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Growth and Development of the Lint Fibers of Pima S-4 Cotton[1]

A. M. Schubert, C. R. Benedict, C. E. Gates, and R. J. Kohel[2]

ABSTRACT

Lint fiber elongation, lint dry weight per seed, and lint dry weight per seed per unit length, was studied from anthesis to maturity in field-grown 'Pima S-4' cotton (*Gossypium barbadense* L.). These parameters of fiber growth and differentiation were plotted against boll age and fitted to best-fit curves by curvilinear regression analysis. The curve fitting procedures yielded mathematical equations which were differentiated to give rate curves. Fiber growth stopped 39 days after anthesis. Splining analysis of fiber dry weight and fiber dry weight per unit length showed that the lint weight per unit length ratio was constant to 17.99 ± 2.35 days. This is the time that secondary wall thickening begins. This analysis also indicates that during the 48-day boll development period, both fiber elongation and secondary wall thickening were occurring simultaneously for 21 days. During the 21-day overlap period, 19.7% of the final fiber length was attained. At the end of fiber elongation, fiber dry weight and fiber dry weight per unit length had reached 90% of their final values. These data show that fiber elongation and secondary wall thickening are not separate phases and a substantial amount of secondary wall thickening occurred before fiber elongation stopped. This degree of overlap of fiber growth and differentiation phases is more striking than for upland cottons and may be related to the fact that the maximum rate of elongation continues for 12 days longer in 'Pima S-4' compared to 'Stoneville 213' cotton (*Gossypium hirsutum* L.). The findings reported here show that deposition of almost pure cellulose in the secondary wall does not immediately stop all of fiber elongation.

Additional index words: Gossypium hirsutum L., Gossypium barbadense L., Lint fiber elongation, Lint dry weight, Lint dry weight per unit of lint length, Fiber primary wall, Fiber secondary wall.

COTTON (*Gossypium* sp.) lint fibers are formed by the elongation of cells in the epidermis of the seed coat of developing cotton seed. Each lint fiber is a single cell, which may be 2.5 cm or more in length and 20 um in diameter (10). The transformation of epidermal cells of the unfertilized cotton ovule to mature seed hairs involves both growth and differentiation. The growth phase consists primarily of cell elongation and the differentiation phase involves the thickening of the fiber wall to occupy more than half the final cell diameter in a mature fiber (8,9). The initial step in fiber development is elongation with secondary wall development beginning later. The generally accepted explanation of fiber development has held that the elongation and wall thickening phases are mutually exclusive (1, 4, 5, 11, 14). Despite the findings of Lang (15) which indicate lint fibers initiate elongation over a fairly short period (2 to 3 days), apparent overlap in time between elongation and wall thickening has been explained as due to variable initiation times and variable growth rates within the population of fibers (2, 3, 6, 11, 19).

Among the earliest research on the growth and development of cotton fibers was by Hawkins and Serviss (14). Their designation of fiber elongation and wall thickening as distinct phases was based on the observation that fiber wall thickening was occurring at only a rather low rate before elongation stopped in Acala cotton (*Gossypium hirsutum* L.) and Pima cotton (*G. barbadense* L.). However their data show that at the time elongation stopped completely, the Acala fibers had attained 21 to 26% of their final wall thickness and the Pima fibers had obtained 60 to 76%.

Benedict et al. (7) found that fiber dry weight was 37 to 59% complete at the time elongation stopped in upland cotton cultivar 'Stoneville 213' (*G. hirsutum* L.). Schubert et al. (17) fitted the growth and differentiation fiber parameters of Stoneville 213 to best-fit curves by curvilinear regression analysis. The data confirmed that fiber weight per seed and fiber weight per unit of fiber length were 42% complete at the time elongation stopped. In this paper similar curvilinear regression analysis procedures are applied to the growth and development parameters of Pima fibers to study the 60 to 70% overlap in wall thickening and fiber elongation reported by Hawkins and Serviss (14).

MATERIALS AND METHODS

The bolls used in this study were from 'Pima S-4' plants grown in the Cotton Variety Test Plots on the Texas A&M Univ. Farm at College Station, Texas. These plants were supplied courtesy of Dr. G. A. Niles. The plants were grown in rows 1 m wide and were thinned to 1 m between plants to permit easy access to the plants for flower tagging and boll collection. The experimental design was a randomized, complete block design with three replications. Fertilization, irrigation, and pest control procedures were those used throughout the test plots, designed to maximize lint yields.

Throughout the growing season, flowers were tagged on the day of anthesis. As nearly as possible, bolls studied were from flowers that bloomed over a narrow time span to minimize differences due to variation in environmental conditions at specific stages of boll development. Boll age was expressed as the number of days past anthesis. Bolls were harvested at 3-day intervals from 6 days after anthesis to maturity.

Boll walls were split with a scalpel and the carpels removed intact. The seeds were separated by a procedure of Morris (16), Gipson and Joham (12), Gipson and Ray (13), Benedict et al. (7) and Schubert et al. (17) as follows: carpels were placed in boiling water for 2 to 5 min to separate the seeds and float them apart. The separated seeds were quickly transferred to cool water. A small amount of concentrated HCl (4 drops/100 ml) was added to the water used to separate the seeds in carpels more than 20 days old. The lower HCl concentration than that frequently used (7, 12, 13), and immediate transfer to cool water seemed to minimize seed coat breakage during subsequent lint removal, while aiding seed separation in the older bolls.

Ten seeds were randomly selected for lint length measurements. Each seed was placed on the convex side of a watchglass and the fibers flared out by a jet of water. A direct measurement of the

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[2] Former graduate student (presently post-doctoral research associate, Dep. of Agronomy, Univ. of Georgia, Athens, GA 30601); professor, Dep. of Plant Sciences, Texas A&M Univ., College Station, TX 77843; professor, Inst. of Statistics, Texas A&M Univ.; and research geneticist, ARS, USDA, College Station, TX 77843.

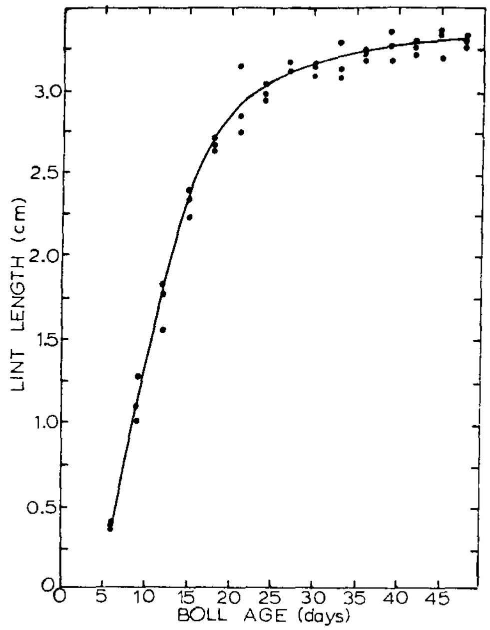

Fig. 1. Best-fit curve for lint fiber elongation in Pima S-4 described by Eq. [1]. $R^2 = 0.992$ and $R = 0.996$.

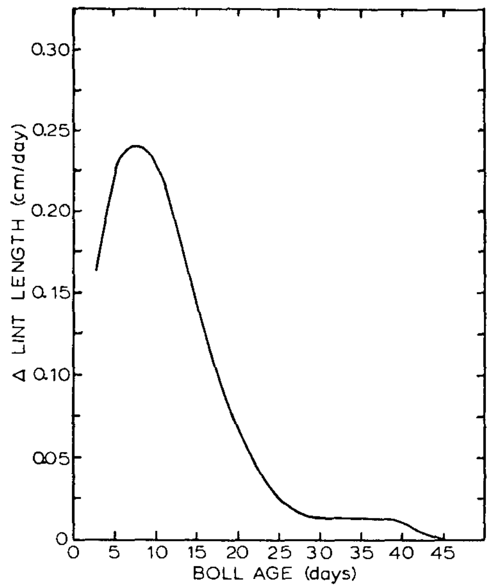

Fig. 2. Rate of elongation of lint fibers in Pima S-4. This curve is described by the derivative of Fig. 1 (Eq. [2]). Elongation was considered complete at 39 days.

halo length was made from the side of the seed, midway between the chalazal and micropylar ends. This measurement site was an arbitrary choice. The fibers were carefully parted during the flaring procedure, so that only those fibers at or very near the reference point on the seed were measured. This greatly reduced the halo effect and simplified measurement. The mean of the 10 seed measurements was used as the fiber length value for that boll.

All of the seeds from each boll were counted and transferred to 80% (vol/vol) ethanol until fiber removal. The fibers were manually removed from the seed using a tearing action with a scalpel. Care was exercised to prevent tearing off pieces of the seed coat and removal of fuzz fibers during this operation. When fiber removal was complete, and the seed allowed to dry, they looked similar to undelinted cotton seed following ginning. The lint was washed with 80% ethanol and dried at 80 C. The dry fibers were weighed, and the weight values expressed on a per seed basis to reduce variability introduced by variation in seed number between bolls. Lint dry weight per seed was used as an indication of cell wall deposition.

Fiber elongation is accompanied by primary cell wall growth and this component of weight gain was removed from total weight gain by dividing lint dry weight per seed by the lint length value of that boll. Lint dry weight per unit lint length is analogous to the linear density measurements once used as a fiber quality indicator. This parameter should indicate cross-sectional wall development, and if fiber elongation and wall thickening are mutually exclusive phases, lint weight per unit length should remain constant throughout the elongation period.

Each fiber growth and development parameter was plotted vs. boll age. Appropriate curves were fitted to the data by curvilinear regression analysis procedures. These procedures provided objective curves describing fiber elongation and weight gain, accompanied by estimates of experimental error. Also, curve-fitting procedures provided mathematical formulas with defined coefficients which were differentiated to yield rate curves. These rate curves were used to identify the fiber age when elongation stopped as well as the specific rates of development at different developmental stages.

RESULTS

Lint fiber elongation proceeded very rapidly early in the boll development period. The graph of data points in Fig. 1 suggested an exponential relationship between boll age and lint length. The data were, therefore, fitted to a polynomial,

$$\hat{Y} = a + bx + cx^2 + \ldots + gx^6 + hx^7 + E \quad [1]$$

where

$\hat{Y}$ =	predicted lint fiber length	d =	-0.287262×10^{-2}
		e =	0.820870×10^{-4}
x =	boll age	f =	-0.993549×10^{-6}
a =	-0.519102	g =	0.214999×10^{-8}
b =	-0.133918×10^{-1}	h =	0.315988×10^{-10}
c =	0.410961×10^{-1}	E =	error of estimation

The Pima S-4 fibers attained a final length of 3.67 cm. The fit of the curve to the data was very good as indicated by a coefficient of determination (R^2) of 0.992, and the multiple correlation coefficient (R), 0.996.

The derivative of eq. [1],

$$d\hat{Y}/dx = b + 2cx + 3dx^2 + \ldots + 6gx^5 + 7hx^6 \quad [2]$$

describes the elongation rate curve seen in Fig. 2. Elongation rate peaked at 7 to 9 days after anthesis and then decreased. By 27 days past anthesis, the elongation rate was low but still proceeded steadily until 39 days. We designated 39 days as the end of elongation, even though elongation may have still been proceeding at a very slow rate.

When lint dry weight per seed was plotted vs. boll age, the relationship was sigmoidal. The data were fitted to a logistic growth equation cited by Snedecor and Cochran (18):

$$\hat{W} = A/(1 + BP^x) \quad [3]$$

where

$\hat{W}$ = predicted dependent variable; lint wt/seed
x = the independent variable; boll age
A = the asymptote ($x \rightarrow \infty$)
B = is related to the W-intercept as follows:

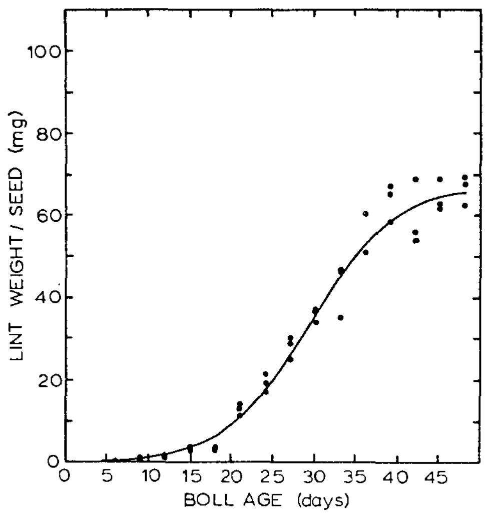

Fig. 3. Lint weight per seed development described by Eq. [3]. The mean deviation from regression is 3.750 mg.

$$W_{X=0} = A/(1 + B)$$

or

$$B = (A/W_{X=0}) - 1$$

P = the shape parameter; indicating the slope of the central portion of the curve.

The logistic growth curve, Eq. [3], was fitted to the data by a nonlinear computer program utilizing Hartley's Modified Gauss-Newton method for nonlinear regression as programed by Dr. E. L. Butler, with documentation modification by Dr. C. E. Gates.

Coefficients defining the curve in Fig. 3 are:

$A = 6.812677 \times 10^1$
$B = 3.174719 \times 10^2$
$P = 8.206194 \times 10^{-1}$.

The mean square deviation from regression is 3.750 mg. At the time elongation ceased, 89.4% of the final lint dry weight per seed had been reached.

Figure 4 shows the rate of lint dry weight per seed gain. This curve was obtained by substituting the above coefficients into the derivative of Eq. [3]:

$$d\hat{W}/dx = -ABP^x\ (\ln P)/(1 + BP^x)^2 \qquad [4]$$

Lint dry weight gain peaked at 29 days, 10 days before elongation stopped.

The data for lint dry weight per unit length were also fitted to Eq. [3]. Figure 5 shows a final value of 20.5 mg/cm was attained. Coefficients for this curve are:

$A = 2.09259 \times 10^1$
$B = 2.275028 \times 10^2$
$P = 8.267596 \times 10^{-1}$.

The mean square deviation from regression is 1.188 mg/cm. At 39 days past anthesis, 89.9% of the final value had been attained.

The rate curve for lint dry weight per unit length (Fig. 6) indicates that the most rapid wall thickening occurred 10 days before elongation stopped completely.

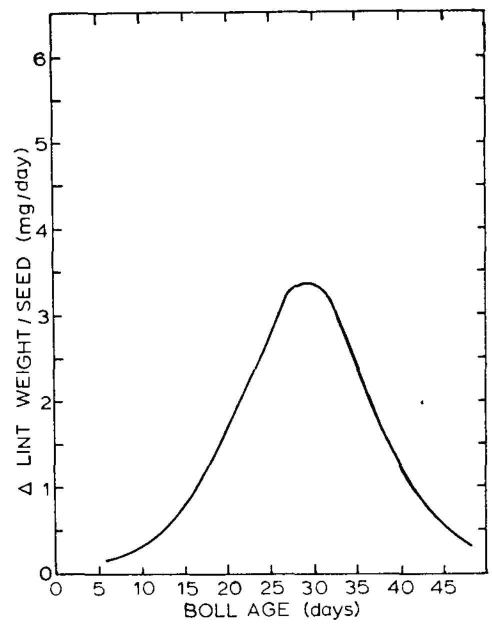

Fig. 4. Rate of lint weight per seed increase as described by Eq. [4]. This curve is the derivative of the curve in Fig. 3.

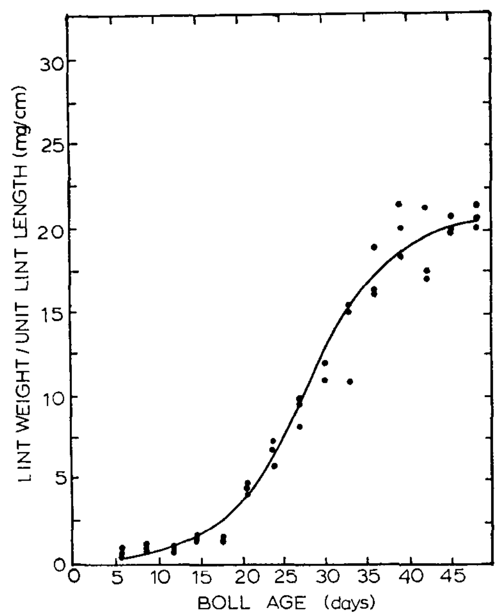

Fig. 5. Lint weight per unit length increase during boll development. The curve is described by Eq. [3]. The mean square from regression is 1.188 mg/cm.

Data for lint dry weight per unit length were fitted to three intersecting straight lines to determine approximately when this ratio ceased to be constant (see Fig. 7). The model for the nonlinear regression analysis contains the defining equations for all three lines with their two intersections (or cuts):

$$\hat{Y} = \lambda_1\ [a + b_1(x - x^*)] + \lambda_2\ [a + b_2\ (x - x^*)] + \lambda_3\ [a' + b_3\ (x - x^+)] \qquad [5]$$

where

$$\lambda_1 = 1 \text{ if } x \leqslant x^*;\ 0 \text{ otherwise}$$

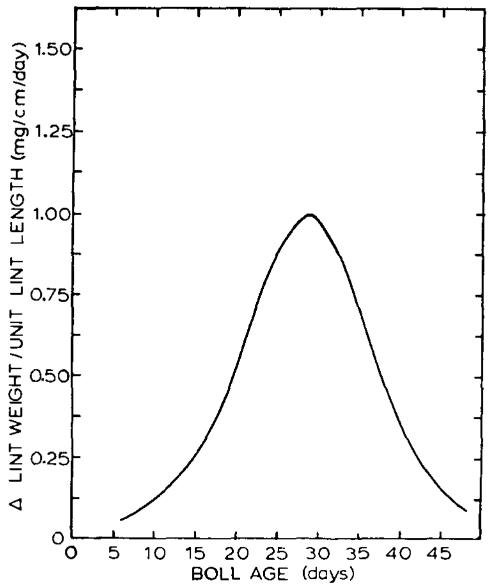

Fig. 6. Rate of lint weight per unit length increase as described by Eq. [4]. This is the derivative curve of that in Fig. 5.

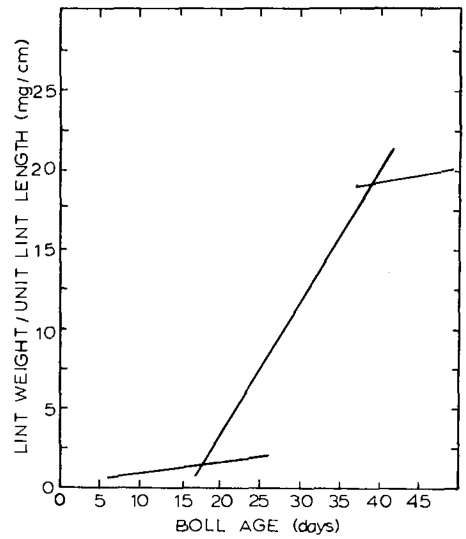

Fig. 7. Splined curve for lint weight per unit length development as defined by Eq. [5]. Note that the ratio was fairly constant until 18 days after anthesis, when wall thickening began.

λ_2 = 1 if $x^* < x \leqslant x^+$; O otherwise
λ_3 = 1 if $x > x^+$; O otherwise
b_1 = slope of the first line
b_2 = $(a' - a)/(x^+ - x^*)$; slope of the second line
b_3 = slope of the third line
a = lint dry wt/unit length at the first cut
a' = lint dry wt/unit length at the second cut
$\hat{Y}$ = predicted lint dry wt/unit length
x = boll age
x^* = boll age at the first cut
x^+ = boll age at the second cut

There are six parameters in the model: b_1, b_3, a, a', x^*, x^+.

Figure 7 shows that early in the developmental period the lint dry weight per unit length ratio was almost constant. However, at 17.99 ±2.35 days, the ratio began increasing rapidly. This indicates that wall thickening began 21 days before elongation stopped. At the time wall thickening began, the fibers had reached 80.3% of their final length (see Fig. 1).

DISCUSSION

This study of the growth and development patterns of Pima S-4 lint fibers reveals a very high degree of overlap between elongation and wall thickening phases. The lint continued to elongate for at least 39 days after anthesis. Wall thickening began about 18 days after anthesis. This indicates that during the 48-day boll development period, both lint elongation and wall thickening were occurring for 21 days. During that 21-day overlap period, a substantial amount, 19.7%, of the final lint length was attained, even though elongation rates were lower than the peak rates seen at 7 to 9 days. At the time elongation stopped, almost 90% of the final values for both weight parameters had been reached. This overlap is even more striking than that indicated by the reexamination of the data of Hawkins and Serviss (14), and that of Benedict et al. (7) and Schubert et al. (17) for upland cottons.

It is important to note that the parameters used in this research reflect known differences between Pima and upland cottons. Compared to the Stoneville 213 reported previously (17), Pima S-4 had longer fibers, 3.27 cm compared to 2.73 cm, and finer fibers, as reflected by a 20.93 ± 1.15 mg/cm lint weight per unit length asymptote vs. 29.86 ± 1.42 mg/cm. A comparison of lint elongation rate curves shows what growth characteristics contributed to the greater fiber length: i) although the peak elongation rates appeared slightly lower in Pima S-4, elongation continued at nearly the maximum rate much longer than in Stoneville 213; and ii) the elongation period was 12 days longer in the Pima S-4.

The findings reported here show that the deposition of the almost pure cellulose of the secondary wall does not immediately stop all fiber elongation. In the case of Pima S-4, the greater overlap may be associated with the fibers' inherently thinner walls. It seems likely that some specific wall thickness might be required to stop elongation completely. In Pima S-4, the lint weight per unit length gain peaked at 1.00 mg/cm/day compared to 1.31 mg/cm/day in Stoneville 213 the previous year.

This research has reopened questions concerning cotton lint fiber development which have been considered closed for years. It points to possible new means of selecting for quality cotton lint under specific growing conditions, since fiber length and wall thickness are important quality factors, and indicates parameters and data handling methods which might be useful in applying the new selection criteria. From a basic standpoint, these findings indicate the need to extend our knowledge of the expansion processes and capacities of cell walls, especially during differentiation phases of development.

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