Leakage of Reducing Sugars and Amino Acids During Cotton (Gossypium hirsutum) Seed Imbibition

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ABSTRACT

Leakage of reducing sugars and amino acids during cotton seed imbibition was evaluated as a possible vigor test. Seed samples from a single cotton seed lot were subjected to accelerated aging at 46°C and 100% R.H. for up to 216 hours.Aged seeds were imbibed at optimal and suboptimal temperatures, and the leakage of total reducing sugars and amino acids into the imbibition solution was quantified spectrophotometrically. Leakage of seed reserves was positively correlated with the duration of accelerated aging, in terms of subsequent germination performance at 30°C and of similar quantity at both imbibition temperatures.

INTRODUCTION

Cool soil temperatures (below 18°C) are particularly stressful to germinating cotton seeds and only the most vigorous seeds emerge under such conditions.

The physiological basis of such temperature sensitivity is not known, but may be related to an increase in cellular permeability which causes leakage of seed reserve nutrients. Under field conditions, such leakage could promote the growth of cotton seed pathogens which in turn would weaken or kill the seeds. Thus, leakage of cotton seed reserve nutrients at low temperatures may correlate with seed vigor under stressful field conditions. To test this, the leakage of total reducing sugars and amino acids was measured in cotton seeds during early imbibition at low temperature.

MATERIALS AND METHODS

All seeds were from a single commercial lot of Deltapine 90 produced in Arizona in 1986 and donated by the Delta Pine & Land, Co. Samples from this seed lot were subjected to accelerated aging at 46°C and 100% relative humidity for up to 216 hours and then air-dried. A standard germination test of 4 days at 30°C in rolled paper towels was then performed on each artificially aged, seed lot sample.

Sixteen randomly selected seeds from each aged sample were imbibed individually in 0.2 ml water containing 0.1 mg kanamycin. Seeds were incubated in darkness for either 15 hours at 8°C or 11 hours at 32°C. A small sample was then removed from the imbibition solution and assayed for total reducing sugars or amino acids. Reducing sugars were assayed with a microassay based on the Nelson and Somogyi method. Amino acids were assayed with a microassay based on ninhydrin reagent. Assays were conducted in microtiter wells which also served as cuvettes for the microtiter plate spectrophotometer.

Results and DiscussionAccelerated aging at 46°C and 100% R.H. for durations greater than 48 hours reduced the germination of cotton seeds (Table 1). Shorter periods of aging were either stimulatory or without effect on germination.

Leakage of reducing sugars was positively related with duration of accelerated aging (Table 2). Seeds subjected to the greatest duration of accelerated aging leaked, on average, the most reducing sugars. Leakage was similar at both optimal and suboptimal imbibition temperatures. Leakage at the lower imbibition temperature was more closely correlated with standard germination results.

Leakage of amino acids was also positively related with duration of accelerated aging (Table 3). The relationship between the total amino acid leakage and the duration of accelerated aging was imbibition temperature dependent. At the optimal imbibition temperature, amino acid leakage increased linearly with the duration of accelerated aging. At the suboptimal imbibition temperature, seeds exposed to 48 h accelerated aging leaked about the same amount of amino acids as un-aged seeds. Longer durations of accelerated aging increased the leakage of amino acids during imbibition.

These results suggest that leakage of reserve nutrients during imbibition correlates with artificially induced vigor differences in cotton seeds. The physiological basis of this relationship remains unclear, however. Since cotton seeds leaked reserve nutrients at both optimal and suboptimal imbibition temperatures, selective promotion of seed pathogen growth under only cool soil temperatures seems an unlikely explanation for differences in cotton seed performance under stressful field conditions.

Table 1. Effect of accelerated aging on the germination percentage of cotton seeds. Seed samples from a single cotton seed lot were subjected to accelerated aging at 46°C and 100% R.H. for the designated intervals. A standard germination test of 4 days at 30°C in rolled paper towels was performed on each artificially aged seed lot sample.

Accelerated aging	Germination
h	% ± s.e.
0	79 ± 1.4
48	89 ± 2.1
96	63 ± 1.7
144	15 ± 1.5
216	0 ± 0.0

Table 2. Leakage of reducing sugars during imbibition at optimal and suboptimal temperatures of cotton seeds subjected to accelerated aging. Cottons seeds from a single seed lot were subjected to accelerated aging for up to 216 h at 46°C and 100% R.H. and then air-dried. Sixteen seeds from each aged sample were imbibed individually in 0.2 ml water at the indicated conditions. Relative amount of reducing sugars excreted from each seed into the imbibing solution was determined by a spectrophotometric microassay.

	Incubation conditions	
Accelerated aging	15 h at 8°C	11 h at 32°C
h	Absorbance, 490 nm ± s.e.	
0	0.13 ± 0.01	0.09 ± 0.04
48	0.12 ± 0.03	0.44 ± 0.19
96	0.31 ± 0.18	0.64 ± 0.21
144	0.51 ± 0.19	0.68 ± 0.16
216	1.65 ± 0.32	1.89 ± 0.21

Table 3. Leakage of amino acids during imbibition at optimal and suboptimal temperatures of cotton seeds subjected to accelerated aging. Cottons seeds from a single seed lot were subjected to accelerated aging for up to 216 h at 46°C and 100% R.H. and then air-dried. Sixteen seeds from each aged sample were imbibed individually in 0.2 ml water at the indicated conditions. Relative amount of amino acids excreted from each seed into the imbibing solution was determined by a spectrophotometric microassay.

	Incubation conditions	
Accelerated aging	15 h at 8°C	11 h at 32°C
h	Absorbance, 405 nm ± s.e.	
0	0.05 ± 0.01	0.09 ± 0.02
48	0.11 ± 0.02	0.82 ± 0.33
96	0.61 ± 0.25	1.37 ± 0.37
144	1.35 ± 0.31	1.85 ± 0.34
216	2.36 ± 0.25	> 3.00 ± 0.00