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Predicting Cotton Seedling Emergence for Cold Tolerance: *Gossypium hirsutum* L.

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Abstract

Breeding cotton for stress tolerance is a priory of most cotton breeding programs. One of the stresses for cotton is cold that affects lint and yield quality. Obtaining a stand of vigorous seedlings is a major problem in cotton production especially in the areas experiencing cool weather temperatures at the planting and seedling stages of cotton. Developing a cold screening technique special for cotton species is important in advanced breeding. A total of 95 cotton genotypes belonging to *G. hirsutum* L. were screened at 5 different laboratory tests to determine the best screening test to predict field emergence. Field emergence percentages ranged from 38% to 77% for the 28th day after planting. A standard germination test was performed at 30°C. Cool germination percentages were measured at 13°C, 15°C and 18°C. Multiple regressions were used to determine the degree of association between variables. Combinations of 30°C and 18°C tests were better than other tests alone in predicting field emergence rates.

Keywords: cotton, *Gossypium hirsutum* L., cold stress, screening

Introduction

Cotton producers have problems when scheduling the planting time. If they plant late in the season when soil temperatures are ideal for seedling emergence and stand establishment, they are faced with reduced fiber and seed quality resulting from maturation under the cool fall temperatures (Kittock *et al*., 1987). Conversely, when they plant early in the season so that crop maturation occurs under warmer fall conditions, seedling emergence and stand establishment are compromised due to the low early spring soil temperatures (Christiansen and Thomas, 1969). On the other hand, early planting showed yield improvement compared to normal planting, if it is not stressed by cold (Bange and Milroy, 2004). So, having good quality cotton seeds that are somehow tolerant to cold stress is an important issue in many breeding programs.

The cool germination test at 18°C was used by several researchers to test cold tolerance of different genotypes (Smith and Varvil, 1984; Durummond and Savoy, 1996; Tolliver *et al*., 1997; Savoy, 2005; Bolek, 2006). The metabolic chill test and imbibitional chill tests on sand are also used to test cold tolerant varieties with cold tolerance rating (Schulze *et al*., 1997; Duesterhaus *et al*., 2000). Cold and warm germination percentages were found better than either component alone (Kerby *et al*., 1989).

In most of the cotton planting areas of Turkey, planting occurs when the night temperatures are well below this minimum temperature requirement. For this reason minimum soil temperature falls below the 15°C. Early plantings when soils were cool generally reduced yields of Upland cotton, but these conditions had little detrimental impact on Pima cotton (Kittock *et al*., 1986).

In the GAP (South East Anatolian Project area) region, major cotton growing region in Turkey, planting usually performed after April 20th and most of the time occurs in the first or second week of May (Çopur, 1999). If more cold tolerant varieties of cotton could be developed, producers could utilize a longer growing season when a good stand establishment would be obtained under cool spring temperatures in addition to the crops ability to mature under the later cool fall temperatures (Buxton *et al*., 1976).

Identification and use of high quality planting seed is a priority of cotton growers in Turkey and in the world as well. Screening for cold tolerance in the field is sometimes not easy due to bad whether conditions in addition to soil borne diseases affecting germination and stand establishment. Using the most predictable laboratory screening technique for cold tolerance will help to save time and increase the efficiency of selection. Since cotton species have different growing habits and genetic construction, it is better to specify screening techniques for different species. *G. barbadense* L. cultivars were screened for five different laboratory tests to predict field emergencies (Bolek, 2006). The purpose of this research was developing a model to predict field emergence rates of *G. hirsutum* L. genotypes from laboratory experiments to relate cold tolerance. Results from this experiment could be used by growers as a tool for scheduling plantings, determining seeding rates and determining seed vigor.

Materials and methods

Field test

Two hundred seed of each 95 *G. hirsutum* L. genotypes were planted on each row on April 7, 2005. This date was

used to determine the response of seeds to low temperature stress. Seeding depth was 25 mm. The optimum planting date for the region is around May 15th. Stand counts were recorded once a week for a month after the planting date. Four counts were made from April 7th to May 4th. Soil temperature in the experimental area was averaged at 12°C for 5 cm in depth.

The experimental design was a randomized complete block design with 2 replications. The rows were 12 m length and 70 cm apart from each other. The soil type was alluvial. Soil moisture was adequate and irrigation was not required for seedling emergence. The field parameter measured for early season in terms of cold tolerance was the Field Emergence Percentage (total percent germination).

Laboratory test

Various laboratory tests were used to evaluate both seed and seedling from the entries. Standard 30°C and cool, 18°C, germination percentages as defined by AOSA (1983), procedures were determined. The tests were conducted on four replications of 50 seeds, which were planted on moistened germination towels (two on bottom, two on top) then rolled. Rolled towels were place upright in a less-than air tight container to prevent towels from drying too rapidly, to maintain high humidity and to provide proper aeration to germinating seeds. These containers were placed in germination equipment capable of maintaining 18°C. The duration of the test was seven days for 18°C and 4 days for 30°C. A final 7 day count was also performed to determine percent germination for 30°C. At this time one count of normal seedlings that had a combined hypocotyl and root length of 3.75 cm (1.5 inch) or longer were made. The root-hypocotyl measurement was made from the point of cotyledon attachment to the tip of the radicle. Standard germination, defined as the percent of healthy seedlings reaching 38 mm after 7 days at 30°C

Tab. 1. Comparison of independent variables against percent field emergence, 7 days after planting

Independent variables and days		r^2	P	Regression equation
13°C Germination Percentage on Paper (Day 7 Counts) (hypocotyl >1 cm)	linear	0.11	0.001	y = 7.0572 + 0.8675x
	quadratic	0.16	0.000	y = 6.6921 + 2.2123x - 0.1419x2
	Cubic	0.17	0.001	y = 6.8254 + 0.8537x + 0.2032x2 - 0.0186x3
15°C Germination Percentage on Paper (Day 7 Counts) (hypocotyl + root length >3.8 cm)	linear	0.12	0.001	y = 5.3476 + 0.1546x
	quadratic	0.17	0.000	y =3.4435 + 0.4585x - 0.0065x2
	Cubic	0.17	0.001	y = 3.0555 + 0.5666x - 0.0118x2 + 6.E5x3
18°C Germination Percentage on Paper (Day 7 Counts)	linear	0.20	0.000	y = 0.5350 + 0.1528x
	quadratic	0.20	0.000	y = 1.8656 + 0.0853x + 0.0007x2
	cubic	0.22	0.000	y = 7.4631 - 0.4255x + 0.0133x2 -9.E5x3
30°C - 18°C Germination Percentage on Paper (Day 4 Counts)	linear	0.12	0.001	y = 12.1885 - 0.1119x
	quadratic	0.15	0.001	y = 10.0915 + 0.0457x - 0.0021x2
	cubic	0.19	0.000	y = 10.3374 + 0.2244x - 0.0103x2 + 8.00001x3
30°C - 18°C Germination Percentage on Paper (Day 7 Counts)	linear	0.17	0.000	y = 14.0002 - 0.1445x
	quadratic	0.17	0.000	y = 13.4820 - 0.1161x - 0.0003x2
	cubic	0.20	0.000	y = 7.1302 + 0.4797x - 0.0155x2 + 0.0001x3
30°C + 18°C Germination Percentage on Paper (Day 4 Counts)	linear	0.18	0.000	y = -7.1065 + 0.1116x
	quadratic	0.20	0.000	y = 20.1206 - 0.3115x + 0.0016x2
	cubic	0.20	0.000	y = 10.8872 - 0.0988x + 3.000009x3
30°C + 18°C Germination Percentage on Paper (Day 7 Counts)	linear	0.18	0.000	y = -9.2416 + 0.1238x
	quadratic	0.20	0.000	y = 19.3787 - 0.3028x + 0.0015x2
	cubic	0.20	0.000	y = 9.5090 - 0.0864x + 3.000006x3
30°C / 18°C Germination Percentage on Paper (Day 4 Counts)	linear	0.08	0.004	y = 10.5448 - 1.1328x
	quadratic	0.16	0.000	y = 15.6826 - 4.7096x + 0.3792x2
	cubic	0.16	0.001	y = 17.6147 - 6.6554x + 0.8768x2 - 0.0330x3
30°C / 18°C Germination Percentage on Paper (Day 7 Counts)	linear	0.09	0.004	y = 10.5896 - 1.0963x
	quadratic	0.18	0.000	y = 16.5012 - 5.0016x + 0.3916x2
	cubic	0.20	0.000	y = 20.4245 - 8.6991x + 1.2744x2 - 0.0550x3
30°C * 18°C Germination Percentage on Paper (Day 4 Counts)	linear	0.21	0.000	y = 1.3136 + 0.0016x
	quadratic	0.21	0.000	y = 2.6259 + 0.0008x + 8.00000005x2
	cubic	0.22	0.000	y = 6.7879 - 0.0033x + 1.000002x2 - 8.E11x3
30°C * 18°C Germination Percentage on Paper (Day 7 Counts)	linear	0.20	0.000	y = 1.0522 + 0.0016x
	quadratic	0.21	0.000	y = 2.6255 + 0.0007x + 9.00000006x2
	cubic	0.21	0.000	y = 5.9297 - 0.0025x + 9.0000003x2 - 6.E11x3

on paper towels. Combinations of 18°C and 30°C tests were also calculated.

Four replications of 50 seed were subjected to a 24 hour imbibition period in rolled foam pads containing 100 ml of 5°C water then planted in a controlled environment room in sand at a constant 18°C to evaluate for early season cold tolerance (Schulze *et al*., 1997). Seeds were planted in plastic boxes on a 3.8 cm layer of sand at field capacity and another 3.8 cm of dry sand covering the seed. Germination counts were made 7th, 14th and 21st days after planting the seeds. Sand was autoclaved before it was used for planting.

Cool, 13°C and 15°C tests were conducted as standard germination test. Counts of germinations were made 7th, 14th and 21st day after planting. Seedlings having hypocotyl length greater than 1 cm and 2 cm were counted on day 7, 14 and 21 days, respectively. Normal seedlings that had a combined hypocotyl and root length of 3.75 cm (1.5 inch) or longer were made at 15°C germination percentages after 7 days of planting.

The multiple regression analysis was used to relate laboratory germinations to field emergence percentages.

Results and discussion

Ninety five *G. hirsutum* L. genotypes were screened for cold tolerance in the laboratory and field to predict the field emergence percentage from laboratory germination and emergence percentages. The following germination conditions were evaluated: 13°C, 15°C and 18°C (both in paper towels and in sand) and 30°C. Combinations of 18°C and 30°C germination percentages were also calculated.

Predicting field emergence 13°C, 15°C and 18°C (on paper towels) yielded significant relations (P=0.05). 22% of the variation in the field emergence at day 7 was explained by 18°C germination percentages on paper towels. The regression equation for this relationship is y=6.7879-0 .0033x+1.000002x^2-8.E11x^3. Having significant relations, variations explained by laboratory tests were low (Tab. 1). This might be caused by different genotypic effects. No significant relations were obtained between 18°C emergence on sand and field emergence 7 days after planting. The variability explained by the all laboratory tests ranged from 11% to 22%. On the other hand, combinations of 30°C and 18°C tests did not increased to predicted values for the 7 days following field emergence (Tab. 1).

Field emergence 14 days after planting was best predicted by 13°C and 15°C germination percentages but the variability explained was between 0.08% and 0.11% (Tab.

Tab. 2. Comparison of independent variables against percent field emergence, 14 days after planting

Independent variables and days		r^2	P	Regression equation
13°C Germination Percentage on Paper (Day 7 Counts) (hypocotyl >1 cm)	linear	0,11	0.001	y = 18.9662 + 1.4587x
	quadratic	0,12	0.004	y = 18.7690 + 2.1851x - 0.0766x^2
	cubic	0,13	0.006	y = 19.0191 - 0.3639x + 0.5707x^2 - 0.0348x^3
15°C Germination Percentage on Paper (Day 7 Counts) (hypocotyl >3.8 cm)	linear	0,08	0.006	y = 16.9362 + 0.2099x
	quadratic	0,11	0.005	y = 14.3596 + 0.6211x - 0.0088x^2
	cubic	0,11	0.012	y = 13.5089 + 0.8582x - 0.0205x^2 + 0.0001x^3
18°C Germination Percentage on Paper (Day 7 Counts)	linear	0,06	0.013	y = 13.4652 + 0.1444x
	quadratic	0,07	0.028	y = 18.7120 - 0.1219x + 0.0028x^2
	cubic	0,09	0.037	y = 28.3168 - 0.9984x + 0.0245x^2 - 0.0002x^3
18°C Emergence Percentage on Sand, (Day 14 Counts)	quadratic	0,07	0.033	y = 13.7419 - 0.2023x + 0.0033x^2
	cubic	0,07	0.033	y = 7.1692x + 0.0559x + 0.00014x^3
30°C + 18°C Germination Percentage on Paper (Day 4 Counts)	linear	0,04	0.049	y = 8.2921 + 0.0903x
	quadratic	0,10	0.009	y = 75.0508 - 0.9472x + 0.0039x^2
	cubic	0,09	0.012	y = 52.3106 - 0.4245x - 9.000004x^3
30°C + 18°C Germination Percentage on Paper (Day 7 Counts)	linear	0,05	0.033	y = 5.5129 + 0.1077x
	quadratic	0,08	0.019	y = 73.6054 - 0.9072x + 0.0037x^2
	cubic	0,08	0.022	y = 50.0910 - 0.3920x + 8.000006x^3
30°C / 18°C Germination Percentage on Paper (Day 7 Counts)	quadratic	0,06	0.054	y = 29.2430 - 5.3627x + 0.4646x^2
	cubic	0,09	0.031	y = 38.6473 - 14.226x + 2.5806x^2 - 0.1317x^3
30°C * 18°C Germination Percentage on Paper (Day 4 Counts)	linear	0,06	0.017	y = 14.4878 + 0.0014x
	quadratic	0,08	0.028	y = 19.9884 - 0.0017x + 3.0000006x^2
	cubic	0,09	0.034	y = 28.4627 - 0.0101x + 2.000006x^2 - 2.E10x^3
30°C * 18°C Germination Percentage on Paper (Day 7 Counts)	linear	0,06	0.016	y = 14.1253 + 0.0014x
	quadratic	0,07	0.030	y = 19.4926 - 0.0015x + 3.0000003x^2
	cubic	0,08	0.051	y = 26.0477 - 0.0078x + 2.0E6x^2 - 1.E10x^3

Tab. 3. Comparison of independent variables against percent field emergence, 21 days after planting

Independent variables and days		r^2	P	Regression equation
13°C Germination Percentage on Paper (Day 7 Counts) (hypocotyl >1 cm)	linear	0,07	0.009	$y = 39.8856 + 1.2926x$
	quadratic	0,07	0.030	$y = 39.7420 + 1.8214x - 0.0558x^2$
	cubic	0,13	0.006	$y = 40.2959 - 3.8245x + 1.3780x^2 - 0.0772x^3$
30°C + 18°C Germination Percentage on Paper (Day 4 Counts)	linear	0,05	0.034	$y = 26.8056 + 0.1069x$
	quadratic	0,07	0.030	$y = 76.7245 - 0.6689x + 0.0029x^2$
	cubic	0,07	0.034	$y = 59.5288 - 0.2758x + 7.0E6x^3$
18°C Emergence Percentage on Sand, (Day 14 Counts)	linear	0,10	0.002	$y = 5.8540 + 0.4222x$
	quadratic	0,10	0.009	$y = -27.016 + 1.2590x - 0.0053x^2$
	cubic	0,10	0.009	$y = -17.537 + 0.8697x - 2.00001x^3$
18°C Emergence Percentage on Sand, (Day 21 Counts)	linear	0,08	0.004	$y = 6.6268 + 0.4043x$
	quadratic	0,09	0.017	$y = -20.989 + 1.0944x - 0.0042x^2$
	cubic	0,09	0.016	$y = -13.415 + 0.7804x - 2.00001x^3$
30°C Germination Percentage on Paper (Day 7 Counts)	quadratic	0,09	0.012	$y = 276.515 - 5.9993x + 0.0373x2$
	cubic	0,10	0.010	$y = 119.068 - 0.0377x^2 + 0.0003x^3$

2). The best regression equation is $y=19.0191-0.3639x+0.5707x^2-0.0348x^3$ for 13°C germination percentages.

Significantly related to field emergence percentages at 21 days after planting were 13°C and 30°C germination as well as 18°C emergence on day 14 and 21 (Tab. 3). Genotypic distribution for field emergence and variables are shown in Fig. 1. Germination percentages at 18°C ranged from 7.78% to 86.67% while they falling between 60.83% and 99.17% at 30°C. On the other hand, 13°C and 15°C germination percentages ranged from 0% to 14.17% and from 0% to 62.50%, respectively (Fig. 1). Averaged percent

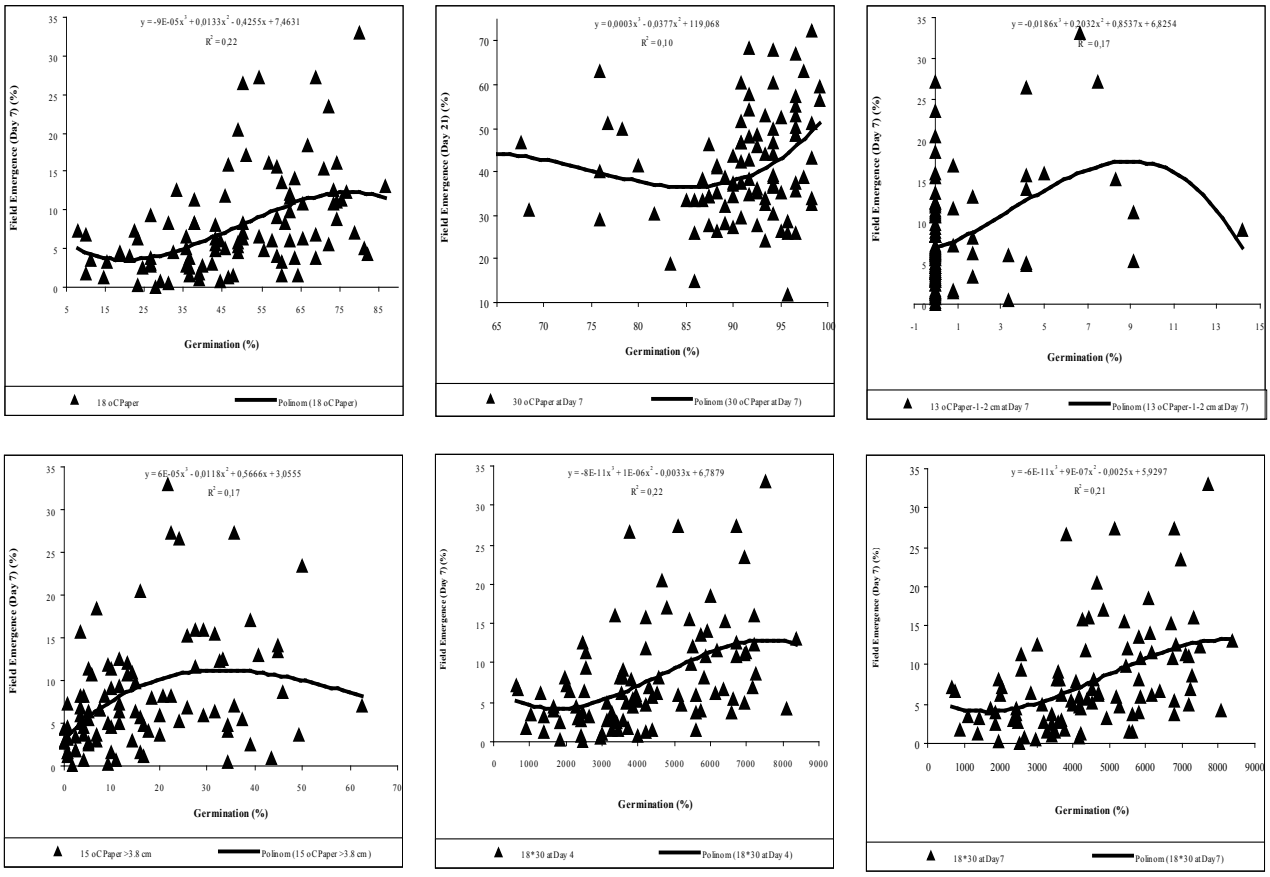

Fig. 1. Field emergence percentages as predicted for 18°C, 30°C, 30°C , 15°C, and 30°C / 18°C germination percentages, 7, 14 and 21 days after planting (DAP)

germinations at 13°C and 15°C were 1.04% and 16.87%, respectively.

Conclusions

Predicting field emergence percentages from laboratory experiments are most reliable if both 30°C and 18°C germination values were used. Relationships were strong at 7 days after planting in the field. Since significant variations still exist on day 21, field emergence counts should be continued until then.

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