

# Seasonal Variation in Flowerbud Gossypol Content in Cotton<sup>1</sup>

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## ABSTRACT

Flowerbud gossypol content has been associated with a degree of host plant resistance in cotton (*Gossypium hirsutum* L.), but efforts to increase and stabilize genetic levels have been hindered by an apparent seasonal variation in gossypol content. A 4-year study (1974 to 1977) of two lines having flowerbuds with high levels of gossypol and two cultivars with normal levels of gossypol revealed a significant change in gossypol contents both within and between seasons. The gossypol contents in 1975 were significantly greater than those in 1974 and 1977 for the two lines having flowerbuds with high levels of gossypol. Also, a significant within-season variation in gossypol contents occurred among all four genotypes that were tested. The coefficient of correlation comparing gossypol contents of the two lines having flowerbuds with high gossypol levels and of the two cultivars were 0.82 and 0.79, respectively. Whereas, the coefficient of correlation comparing the gossypol contents of the cultivars with the high gossypol lines were all less than 0.55. These data show that comparisons of flowerbud gossypol levels within a season should be based on seasonal averages or should be made as close to the same date as possible. There should not be more than a 7-day interval between dates if direct comparisons are to be made among entries within a single season.

Additional index words: Insect resistance, Terpenoid aldehydes, Polyphenolics, Breeding stocks, Cultivars, Race stocks, *Gossypium hirsutum* L.

THE above-ground plant parts of cotton (*Gossypium hirsutum* L.) possess natural pigment glands which contain gossypol, a polyphenolic yellow pigment, and other related terpenoid aldehydes (9). Gossypol has been recognized for many years as a substance toxic to several insect pests (11). Thus, it seemed desirable to develop upland cultivars with increased levels of total gossypol in the flowerbuds (squares). However, it has proven difficult to develop upland breeding stocks with more than 1.0% flowerbud gossypol combined with adequate agronomic and fiber properties (7, 12). A limited germ plasm pool having high levels of flowerbud gossypol has been a hindrance until new sources were recently identified and made available to breeders (1).

Efforts to increase and stabilize levels of flowerbud gossypol have been hindered by an apparent seasonal variation in gossypol content. Several workers have reported seasonal variation in gossypol content in cotton.

However, most of the plants used in previous studies have been greenhouse-grown, 'stubbed-off' transplants, cotton grown under atypical conditions, or plants not possessing increased levels of flowerbud gossypol (2, 3, 5, 12). This diversity makes the interpretation of these data for breeding purposes difficult.

The study reported in this paper was designed to determine the within and between seasonal variation in flowerbud gossypol of two cultivars with normal levels and two lines with increased levels of flowerbud gossypol.

## MATERIALS AND METHODS

This study was conducted over a 4-year period (1974 to 1977). We used two cultivars ('Stoneville 213' and 'Deltapine 16') with normal levels of flowerbud gossypol, and two breeding lines (HG-44 and HG-46) with increased levels of gossypol. The two breeding lines, developed by the authors, are sister-line selections from a cross of Socorro Island Wild cotton (Texas 934) with Stoneville 213.

The experimental design used was a randomized complete block with four replications. The tests were planted on the same type of soil classified as a Laredo-Reynosa complex which is a fine-silty, mixed, hyperthermic soil on 29 March 1974, 12 March 1975, 12 March 1976, and 21 March 1977. Plots were hand thinned to about 10 plants/m when the plants had about three to four true leaves. None of the plots were irrigated during the 4-year period. Flowerbuds for gossypol analysis were harvested at random from the plants, placed on dry ice, debarked, freeze-dried, and ground through a 0.30 mm screen and stored in a laboratory freezer for subsequent analysis. Sampling dates were as follows: 24 May, 29 June, and 22 July 1974; 19 May, 25 June, and 19 July 1975; 25 May, 1, 8, 15, 22, and 29 June and 13 July 1976; and 3 and 17 June and 15 July 1977. The level of gossypol, plus related terpenoid aldehydes, were determined for each of the flowerbud samples by a method described by Smith (8).

The data were analyzed and the means were compared with Duncan's multiple range test. In order to have an equal number of harvest dates for the four-year combined analysis, the results were used from only three (25 May, 29 June, and 13 July) of the seven harvest dates in 1976. These dates were selected because they were similar to the harvest dates of 1974, 1975, and 1977. However, data from all seven dates were used for the analysis of variance for 1976 only.

The earliest common harvest date for the 4-year period was

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74 days after planting. The gossypol contents of the three harvest dates in 1974, 1975, and 1977 and the seven harvest dates in 1976 were plotted and the number of days after planting was superimposed over the actual harvest dates. The gossypol contents at each of the four dates (74, 88, 102, and 116 days after planting) taken at 2-week intervals were not used for analysis but only to depict changes in graphic form for the 4 years. All statistical analyses were based on gossypol contents from actual harvest dates.

## RESULTS AND DISCUSSION

A combined analysis of variance over years and entries showed significant differences due to years, entries (cultivars vs. lines), and harvest dates. Figure 1 depicts the differences and similarities between years and Figs. 2 to 5 depict the differences and similarities between entries and also harvest dates in graphic form. Means are shown in Table 1. These data reveal that there are many similarities between and within years and entries in levels of gossypol, but more importantly there are many significant differences that occur both between and within years and entries. Furthermore, if these differences in levels of gossypol are known to exist and become part of the experimental design for evaluating breeding stocks then they can serve as a tool in breeding for high flowerbud gossypol cotton.

When the entries were evaluated within years (means within the same entry within the same year; Table 1) there was no seasonal change in gossypol level in 1974 in three of the four lines. Stoneville 213, however, showed a significant decrease at the last harvest date. In 1975, both HG-44 and Deltapine 16 showed a significant seasonal change, an increase from first to third harvest for HG-44 and an increase from second to third harvest for Deltapine 16. In 1976, there was no change in the gossypol level in Deltapine 16 during the season, but there was a slight upward trend in the other three as the season pro-

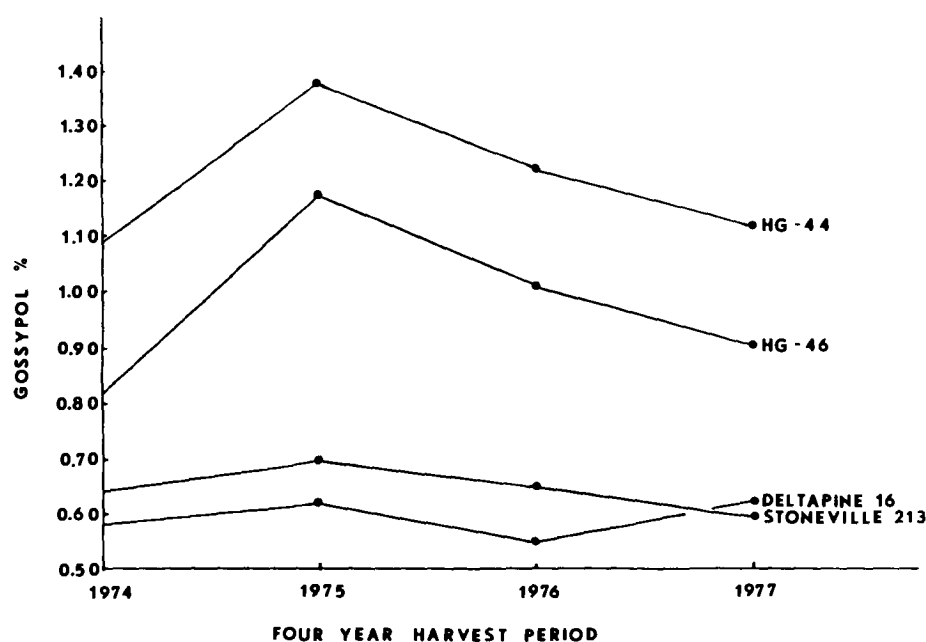
gressed. The highest gossypol percentages occurred 109 and 123 days after planting. In 1977, there was no change in the gossypol level during the season in HG-46, a slight decrease in HG-44 at the middle harvest date and a significant decrease in the two cultivars as the season progressed.

When the data from each entry are compared over the 4-year test the results from Table 1 show that there were significant differences in gossypol contents within each

**Table 1. Gossypol percentages over a 4-year period for two cultivars and two high gossypol (HG) cotton lines harvested on different dates.**

Harvest date	Gossypol				Days after planting
	Stoneville 213	Deltapine 16	HG-44	HG-46	
	%				
No.					
1974					
24 May	0.69 a*	0.61 a	1.01 a	0.84 a	56
29 June	0.69 a	0.63 a	1.11 a	0.76 a	92
22 July	0.54 b	0.50 a	1.15 a	0.84 a	115
1975					
19 May	0.70 a	0.64 ab	1.28 b	1.15 a	68
25 June	0.66 a	0.56 b	1.39 ab	1.24 a	105
19 July	0.74 a	0.70 a	1.49 a	1.16 a	129
1976					
25 May	0.56 bc	0.50 a	1.04 c	0.89 b	74
1 June	0.54 c	0.47 a	1.02 c	0.98 b	81
8 June	0.65 ab	0.54 a	1.12 bc	1.02 b	88
15 June	0.54 c	0.52 a	1.13 bc	0.97 b	95
22 June	0.56 bc	0.49 a	1.16 bc	0.92 b	102
29 June	0.72 a	0.58 a	1.29 ab	1.21 a	109
13 July	0.66 ab	0.57 a	1.35 a	1.24 a	123
1977					
3 June	0.72 a	0.75 a	1.19 a	0.99 a	74
17 June	0.59 b	0.60 b	1.04 b	0.87 a	88
15 July	0.48 c	0.53 b	1.14 ab	0.88 a	116

\* Means followed by the same letter within the same entry and year are not different at the 0.05 probability level according to Duncan's Multiple Range Test.



**Fig. 1. Seasonal averages of flowerbud gossypol percentages for two HG stocks and two cultivars over a 4-year period.**

season in 3 of the 4 years for Stoneville 213 and HG-44, in 2 years for Deltapine 16, and in 1 year for HG-46.

The four lines held their relative position except for the two cultivars in 1977 (in that year, Stoneville 213 was slightly lower than Deltapine 16 in gossypol content; Fig. 1). Based on seasonal averages, HG-44 was always higher than HG-46 in gossypol content and the two HG lines could be separated from the two cultivars in 3 of the 4 years (HG-46 was not significantly different from Stone-

ville 213 in 1974). However, the important point is that individual samples cannot be harvested at random during a season and compared with other samples harvested at random during that season or some other year. For example, in 1974 HG-44 was significantly higher than HG-46 based on seasonal averages, however, there was no significant difference between HG-44 harvested on 24 May and HG-46 harvested on 24 May or 22 July. Results were similar during the other 3 years, also. That is

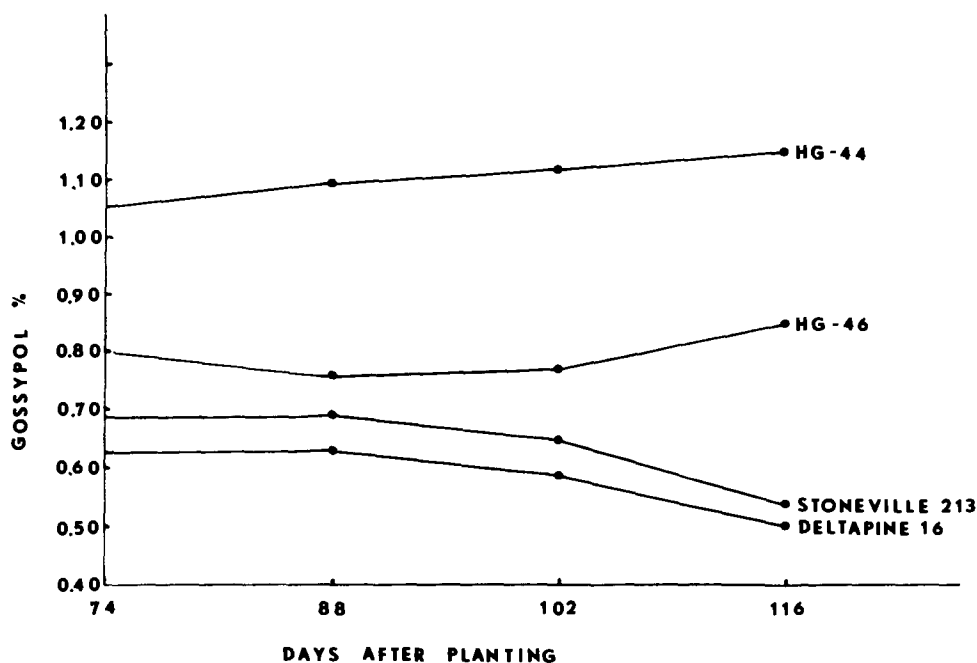


Fig. 2. The average flowerbud gossypol contents of two high gossypol stocks and two cultivars in 1974.

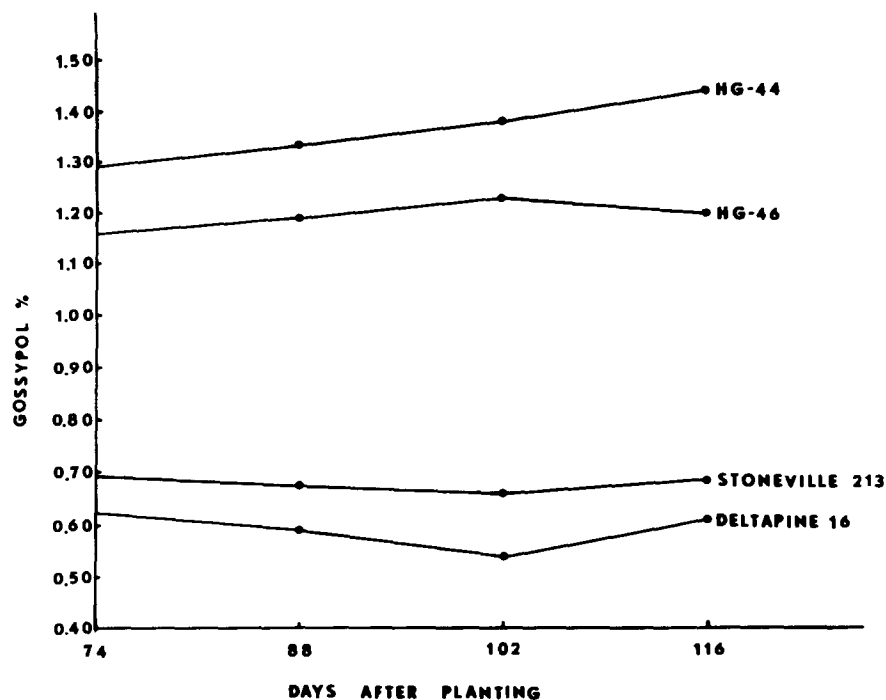


Fig. 3. The average flowerbud gossypol contents of two high gossypol stocks and two cultivars in 1975.

seasonal means were significantly different among entries for the most part, but means at certain dates were not.

In 1976, gossypol contents showed a marked increase between days 102 and 109. This result suggests that, if seasonal averages cannot be obtained, comparisons between lines should be made from samples picked on the same date or no more than 7 days apart. Year-to-year variation in seasonal mean gossypol levels among the four lines indicate that the cultivars were more stable than were the two HG lines. One possible explanation for the differential response of the Socorro Island derivatives as

compared to the standard cultivars may be that different genetic mechanisms are responsible for gossypol production in these cottons. The two major genes controlling gossypol production in the cultivars have been designated  $G1_2$  and  $G1_3$ . Wilson and Smith (10) reported a high potency allele at the  $G1_3$  locus in a derivative of Socorro Island Wild cotton. Also, Lee (4) reported an additional allele in another Socorro Island derivative which is responsible for the rugate boll ( $G1_3$ ) character.

The slight decrease in gossypol content in the two cultivars in 1974 and 1977, and the slight increase in 1975

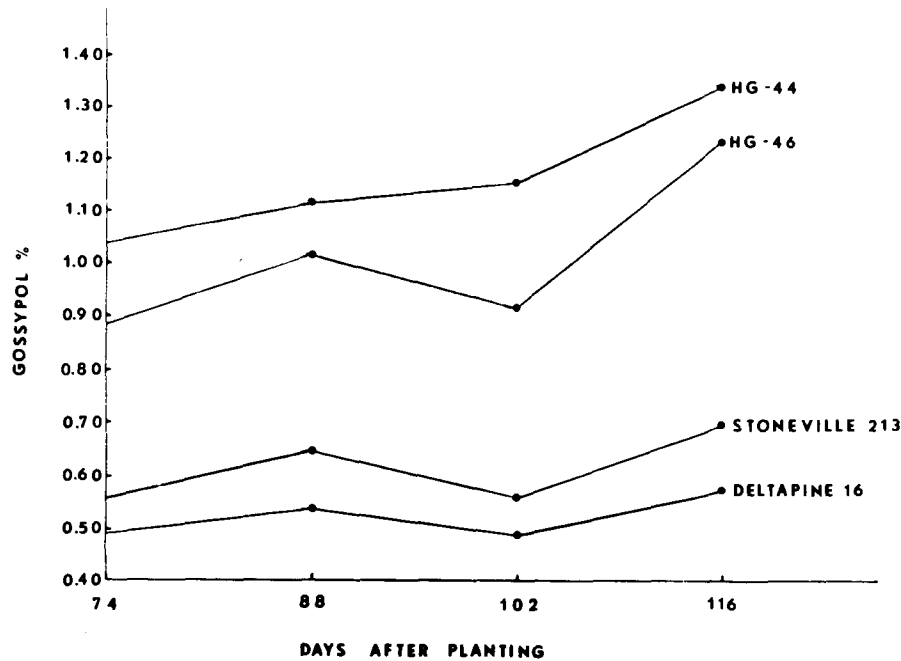


Fig. 4. The average flowerbud gossypol contents of two high gossypol stocks and two cultivars in 1976.

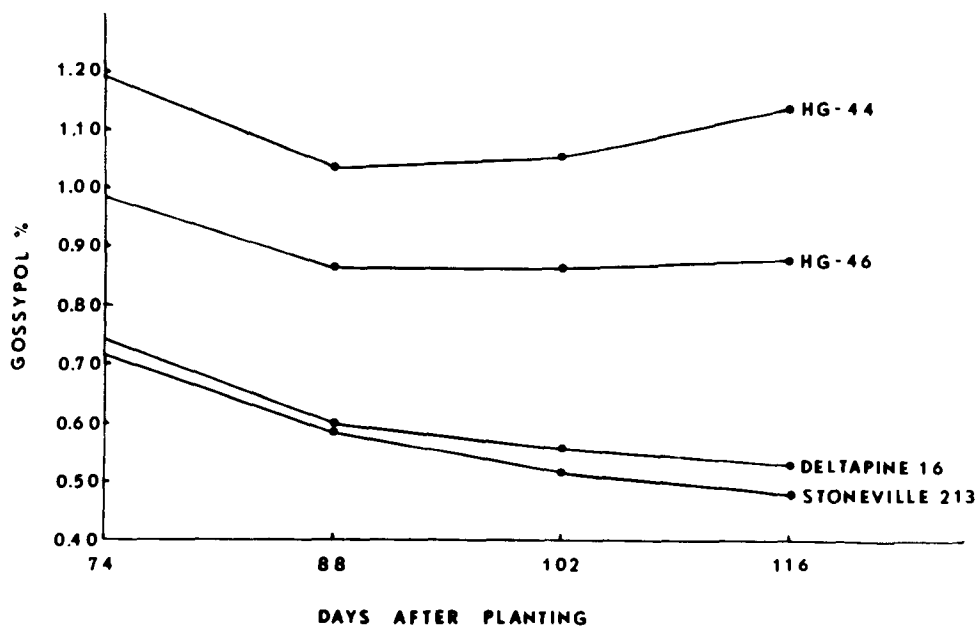


Fig. 5. The average flowerbud gossypol contents of two high gossypol stocks and two cultivars in 1977.

**Table 2. Coefficients of correlation for gossypol percentages in the two cultivars and the two high gossypol lines for the 4-year period, 1974 to 1977.**

Entry	Deltapine 16	HG-44	HG-46
Stoneville 213	0.79**	0.53*	0.48
Deltapine 16		0.39	0.18
HG-44			0.82**

\*,\*\* Significant at the 0.05 and 0.01 probability levels (14 df), respectively.

and 1976 as the season progressed indicate the extent to which environmental factors affected the amounts of this substance. Pons et al. (6) found a negative correlation between temperature and gossypol content of cottonseed but a positive correlation between rainfall and the latter. Our study was not designed to determine the interrelationship of gossypol content with environmental factors. However, both our data and the data of Pons et al. (6) show significant changes within and between seasons. This result suggests that environmental factors could play an important role in the levels of gossypol in both the seed and vegetative plant parts of cotton.

Our data suggest that the breeder who desires to produce lines of cotton having high flowerbud gossypol content should be able to separate those having >1.0% gossypol from those having <0.7% gossypol in a single season based on either seasonal averages or on individual harvests as long as the harvests are no greater than 7 days apart. Variation in gossypol content among lines and between years, combined with variation within a single season (e.g. HG-46, 0.89 to 1.24% and HG-44, 1.02 to 1.35% in 1976), make comparisons among lines and between years risky. However, our data suggest that these comparisons are possible if several harvests are made and seasonal averages are compared, or if all of the lines are harvested no more than 7 days apart and compared to a

standard which is grown each year. Furthermore, one should expect a greater year-to-year variation in high gossypol lines than in standard cultivars, until or unless genetically stable high gossypol lines are developed.

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