Inheritance and Combining Ability of the Stay Green Trait in Sunflower

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Abstract

Plants which retain their green color at physiological maturity are referred to as stay green. Research on inheritance of the stay green/early senescence trait may provide valuable information to plant breeders for developing new cultivars with better resistance to stress environments and pests. This study was conducted to investigate inheritance of the stay green trait among two female and two male sunflower (*Helianthus annuus* L.) inbred lines and determine the general (GCA) and specific (SCA) combining abilities of different female and male inbred lines in F_1 hybrids for this character. Change in stem color was used as a criterion for the stay green/early senescence characteristic. Results of both studies indicated that additive gene effects were more important than nonadditive gene effects in controlling stay green trait in sunflower. Since additive effects were predominant, selection for this trait can be made in early generation segregating populations. Significant, but small correlation coefficients for stem color observed at different sampling dates indicated that selection would be most effective at physiological maturity.

Key Words: Sunflower, *Helianthus annuus*, breeding, genetics, early senescence.

Introduction

The stay green trait in sorghum [*Sorghum bicolor* (L.) Moench] and corn (*Zea mays* L.) is often associated with good plant health and increased plant resistance to insects and diseases (Walulu, 1994; Duvick, 1992). In sorghum, stay green hybrids also tolerated post-flowering drought conditions better than hybrids which senesce at physiological maturity. The mode of inheritance of stay green in sorghum differed among genotypes (Rosenow et al., 1988).

Although differences for the stay green character have been observed among sunflower inbred lines and hybrids, no research has been conducted to study the effect of this trait on the sunflower crop. Also, differences in seed moisture content were observed among sunflower genotypes (Chervet et al., 1988). However, the relationship between the stay green trait and seed moisture content is not known. The main objectives of this study were 1) to investigate the genetic factors controlling the stay green character among two female (B) and two male (R) sunflower inbred lines, 2) to determine the general and specific combining abilities of different female and male inbred lines in F_1 hybrid combinations for the stay green character and seed moisture content, and 3) to determine the relationship between stem color and seed moisture content at different developmental stages.

Materials and Methods

Crosses were made between two B-lines, HA234 and HA290, and two R-lines, RHA377 and RHA274, to study the inheritance of the stay green characteristic in sunflower.

HA234 and RHA377 had the stay green characteristic, whereas HA290 and RHA274 possessed early senesce. Parental, F_1, F_2, BCP_1 (P_1 x F_1), and BCP_2 (P_2 x F_1) generations were grown in the field at Fargo, ND, in 1992. Change in stem color was used as a criterion for the stay green/plant senescence characteristic. Stem color was analyzed by a computer program called Map and Image Processing System (MIPS). MIPS was based on a color palette ranging from 1 (dark green) to 40 (dark brown).

The continuous distribution of the segregating F_2 populations led to the difficulty of placing plants into phenotypic classes. Consequently, information on the nature of the gene effects involved in stem color was obtained by a weighted least square analysis with joint scaling tests based on the methods established by Hayman and Mather (1955) and Mather and Jinks (1971). Genetic parameters based on this weighted least square analysis were estimated according to the expectations derived by Hayman (1958), in which the F_2 is the reference population.

For the combining ability study, 36 hybrids were developed by crossing six cytoplasmic male sterile female lines (A-lines) to six male lines (R-lines) in a factorial mating design or Design II of Comstock and Robinson (1948). The 36 hybrids were planted at Casselton and Edgeley, ND, in 1991, and Casselton and Sheldon, ND, and Glyndon, MN, in 1992. The experimental design was a 6 x 6 lattice with three replicates. Three plants from each plot were selected for color evaluation of mid-stem at the R7, R8, and R9 plant stages (Schneiter and Miller, 1985). Also, seed samples from the same plants were taken at the R8 and R9 stages to determine seed moisture content. The whole plot was included in determination of seed moisture content at harvest.

The hybrid sum of squares was partitioned into variation due to females, males, and the female x male interaction. The main effects of females and males are equivalent to general combining ability (GCA), and the female x male interaction is equivalent to specific combining ability (SCA) effects. For each character, general combining ability effect for each line and specific combining ability effect for each F_1 hybrid were calculated according to Beil and Atkins (1967). Standard errors for GCA effects of female and male lines and SCA effect were calculated by using the method described by Cox and Frey (1984). Two-tailed *t*-tests were used to test the significance of the GCA and SCA effects, where $t = GCA/SE_{GCA}$ and $t = SCA/SE_{SCA}$, respectively (Singh and Chaudhary, 1977). The Pearson product-moment correlations were calculated for selected traits to study the interrelationships between these traits.

Results and Discussion

Inheritance Study

Generation mean analyses tested three- and six-parameter models for the best fit to explain control of stem color in the RHA377 x RHA274 and HA234 x HA290 crosses. Effects were estimated, and the model was developed by sequentially adding parameters into the model. Chi-square values for the three-parameter model were checked for the probability of obtaining a larger χ^2 value. The three-parameter model was found to sufficiently explain the control of stem color in the HA234 x HA290 cross, but not in the RHA377 x RHA274 cross (Table 1). Epistasis was not involved in the inheritance of the trait in the HA234 x HA290 cross. Additive effects (-6.00 ± 0.35) appeared to be the most important factor contributing to the genetic control of stem color in this cross. Dominance effects were positive, and the estimated value was 2.18 ± 0.79. These results indicated partial dominance for stem color, but a predominance of additive action in control of this trait.

The results of the three-parameter model analysis indicated that epistasis was important in the RHA377 x RHA274 cross (Table 1). Therefore, the six-parameter model was fitted to determine the type and magnitude of gene action involved in the inheritance of stem color. Both additive (-4.03 ± 0.40) and dominance (-4.87 ± 1.37) effects contributed significantly to the inheritance of stem color in this cross. The additive x dominance effects were the only epistatic estimate (1.90 ± 0.53) that was significant.

The negative sign indicated for d, additive effects, was of no significant importance as it was a function of which parent was chosen as P_1. The sign of h, dominance effects, was a function of the F_1 mean value in relation to the mid-parent value and indicated which parent was contributing to the dominance effect. Dominance effects in the RHA377 x RHA274 cross were contributed by the genes differing in the RHA377 parent whereas in the HA234 x HA290 cross it was contributed by the HA290 parent. All estimated effects were cumulative or pooled over all loci which differed between the two parents.

Variability accounted for by the estimated effects was examined (data is not presented). Additive effects, d, accounted for the largest portion of the variability, 90% in the RHA377 x RHA274 cross and 97% in the HA234 x HA290 cross. The six-parameter model in the RHA377 x RHA274 cross explained more than 99% of the variability.

Combining Ability Study

Significant differences among hybrids were observed for stem color at physiological maturity (the R9 plant stage) and seed moisture content at harvest (Table 2). Significant hybrid x environment interactions suggested the need for evaluations of hybrids in different environments for these traits.

Both general combining ability effects of males and females, and specific combining ability effects were significant for stem color at the R9 plant stage and harvest seed moisture content (Table 2). High ratios of GCA to SCA effects suggested that additive gene effects were relatively more important than nonadditive gene effects in the variation expressed among hybrid combinations. However, significant SCA effects also implied the contribution of the nonadditive gene effects to the variation expressed among hybrids.

At physiological maturity (the R9 plant stage), stem color of hybrids ranged from 21 (light green) to 31 (brown). Crosses with HA234 had the greenest stem color, except for RHA274 and RHA299 combinations (Table 3). If hybrids with stay green character are of interest, then a negative GCA effect of lines would be desirable for breeding. In other words, lines with negative GCA effects will be good combiners for the development of stay green hybrids. The hybrid combination HA89 x RHA271 had the highest negative SCA effect. The hybrid HA89 x RHA377, on the other hand, had the highest positive SCA effect, meaning that it senesced earlier than expected based on the average performance of hybrids with those parental lines.

Similar to stem color, variation in seed moisture content among hybrid combinations was mainly due to additive gene effects (Table 2). These findings agreed with the conclusions of Chervet et al. (1988) who studied genetic control of seed moisture content and its relationship with other agrophysiological traits in sunflower. In contrast to stem color, the male mean square was larger than the female mean square at harvest, indicating that the males in this study had greater influence on seed moisture content. At harvest, the female line HA821 had the highest negative GCA effect indicating lower seed moisture content than average (Table 4). The male lines RHA299 and RHA274 had the highest negative GCA effects. Hybrids with RHA298 and RHA273, on the average, had higher seed moisture content at harvest than hybrids with other male lines. If the main objective of a breeding program is to develop hybrids that senesce early and have low seed moisture content at harvest, the female line HA821 and the male lines RHA274 and RHA299 could be considered as good combiners. On the other hand, it was possible to develop hybrids, such as those with HA234 as a parent, which had the stay green trait as well as low harvest moisture content. The highest negative SCA effect at harvest was observed with hybrid HA234 x RHA298, whereas the crosses with the highest positive SCA effects were HA234 x RHA273 and HA234 x RHA271 (Table 4).

The results of both studies showed significant correlation between the first and third sampling dates for stem color; however, the degree of association was not very strong. Therefore, determination of stem color should be made at a later stage. No significant correlation was found between seed moisture content and stem color at any sampling date. Hybrids can be developed with the stay green trait and still have low harvest moisture.

Since both studies indicated predominantly additive gene effects in controlling stem color, breeding schemes that take advantage of additive gene action can be applied to develop lines with the stay green trait. The variability among the inbred lines made it possible to develop hybrids for diverse environmental conditions without sacrificing yield or oil content. It also is important that hybrids which senesce early might be developed for short-season production areas without yield loss.

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Table 1. Estimates of parameters, with standard errors, and χ^2 values for three- and six- parameter models for stem color of plants observed at physiological maturity for HA234 x HA290 and RHA377 x RHA274 crosses grown at Fargo in 1992.

Model and	Cross	
effect†	HA234 x HA290	RHA377 x RHA274
Three-parameter		
m	24.26 ± 0.16	21.20 ± 0.13
d	-6.00 ± 0.35	-4.98 ± 0.26
h	2.18 ± 0.79	-3.09 ± 0.66
χ^2	1.51	17.16
P‡	0.50-0.75	< 0.005
Six-parameter		
m	24.36 ± 0.21	21.50 ± 0.22
d	-5.63 ± 0.59	-4.03 ± 0.40
h	1.54 ± 1.75	-4.87 ± 1.37
i	-0.73 ± 1.44	2.10 ± 1.14
j	0.72 ± 0.75	1.90 ± 0.53
l	0.29 ± 3.18	2.94 ± 2.29

† Parameters estimated were m = mean of F_2, d = sum of additive effects, h = sum of dominance effects, i = sum of additive x additive epistatic effects, j = sum of additive x dominance epistatic effects, and l = sum of dominance x dominance epistatic effects.

‡ P value is probability of obtaining a larger χ^2 value.

Table 2. Analysis of variance combined across five environments for stem color at physiological maturity (the R9 plant stage) and harvest seed moisture content in 36 hybrids.

Source of variation	df		Mean square	
			Stem color	Seed moisture
Environments (E)	4		123.376	39456.566
Hybrids (H)	35		94.376**	2451.435*
Female (F)		5	311.581**	4590.330**
Male (M)		5	237.721**	8553.221**
F x M		25	22.266**	803.299**
H x E	140		23.008**	621.062**
F x E		20	62.342**	1190.048**
M x E		20	44.257**	1348.515**
F x M x E		100	10.892**	361.775**
Pooled error	245		3.134	60.540

*, ** Significant at the 0.05 and 0.01 probability levels.

Table 3. Stem color, specific combining ability effects, and general combining ability effects of sunflower inbred lines crossed to produce hybrids grown in five environments in 1991 and 1992.

Female	Male line						
line	RHA271	RHA274	RHA299	RHA377	RHA273	RHA298	GCA_f[§]
HA89	23[†] -2.28[‡]	28 -0.55	29 0.18	30 2.92	24 -0.76	26 0.5	0.77
HA821	26 0.36	29 -0.06	27 -1.61	28 1.04	25 -0.02	27 0.29	1.16
HA124	24 0.63	25 -1.70	27 0.69	23 -1.35	24 1.71	24 0.03	-1.48
HA372	24 -0.14	29 1.57	27 -0.46	25 -0.98	23 -0.24	25 0.26	-0.37
HA234	21 -0.53	26 1.04	25 0.43	22 -1.05	21 0.51	22 -0.41	-2.76
HA290	29 1.97	30 -0.30	31 0.77	28 -0.57	25 -1.19	27 -0.67	2.68
GCA_m[¶]	-1.34	1.99	1.86	0.36	-2.18	-0.70	

[†] LSD (0.05)= 3.64.
[‡] SCA effects: SE_{SCA}= 0.75, SCA= ±1.48.
[§] Female line GCA effects: SE_{GCAf}= 0.80, GCA_f= ±1.66.
[¶] Male line GCA effects: SE_{GCAm}= 0.67, GCA_m= ±1.40.

Table 4. Seed moisture content (g kg^{-1}), specific combining ability effects, and general combining ability effects of sunflower inbred lines crossed to produce hybrids grown in five environments in 1991 and 1992.

Female	Male line						
line	RHA271	RHA274	RHA299	RHA377	RHA273	RHA298	GCA_f[§]
HA89	114.73[†] 6.93[‡]	85.01 -5.65	96.32 5.91	86.53 -10.81	114.86 6.01	112.24 -2.40	6.87
HA821	82.72 -6.72	78.28 5.97	75.79 3.74	82.27 3.29	86.12 -4.38	94.38 -1.91	-11.49
HA124	110.00 1.80	91.17 0.11	85.56 -5.24	102.75 5.01	103.33 -5.92	119.26 4.23	7.25
HA372	95.96 -8.85	84.33 -3.34	89.23 1.82	94.51 0.17	106.07 0.21	121.62 9.98	3.86
HA234	112.62 12.31	81.02 -2.15	76.90 -6.02	89.46 -0.39	113.71 12.34	91.05 -16.10	-0.63
HA290	89.63 -5.47	83.01 5.05	77.48 -0.22	87.35 2.72	87.87 -8.27	108.11 6.18	-5.85
GCA_m[¶]	6.19	-10.95	-11.21	-4.28	7.24	13.02	

[†] LSD (0.05)= 18.89.
[‡] SCA effects: SE_{SCA}= 4.30, SCA= ±8.54.
[§] Female line GCA effects: SE_{GCAf}= 3.48, GCA_f= ±7.27.
[¶] Male line GCA effects: SE_{GCAm}= 3.71, GCA_m= ±7.74.