetic and environmental factors, the elongation phase lasts for about 20-25 days, by which time the fibers attain a maximum length. The maximum growth rate occurs around the 8th to the 10th day postanthesis.

Many reports indicate that gibberellins, auxins and abscisic acid affect fiber elongation, the first two inducing and the third inhibiting it (Bhardwaj and

Sharma, 1971; Beasley, 1973; Beasley and Ting, 1973, 1974; Singh and Singh, 1975; Baert *et al.*, 1975; Dhindsa *et al.*, 1976; Kosmidou, 1976; Babaev and Agakishiev, 1977; Bazanova, 1977; DeLanghe *et al.*, 1978). When treated with GA_3 (0. 1 ml 10^{-2}M/flower), auxins (IAA or 2,4-D 0.1 ml 10^{-3}M/flower) and ABA (0.1 ml 10^{-3}M/flower), each separately or in combinations on the morning of anthesis, fibers of non-fertilized ovules *in situ* elongated with different patterns which influenced the final length distribution (Figure 3). GA_3, besides preventing

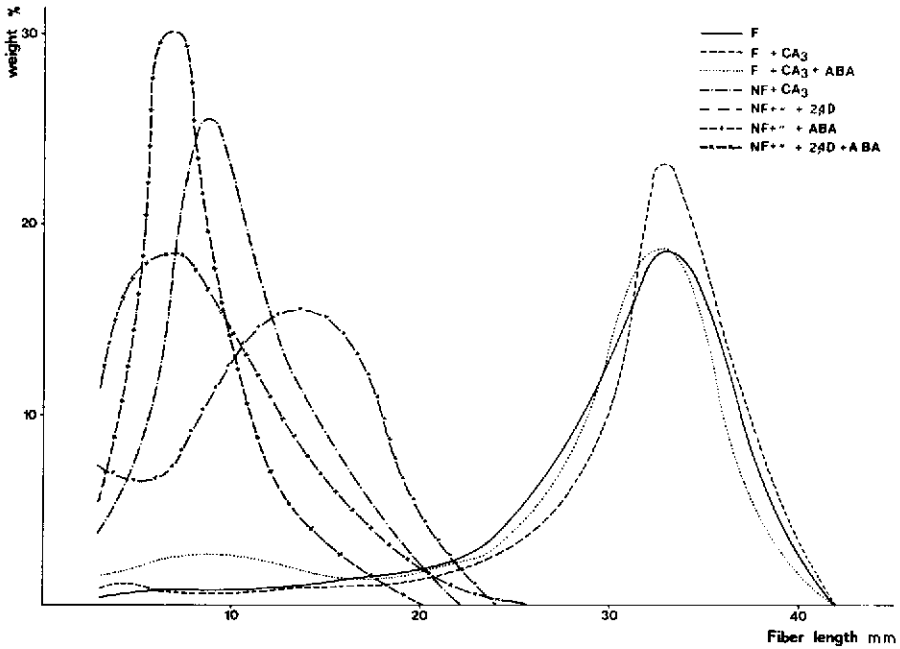

Figure 3. Hormonal effect on cotton fiber length distribution *in situ*. Fertilized (F) and unfertilized (NF) ovules, *G. hirsutum* cv B49.

fruit drop, stimulated a certain degree of fiber elongation. Auxin had a marked positive effect which was additive to the GA_3 stimulation, and ABA counteracted the effect of the other two (Kosmidou, 1976). The auxin stimulus during the elongation phase of fiber is complementary to the work of Jasdanwala *et al.* (1977) who have shown that under natural conditions IAA catabolism is low during this phase. Also, Rodgers (1981a) identified in lint + seed extracts an increased auxin content during the elongation phase with a 6-fold peak at day 15 after anthesis.

Auxins and GA_3 act synergistically *in vitro* on the fiber capacity for primary

wall synthesis and elongation during the early elongation stages (Beasley and Ting, 1973, 1974; Kosmidou, 1976). GA_3 could not affect fiber elongation beyond about the 8th day postanthesis stages. Its positive effect on elongation was greater the earlier the fiber growth stage. On the contrary, auxin enhanced elongation in later stages, too (Kosmidou, 1976). Ethylene (ethephon) in 2-day-old ovules cultured *in vitro* inhibited fiber growth (Hsu and Stewart, 1976).

The work of Beasley and co-workers (Beasley, 1973, 1977a,b; Beasley and Ting, 1973, 1974; Beasley *et al.*, 1974; Dhindsa *et al.*, 1976) *in vitro* has shown that:

1. Two-day-old fertilized and isolated cotton ovules appeared to be deficient in their capacity to synthesize optimum levels of gibberellins, sufficient in their production of cytokinins, and optimum or near optimum in the production of IAA. ABA was not essential for fiber elongation. They, as well as other authors (DeLanghe, 1973; Baert *et al*., 1975; Kosmidou, 1976), concluded that an increase in IAA and GA following fertilization permits ovule and fiber growth.
2. In two-days postanthesis unfertilized ovules, IAA had a marked positive effect on the production of fibers, kinetin supported no fiber elongation, combinations of IAA and GA_3 produced additive amounts of fibers, ABA reduced the amount of fibers induced by IAA, and kinetin partially overcame the inhibition caused by ABA.
3. Unfertilized ovules transferred to culture on the day of anthesis or one to two days preanthesis responded to IAA and/or GA_3 by producing fibers, but not on the fourth (or earlier) day preanthesis. Dhindsa *et al.* (1976) found that ABA inhibited the growth of fiber only when applied during the first four days of culture, and they suggested the possibility that ABA causes this inhibition by interfering with malate catabolism. In another study Dhindsa (1978b) presented evidence that GA_3 causes a large increase in the activities of malate synthesizing enzymes in unfertilized ovules. Furthermore, ABA inhibits the increase in enzyme activities and lowers the ability of ovules to fix CO_2 in dark, apparently by counteracting the GA_3 effect. The same author (1978a) studied the effect of bromodeoxyuridine (BUdR, known to prevent differentiation), AMO-1618 (a growth retardant) and p-chlorophenoxyisobutyric acid (PCIB, an antiauxin) on preanthesis ovules cultured *in vitro*. In the presence of PCIB alone and in combination with GA_3 or GA_3 plus IAA, ovules do not produce fibers and ovule growth appears to be predominantly determined by gibberellin while fiber growth is largely dependent on the availability of auxin.

The identification of considerable amounts of GA's, auxin and cytokinins in lint + seed extracts during the fiber elongation phase which have their peak activity early in this phase (Rodgers, 1981a,b,c), together with the above *in situ* and *in vitro* results, show the importance of hormones on fiber elongation, especially that of auxin and gibberellin. Relatively high concentrations of ABA occur

in bolls and seeds around 5 days postanthesis, which coincides with the high boll abscission rate (Rodgers, 1980). Abscisic acid (ABA), besides its role on boll abscission and inhibition of embryo germination, seems to be a prime candidate as the agent responsible for altering the fiber promoting effects of auxin and gibberellins.

Although the mode-of-action of auxin-regulated elongation is not yet clear, research results in many tissues support the hypothesis that auxin regulates and coordinates both wall loosening and supply of wall materials in elongation (Vanderhoef and Sathl, 1975). Various theories of auxin-stimulated wall acidification and loosening (Cleland, 1979; Ray, 1980), auxin action in two biochemically distinct phases involving rapid lowering of pH and delayed but sustained steady state growth (Vanderhoel and Dute, 1981) and the auxin receptors (Naryanan, 1981; Rubern, 1981; Vreugdenhil *et al.*, 1981; Walton and Ray, 1981) have received much support. Auxin activity in gene expression for sustained cell elongation also appears to be a real possibility (Bevan and Northcote, 1981; Vanderhoef and Dute, 1981; Zurfluh and Guildfoyle, 1980, 1982).

The role of gibberellins in promoting cell enlargement and division in intact plants is recognized, as is the importance of certain tissues like the intact subapical region and the cereal aleurone layer, as target sites for GA action (Rappaport, 1980). Existing information favors a role for GA's in stimulating synthesis of mRNA and intervening in transcription or translation (Rappaport, 1980).

Despite the lack of knowledge of detailed mechanisms so far, the early requirement of GA_3 and IAA for fiber development is recognized, as is the pronounced deleterious effect of ABA.

EFFECT OF HORMONES ON SECONDARY WALL FORMATION

Secondary wall formation begins towards the end of the elongation phase and it is the last fiber developmental stage before maturation. An increase in the number of ER-associated polysomes occurs during this stage (Berlin and Ramsey, 1970; Westafer and Brown, 1976). The formation of secondary wall in fibers of unfertilized ovules has been found to be influenced by auxins and ABA *in situ* (Kosmidou, 1976). 2,4-D promoted the synthesis of cellulose and increased the percentage of mature fibers. Jasdanwala *et al.* (1977) also found that auxin was necessary for secondary wall biogenesis. ABA had the opposite effect of auxin in both fertilized and unfertilized ovules (Figure 4). Abscisic acid could not be detected in 20 DPA seeds *in situ* (Davis and Addicott, 1972) and only at about 30 DPA does it appear to accumulate (Choinski *et al.*, 1981).

Fibers developing from unfertilized ovules cultured *in vitro* produce cell walls very similar to those of plant grown fibers, both with respect to composition and changes in the composition as a function of development (Meinert and Delmer,

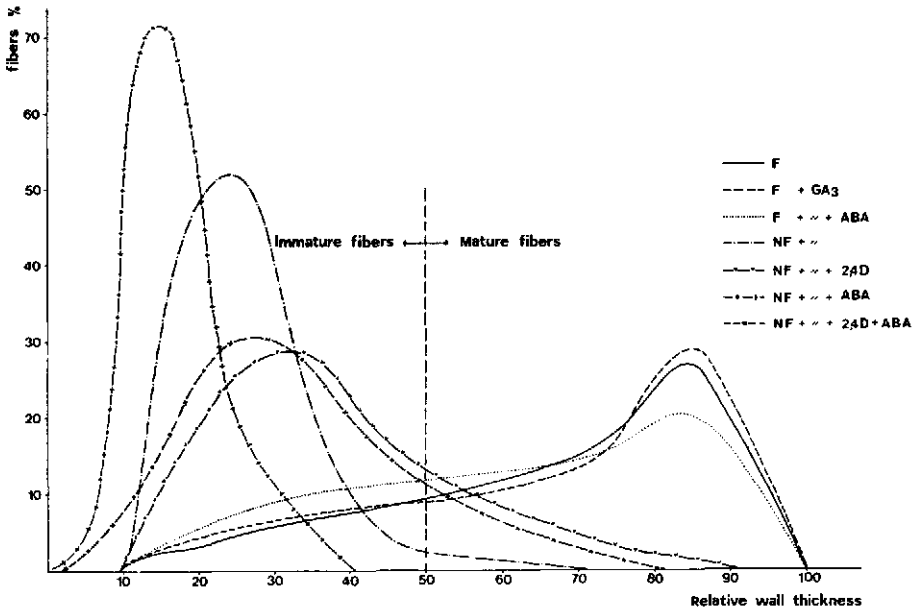

Figure 4. Hormonal effect on cotton fiber relative wall thickness distribution *in situ*. Fertilized (F) and unfertilized (NF) ovules, *G. hirsutum* cv B49.

1977; Carpita and Delmer, 1981). When unfertilized or fertilized ovules were transferred from plants at 5-14 days postanthesis to *in vitro* culture without hormones their fibers could produce secondary walls (Kosmidou, 1976). Auxin (NAA 10^{-5}M and 10^{-5}M) increased the amount of cellulose formed by these fibers and consequently their maturity, while ABA (10^{-6}M) reduced it significantly. In the same study no significant effect of GA_3 (10^{-7}M, 10^{-6}M, 10^{-5}M) was detected.

According to Tarchevskii *et al.* (1980) inhibitors of ATP formation in the course of oxidative phosphorylation (rotenone and antimycin A) and glycolysis (sodium fluoride and monoiodoacetate) powerfully depressed cotton fiber cellulose synthesis. Cellulose synthesis in cotton fibers is limited by the content of macroergic phosphates, and modification of the cell energy regime constitutes one of the ways in which synthesis of cotton cellulose can be effectively regulated. Other inhibitors of cellulose synthesis in cotton fibers have been characterized also (Montezinos and Delmer, 1980). Understanding the process of cellulose biosynthesis may help to understand the hormonal influences in it.

Ethylene is known to be implicated in the control of fruit ripening. In cotton it has received much attention as one of the causal agents in young fruit abscission (Guinn, 1979). Maximum rates of ethylene evolution in cotton fruits coincide with the highest incidence of young fruit abscission (Guinn, 1982). Ethrel, which causes rapid release of ethylene in the plant, has been found to stimulate the

translocation of assimilates into cotton bolls, particularly into fibers, during the six days following ethrel treatment (Yue-Quing *et al.*, 1980a). According to this report, the result of this extra translocation was the improvement of fiber quality and fullness, implicating thicker secondary walls. Ethylene also exerts an effect on the orientation of microtubules and cellulose microfibrils in pea epicotyl and stem tissues (Steen and Chadwick, 1981; Lang *et al.*, 1982). Recent progress in the identification of the steps in ethylene biosynthesis in plant tissues (Adams and Yang, 1979; Lürsen *et al.*, 1979), as well as in ways to prevent or inhibit ethylene action, provides the means for detailed investigations regarding the involvement of ethylene in cellulose microfibril deposition and secondary wall formation.

SUMMARY

Research into the influence of hormones on cotton fiber development made significant progress in the last decade, mainly due to the use of the *in vitro* culture of cotton ovules with their associated fibers. Identification of the naturally occurring hormones in the cotton ovule, and fundamental work on the mode of hormone action in other plants, complemented our understanding on this subject, although not all mechanisms and maybe not all hormonal influences are known. More information on the differences in hormonal balance and changes during all fiber growth stages, in fibers of short versus long cottons, and in the detailed mechanisms of hormone action, would be helpful.

The involvement of phenolic substances in the process of fiber differentiation and the mediation of gibberellins and auxin during fiber differentiation and initiation have been proved. Evolution and structure of fiber nucleoli are changed by the influence of auxins, GA_3 and ABA, and it is speculated that these organelles play a direct or indirect role in hormonal action during fiber development. Hormonal balance seems very important, especially during the early stages of fiber growth.

During the fiber elongation phase, a synergistic effect of GA_3 and auxin seems to play an important role. There is evidence that GA exerts its action during the early days following anthesis, while the presence of auxin seems to be necessary from the early stages through secondary wall formation.

During the secondary wall formation, auxin increases the amount of cellulose deposited in the fiber walls. Limited evidence suggests ethylene involvement during the same fiber growth stage, and further investigation on its exact action, interactions and all possible influences is important.

Abscisic acid counteracts the GA and auxin effects on fiber development, and it has been proposed as the prime candidate responsible for altering the fiber promoting effects of these two types of hormones.

Although considerable information is now available on the involvement of hormones in fiber differentiation and development, a more thorough understanding of the mode of hormonal action, sequential changes, balance and interactions during cotton fiber growth is needed.