FATTY ACID COMPOSITION OF LIPID FRACTIONS IN GERMINATING COTTON AS AFFECTED BY TEMPERATURE B. Lauterbach, D.R. Krieg and G. Jividen Texas Tech University and Cotton Incorporated Lubbock, TX and Raleigh NC

<u>Abstract</u>

Chilling injury occurs in cotton seedlings whenever the temperature drops below 15°C for a few hours during the first few days of germination. The fatty acid composition of the lipid components is proposed to be related to chilling sensitivity. Plants with more unsaturated fatty acids in the lipid components are believed to have greater cold tolerance. An experiment was designed to test the effect of temperature on the fatty acid composition of germinating cotton seedlings. Seed were germinated in growth chambers under eight different day/night temperature regimes. Seedlings were harvested 4, 6, 8 and 10 days after planting, separated into three vigor classes based on hypocotyl length, and cut into cotyledon, hypocotyl and radical fractions. The phospholipid fractions were extracted from the hypocotyls, and the fatty acids were methylated to form fatty acid methyl esters which were analyzed by gas/liquid chromatography. Linolenic acid (18:3) concentrations were higher in the longest seedlings regardless of temperature. Fatty acid composition and growth were correlated, suggesting that linolenic acid concentrations are related to growth rather than to temperature. The unsaturated/saturated (U/S) ratio did not appear to be related to seedling growth nor to temperature.

Introduction

Cotton (Gossypium hirsutum L.) is very sensitive to cool temperatures during germination. The current theory of cold tolerance in plants is based on the fatty acid composition of the lipid components. Based on this theory, the sensitivity of cotton seedlings to cool temperatures could either be due to the fatty acid composition of the storage oil in the seed or to the composition of the membrane lipids. Field germination of Pima cotton (Gossypium barbadense) at low soil temperatures was correlated to higher U/S ratios in the storage oil (Bartkowski, 1977). However, Speed (1995) found that germination of cotton (Gossypium hirsutum) at cool temperatures was related to higher levels of saturated fatty acids in the oil Seed with higher levels of saturated fatty acids were also more mature. This was corroborated by another study (Borth, 1997) analyzing the fatty acids of developing cottonseed. Immature seed had high levels of linolenic acid which decreased as the seed matured. The more mature seed had higher germination percentages than the immature seed. This seems to indicate that during germination, fatty acid composition of the storage oil has only a minor role in cold tolerance.

Cotton root tips show elevated levels of linolenic acid when cold acclimated (St. John, 1976). Work done in other species also suggests an important role for membrane fatty acids. It was found that chilling-sensitive lima beans had a lower U/S ratio than the more chilling-resistant broad beans and peas (Dogras, 1977). A study involving two potato species found that both species showed an increase in the U/S ratio after cold acclimation (Palta, 1993). Based on these studies, it was hypothesized that cotton grown under cooler temperatures would exhibit a higher U/S ratio and higher levels of linolenic acid than cotton grown under warmer temperatures. Hypocotyl tissue was analyzed because it is a frequent site of chilling injury.

Materials and Methods

Cottonseed (Paymaster HS26) were germinated in growth chambers under eight different day/night temperature regimes: 30/25, 30/20, 30/15, 30/10, 25/25, 25/20, 25/15 and 25/10. Seedlings were harvested 4, 6, 8 and 10 days after planting and separated into three vigor classes based on hypocotyl length. The phospholipid fraction in the hypocotyls was extracted using a chloroform:methanol (2:1) solvent. The fatty acids were hydrolyzed and methylated for analysis by gas/liquid chromatography. Regression analysis was used to correlate fatty acid composition with growth of the seedling.

Results

Growth increased over time, and rate of growth increased with temperature. Using Pearson correlation, percentages of oleic (18:1) and linoleic acid (18:2) were negatively correlated to growth while linolenic acid concentration was positively correlated to growth. The U/S ratio was not correlated to growth. The correlation coefficient of linolenic acid increased when it was expressed as a proportion of total unsaturated fatty acids (18:3/US) and as a ratio to linoleic acid (18:3/18:2) (Table 1). This suggests that the increase in linolenic acid occurs at the expense of other unsaturated fatty acids rather than changing the U/S As expected, linolenic acid concentration was ratio. highest in the longest vigor class under every temperature regime and harvest date. Linolenic acid also increased over time under each temperature regime (Tables 2and 3). Increase was more pronounced under cooler temperatures. At warmer temperatures, linolenic acid was higher at the first harvest date and remained high. This was probably due to the fact that the first harvest dates of plants grown under cooler temperatures represent an earlier stage of development than the same harvest dates under warmer temperatures. By days 8 and 10, all treatments had similar linolenic acid concentrations. This seems to relate linolenic acid concentration to growth and development

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rather than to temperature. Unsaturated/saturated fatty acid ratios were similar over time, under all temperatures, and across all vigor classes.

Conclusions

Linolenic acid concentration was related to seedling growth across all temperature conditions. Unsaturated/saturated fatty acid ratio was not related to growth or to temperature. Neither of these factors are able to explain cold tolerance, but linolenic acid concentration seems to be related to vigor. It is not possible to say at this point whether seedlings are more vigorous because they produce more linolenic acid or whether more vigorously growing seedlings are able to produce more linolenic acid.

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Table 1. Pearson correlation of fatty acids and growth. All correlation coefficients are significant (P<.05).

	Hypocotyl Length	Hypocotyl Dry Wt.		
% 18:3	.634	.700		
18:3/US	.647	.702		
18:3/18:2	.683	.692		
U/S	.174	.253		

Fable 2. % Linolenic acid at 25/10.						
Vigor Class						
Short	Intermediate	Long				
6.93	8.69	9.18				
8.17	10.35	11.55				
10.29	13.18	13.8				
12.59	12.38	14.55				
	Short 6.93 8.17 10.29	Vigor Class Short Intermediate 6.93 8.69 8.17 10.35 10.29 13.18				

Table 3.	%Linole	enic acid a	at 25/25.	

	Vigor Class		
Harvest Day	Short	Intermediate	Long
4 day	9.33	12.16	13.21
6 day	12.39	13.65	14.76
8 day	12.71	13.39	13.9
10 day	13.03	13.44	14.54