

Diallel analysis of grain filling rate and grain filling period in tropical maize (*Zea mays* L.)

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Abstract Grain yields of maize in the lowland tropics are generally limited by short days and high temperatures that minimize durations of incident light. Little has been published on the effects of this limiting environment on the genetics of grain filling rate (GFR), and grain filling period (GFP) in tropical maize germplasm. This study sought to address these limitations. A set of 8 elite maize inbreds of tropical origin and their 28 diallel hybrids were grown in three seasons at Waimanalo, Hawaii, USA. Seasonal differences included > 100% differences in values of photosynthetic active radiation (PAR) during grain formation. Information was sought on the performance variations and the genotype by season interactions for GFR, GFP, days to mid-silk (DTS), kernel weight, with estimates of general (GCA) and specific combining ability (SCA) and their interactions with seasons. Significant differences occurred for inbreds, hybrids, and genotype by season interactions, GCA and SCA effects and their interactions with seasons, which could be attributed primarily to the differences in PAR values among seasons in Hawaii during grain filling. Additive genetic effects predominated for GFR

and GFP. Breeding approaches that take advantage of additive gene effects including hybrid breeding with evaluations in multiple Hawaii seasons may be used to alter GFR and GFP.

Keywords General combining ability (GCA) · Grain filling rate (GFR) · Grain filling period (GFP) · Kernel weight (KW) · Photosynthetic active radiation (PAR) · Specific combining ability (SCA)

Introduction

Light is a major limiting factor for maize grain yields in the tropics. Short days combine with high cloud cover during rainy seasons to minimize photosynthetically active radiation (PAR). Summer yields in Hawaii are almost double those of winter (Jong et al. 1982), reflecting the coincidence of Hawaii's short winter days with the rainy season. Light intercepted by plants during grain filling is reduced up to 50% in winter (Brewbaker 2003). A breeding approach that has been suggested to increase yield involves increasing the effective grain-filling period (GFP) (Carter and Poneleit 1973; Poneleit and Egli 1979). Alternatively, breeders might attempt to increase grain-filling rates (GFR) (Daynard and Kannenberg 1976; Jones et al. 1979). Gasura et al. (2014) found that selection for increased yield in early maturing tropical maize

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germplasm in Zimbabwe could be achieved by selecting for higher GFR and GFP.

Grain development in maize involves a short lag period (LAG) followed by an essentially linear phase of the GFP. The LAG extends from syngamy until the start of dry matter accumulation involving endosperm cell development and sugar accumulation prior to starch synthesis (Reddy and Daynard 1983). A standardized system of nomenclature in maize grain filling was proposed by Salvador and Pearce (1995) dividing LAG into a dilatory phase characterized by slow dry matter accumulation, and an exponential phase in which dry matter accumulation starts to increase, progressing from the base to tip of the ear. This is followed by the linear filling phase where there is rapid and constant accumulation of dry matter, and finally the attenuation phase characterized by the onset of kernel black layer formation. Mathematically, Johnson and Tanner (1972) defined GFR as the slope of the linear regression of kernel weight over time until kernel physiological maturity. A previous study characterizing field corn inbreds in Hawaii revealed GFRs of $\sim 7 \text{ mg kernel}^{-1} \text{ day}^{-1}$ at an average temperature of $28 \text{ }^\circ\text{C}$, declining to $\sim 4 \text{ mg kernel}^{-1} \text{ day}^{-1}$ at $22 \text{ }^\circ\text{C}$ (Fahrner 1991).

Determination of physiological maturity and termination of GFP has been approached with different methods. A common approach to determining maturity established by Daynard and Duncan (1969) is to use the grain “black-layer”, an abscission layer that blocks translocation of assimilates to the kernel. However, black layer is often difficult to see in tropical flint inbreds in Hawaii, as cited also by Maddonni et al. (1998) in Argentina. Daynard (1972) also indicated that black layer could be induced prematurely by low temperatures. Alternatively, Sala et al. (2007) demonstrated that physiological maturity was attained at 35% grain moisture estimated using a wide range of maize genotypes. We chose to use the method of Johnson and Tanner (1972), a statistical approach determined by dividing final kernel weight (KW) by the GFR.

Several studies in maize have revealed that GFR and GFP are controlled predominantly by additive gene action (Cross 1975; Ottaviano and Camussi 1981; Katsantonis et al. 1986; Wang et al. 1999). These studies however are associated mainly with temperate germplasm and current information on the genetic control of GFR and GFP in Hawaii tropical

maize germplasm representing diverse maturities, and seasons are limited (Fahrner 1991). Cross (1975) analyzed a 7 entry diallel in North Dakota where incident light values approximately double those in Hawaii. In this study, general combining ability (GCA) effects predominated for GFR and GFP. Ottaviano and Camussi (1981) in Italy found significant GCA and specific combining ability (SCA) effects, with high GCA to SCA ratios for both GFR and GFP. The GCA mean squares were also significantly greater than SCA for yield per plant, kernel row number and kernels per row. Wang et al. (1999) examined GCA and SCA effects for GFR and GFP in 8 inbred lines crossed in a 4×4 mating scheme (North Carolina Design II) at Baton Rouge, Louisiana. The GCA again explained most of the variation in GFR, GFP and black layer maturity, while SCA was more significant for kernel numbers.

Maize yields are limited greatly in Hawaii by reduced day lengths and cloud cover during the winter rainy seasons (Jong et al. 1982). Because it is known that GFR and GFP are correlated with maize grain yield (Daynard and Kannenberg 1976; Jones et al. 1979; Gasura et al. 2014), it may be conjectured that yields under these conditions in Hawaii can be improved significantly by breeding for increased GFR or GFP. There is very limited data describing the genetics of GFR and GFP in tropical Hawaii maize with broader maturities and in multi-season growing environments with significant light and temperature variations.

The objectives of the present study of tropical maize germplasm in Hawaii were to, (1) characterize seasonal variation in PAR and temperature during kernel filling, (2) determine variation in entry and entry by season interactions for GFR, GFP, days to mid-silk (DTS), and KW, (3) characterize individual inbred and F_1 hybrid performance for these traits, and (4) to determine GCA and SCA effects for GFR, GFP, DTS and KW.

Materials and methods

Genetic materials

Eight elite maize inbreds released by Hawaii Foundation Seeds (HFS) of the College of Tropical Agriculture and Human Resources of the University of Hawaii at Manoa (UHM), representing diverse heterotic

groups were chosen as parents for the diallel analyses in this study (Table 1). Each inbred was an isogenic line of an elite inbred converted through six or more backcrosses to resistance for maize mosaic virus in Hawaii (Brewbaker 1997; Brewbaker and Josue 2007). All eight have been used in tropically-adapted Hawaiian hybrids of high yield potential. Inbred Hi26 was based on CM202 from India, which in turn was derived from M. T. Jenkin's denty CI21E. Inbred Hi53 derives from one of F. Arboleda's most significant Colombian flint inbreds, while Hi57 is from Thailand's Ki9 (formerly KU1409). Inbred Hi60 was derived from M. Zuber's famous inbred Mo17. Inbred Hi61 was based on H. C. Arnold's inbred N3, female parent of the Southern Rhodesian white dent hybrid SR52 that revolutionized African corn agriculture (McCann 2005). Inbreds Hi62, Hi65 and Hi67 were of similar importance in the Philippines, southern USA and West Africa, respectively.

The eight inbreds were crossed in a diallel excluding reciprocals (Griffing 1956) in August 2003 at Hawaii's Waimanalo Research Station. The 28 F_1 crosses and the 8 inbred parents were evaluated in a modified RCB with two replications. The F_1 hybrids and inbreds were blocked separately and randomized separately within blocks.

Complete fertilizer (16–16–16) was applied as a basal application prior to planting at a rate of 80 kg of N and 80 kg of P_2O_5 and 80 kg K_2O ha^{-1} . Alachlor[®] and Atrazine[®] were applied as pre-emergence herbicides and post-emergence herbicides were applied as needed. Trials were irrigated using a drip tube system. Entries were grown in two row plots 5 m long spaced 0.75 m apart. Two to three seeds were planted per hill using a hand jab planter at 0.25 m spacing and thinned to one plant (53,333 plants ha^{-1}) at around the V5–V8 leaf stage. Urea (46–0–0) was applied after thinning at

a rate of 160 kg ha^{-1} . Diallel entries were grown and evaluated in a series of plantings in 2004 on July 7 (M1) and August 30 (M2), and in 2005 on March 16 (M3) and May 15 (M4). The trial planted on July 7 (M1) was excluded in the final analysis because of some missing data.

Climate conditions

All research was conducted at the Waimanalo Research Station (21°20'N latitude, 157°43'W longitude, 18.3 m above sea level), UHM in the island of Oahu, Hawaii from 2003 to 2006. The soil at the Waimanalo station is a Vertic Haplustoll (pH 6.0) derived from coral and lava intrusions. Allocated plots for all the trials in this study had been grown exclusively to corn since 1965 (Brewbaker 1985). Mean monthly temperatures during the growing period from 2003 and 2005 ranged from 22.0 °C in winter to 26.7 °C in summer with mean temperatures slightly higher than usual in February, while succeeding monthly temperature trends followed closely those of the preceding 20 years (Fig. 1).

Average rainfalls were very high from 2003 to 2005 (Fig. 2) and accompanied by much cloud cover and reduced incident light. Average rainfall in 2004 was highest in the months of January (424.2 mm) and February (368.3 mm). The mean amount of rainfall in January (2003–2005) was almost double that of rainfall recorded from 1980 to 2000 (175.3 mm), and reflected in high cloud cover.

Photosynthetic active radiation (PAR) ($mol\ m^{-2}\ day^{-1}$) was measured monthly in 2005 and 2006 throughout the growing period (Fig. 3). Readings were not available during the 2003 and 2004 plantings. Mean PAR across years was unusually higher in the month of February compared to January and later in

Table 1 Maize inbred lines selected for diallel analysis

Inbred	Source Code	Origin	Seed type	Breeder
Hi26	Hi26	CM202 (= CI21E)	Southern dent	M. T. Jenkins
Hi53	ICA L210	Cuban Flint 5832#	Tropical flint	F. Arboleda
Hi57	Ki9	Suwan 1(S)C4 (= KU1409)	Tropical flint	Sujin/Sutat
Hi60	Mo17	CI187-2 × C103	Corn belt dent	M. Zuber
Hi61	N3y	Salisbury White (= SR52F)	Southern dent	H. C. Arnold
Hi62	Pi17	Tropical × Temp	Tropical flint	M. Logroño
Hi65	Tx601	Yellow Tuxpan	Tropical dent	A. Bockholt
Hi67	Tzi18	SeteLagaos TZSR × 7729	Tropical flint	S. K. Kim

Fig. 1 Average temperatures from 2003 to 2005 and for the 20-year period (1980–2000) at Waimanalo Hawaii

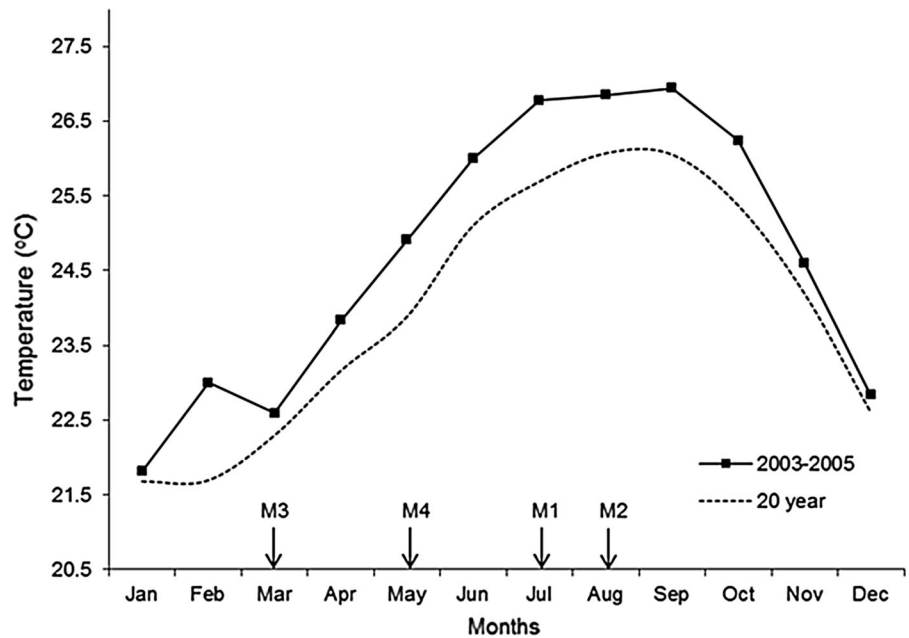
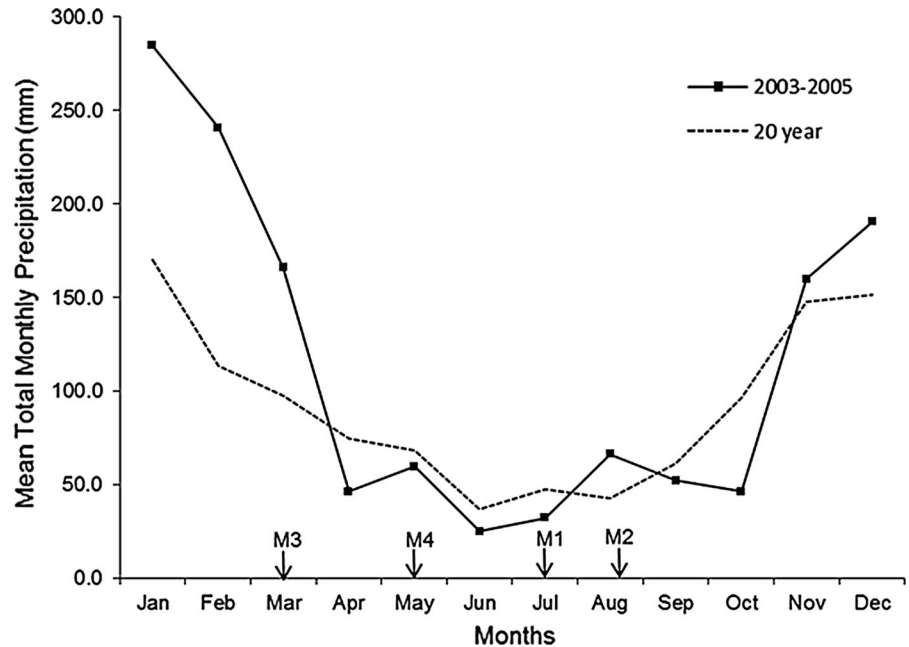


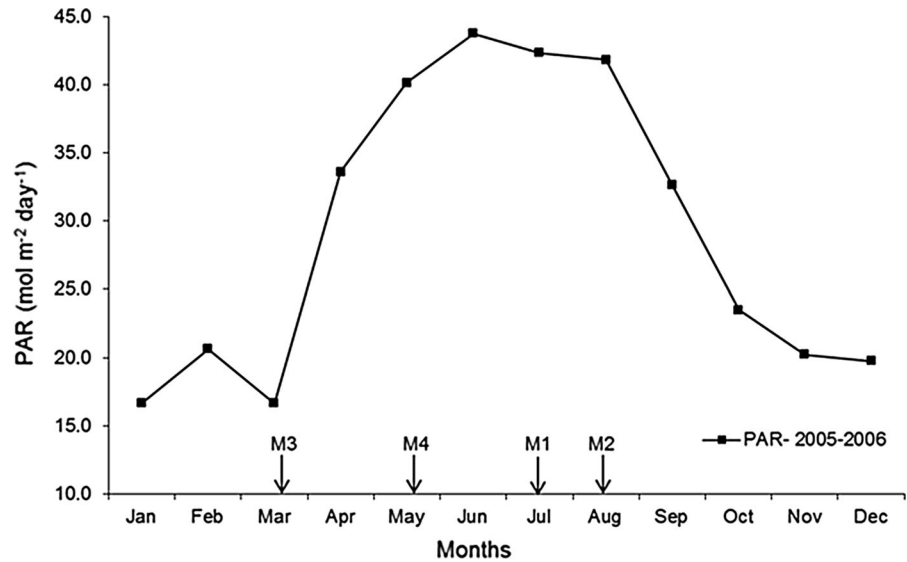
Fig. 2 Average monthly precipitation from 2003 to 2005 and for the 20-year period (1980–2000) at Waimanalo, Hawaii



March. The lowest PAR reading occurred in January ($16.7 \text{ mol m}^{-2} \text{ day}^{-1}$) while the highest readings were observed between June ($43.8 \text{ mol m}^{-2} \text{ day}^{-1}$) and August ($42.7 \text{ mol m}^{-2} \text{ day}^{-1}$), declining as the short-day winter season with heavy rain and cloud cover approached. Jong et al. (1982) found that maize gain yields fluctuated significantly with changes in the

amount of solar radiation in Waimanalo, HI, indicating that light was the most significant environmental factor affecting yields of tropical maize hybrids.

Fig. 3 Average monthly photosynthetic active radiation (PAR) at Waimanalo, Hawaii from 2005 to 2006



Traits measured

Days to mid-silk (DTS) was recorded when 50% of the plants in each plot had emerged silk. The GFR was determined from a random sample of five ears per plot. Harvesting began at 14 days after DTS and continued at 21, 28 and 35 days with a final harvest at maturity (> 40 days post DTS). Harvested ears were dried in a cabinet-type dryer at room temperature for a week. Kernels were shelled from the middle of the ear and dried further at 70 °C in an oven dryer to constant weight. A total of 100 dry kernels were sampled and weighed from each ear. The linear regressions of KW over time were equated to GFR values and expressed as mg kernel⁻¹ day⁻¹.

The GFP was calculated by dividing the physiologically mature KW by GFR (Johnson and Tanner 1972) and expressed as days. The lag periods (LAG) were also estimated for all entries in the three seasons which was determined by dividing the absolute value of the intercept (*a*) of the GFR regression line with its slope (*b*), while KW was expressed as grams per 100 kernels (g 100 k⁻¹). Due to the large error variances associated with determining LAG and their non-significant Analysis of Variance (ANOVA) values, LAG was presumed to have little genetic significance and was excluded in this study. Yields could not be accurately determined for inbreds and hybrids because of the destructive nature of the ear sampling process to

determine GFR and GFP which reduced the total number of ears per plot.

Statistical analysis

To characterize the climate conditions during the growing period, means and standard deviations of temperature (°C) and PAR among the three seasons were calculated exclusively during the GFP for each entry. The Students t test was used to compare the means ($\alpha = 0.05$). Combined ANOVA of the seasons was preceded by a test for the homogeneity of variance using Bartlett's test. The ANOVA was conducted for each trait using the Generalized Linear Model (PROC GLM) procedure of SAS (SAS 1996) and spreadsheet methods of Brewbaker (2004). All F-tests were based on a Fixed Effects Model for entries (inbreds and hybrids). Seasons was considered as a random sample of environments in Hawaii because the average seasonal weather patterns such as temperature and incident light in relation to maize yield in Hawaii are generally predictable all year round (Jong et al. 1982). Replications were considered random. The ANOVA for entries was partitioned into variation among inbreds, among hybrids, and for inbreds vs. hybrids, with similar treatment of the interaction of entries with seasons. Means of entries were adjusted using the Standard Least Squares (LS) method. Diallel analysis among individual seasons and for the combined analysis was conducted using Method 2, Model 1, of

Griffing (1956). Using the LS means, calculations of the ANOVA for GCA, SCA, their individual fixed effects, and standard errors (σ) were performed on spreadsheet according to the procedures outlined by Brewbaker (2004). Mid-parent heterosis (MPH) was calculated by subtracting the mean performance of the parents from their F_1 divided by the mean performance of the parents and expressed as percentage. The relative contributions of GCA and SCA effects controlling a trait were calculated using the direct ratio of GCA to SCA mean squares (MS_{GCA}/MS_{SCA}).

Results

Seasonal variations in PAR and temperature

Values for PAR and temperature during grain filling were significantly different for the three sampled seasons (Table 2) reflecting the changes in the growing environmental conditions among seasons in Waimanalo, HI. Mean temperature and PAR were both minimized in the autumn season (planting in August), and were identical between the spring and summer seasons during grain filling for the diallel set of 8 inbreds and 28 hybrids. For the autumn planting, grain filling was expected to coincide with lower mean temperatures which occurred during the months of October and November.

ANOVA of traits

Data from the three seasons were combined for ANOVA (Table 3) after tests for homogeneity of variances revealed no significant heterogeneity ($\chi^2 < 0.01$). The effects of seasons and inbred vs. hybrid comparisons dominated the significance tests. Most measured traits showed no significant replication effects, a reflection of the high homogeneity of corn research fields in Waimanalo, HI (Brewbaker 1985). The mean squares for inbreds vs. hybrids were significant for all measured traits. The ANOVA for GFR revealed highly significant differences among seasons, inbreds, hybrids and for inbreds vs. hybrids (Table 3). Highly significant differences were also found for seasons and the relevant interactions for GFR values. The extremely low PAR value of the autumn planting during grain filling largely accounted for the seasonal differences in GFR.

The ANOVA of GFP values resembled those of GFR values for all main effects. However, the interactions with season were generally not significant (Table 3). It was concluded that growing conditions prior to silking played a major role in accounting for the absence of clear trends in GFP. Days to mid-silk varied significantly for all tested main effects and interactions. The ANOVA values for KW similarly showed high significance for all main effects and interactions (Table 3).

Mean performances of genotypes (inbreds and hybrids) across seasons for GFR, GFP, DTS and KW

Table 2 Mean seasonal PAR and temperature during the grain filling period among inbreds and hybrids

Planting Month/year	Season	Photosynthetic active radiation ^a	Temperature ^b
Inbreds (N = 8)			
August 2004	Autumn	17.6e ^c	24.2f
March 2005	Spring	42.8a	26.4c
May 2005	Summer	40.9b	26.9a
Grand mean		33.8	25.8
SD		11.7	1.2
Hybrids (N = 28)			
August 2004	Autumn	18.3d	24.3e
March 2005	Spring	42.7a	26.3d
May 2005	Summer	40.4c	26.8b
Grand mean		33.8	25.8
SD		11.1	1.1

^{a,b}Calculated exclusively during the GFP for each inbred and hybrid

^cMeans with a common letter are not significantly different (Students *t*, $\alpha = 0.05$, $t = 1.97149$)

Table 3 ANOVA combined across seasons showing the mean squares for each trait

Source	df	Grain filling rate	Grain filling period	Days to mid-silk	100 kernel weight
Season	2	42.2**	1380.8**	1986.9**	2992.6**
Reps in season	3	1.0	2.5	2.5	17.0**
Entries	35	9.5**	34.5**	55.5**	139.3**
Inbreds	7	9.5**	40.4**	62.4**	102.7**
Hybrids	27	6.1**	24.7*	15.0**	73.1**
Inbreds versus hybrids	1	100.3**	257.1**	1101.4**	2181.8**
Entry × season	70	2.4**	14.1*	3.6**	24.2**
Inbreds × season	14	2.2**	8.1	5.0**	21.8**
Hybrids × season	54	2.5**	14.9*	2.8*	23.4**
(Inbreds vs. Hybrids) × season	2	3.0**	35.8*	14.7**	61.3**
Pooled error	102 ^a	0.6	9.0	1.8	2.4
Total	212				

^aPooled error adjusted to reflect missing data

*, ** Significant at the 5 and 1% levels of probability, respectively

indicated majority of interactions were based on magnitude with some evidence of ranking (Fig. 4). Interactions based on ranking were most evident in some inbreds between the autumn and the favorable growing seasons of spring and summer in Hawaii. For instance, Hi61 a large seeded inbred was superior in performance for GFR across spring and summer seasons, but was outranked by Hi53 in the autumn. Interactions based on magnitude were most pronounced in GFR, GFP, DTS and KW for most hybrid genotypes across the spring and summer seasons. The seasonal differences in PAR and temperature in Waimanalo, HI accounted for the changes observed in genotype performance.

Inbred and hybrid performance

Mean GFR for spring and summer seasons did not differ significantly, both representing similarly high levels of PAR and temperature during grain filling (Table 4). The 28 hybrids consistently had higher GFR ratings than their 8 inbred parents, a 19.6% advantage across all seasons. There was a reduction in mean GFP to a total of approximately 4 weeks in the autumn planting under conditions of the very low PAR values. Mean GFP values for summer period averaged about 5 weeks total and exceeded those of the spring planting date, although PAR values during grain fill

were similar. Mean DTS were much longer in the spring than in the summer trial (Table 4), reflecting the lower mean temperatures of spring in Hawaii. These DTS values evidently affected the GFP and were evident in correlations of PAR, temperature, and GFP values (Josue 2007). Hybrid GFP means also exceeded those of the inbreds by 8.7%, and maximized for the summer planting.

Mean GFR values for Hi61 hybrids were generally outstanding, averaging 11.17 mg kernel⁻¹ day⁻¹ (Table 4). Hybrids with comparably and closely identical high GFR values were Hi53 × Hi61 (11.42 mg kernel⁻¹ day⁻¹) and Hi57 × Hi61 (11.41 mg kernel⁻¹ day⁻¹), both also with high KW. Later maturing inbreds were Hi65, Hi26, and Hi62 based on their higher DTS values. Prolonged DTS was mostly evident in Hi26 hybrids, averaging 59 days. Mean KW (Table 4) across seasons was lowest for Nigerian flint inbred Hi67, and highest for Rhodesian large seeded white dent Hi61. Kernel weights of the 28 hybrids averaged 32.7 g 100 k⁻¹, exceeding the inbreds by 30.8% (Table 4). The large-seeded white dent Hi61 produced hybrids with higher KW, for example in the dent by flint hybrid Hi61 × Hi62 (38.9 g 100 k⁻¹).

Fig. 4 Inbred and hybrid array mean performance across seasons, respectively for **a, b** grain filling rate, **c, d** grain filling period, **e, f** days to mid-silk, and **g, h** 100 kernel weight

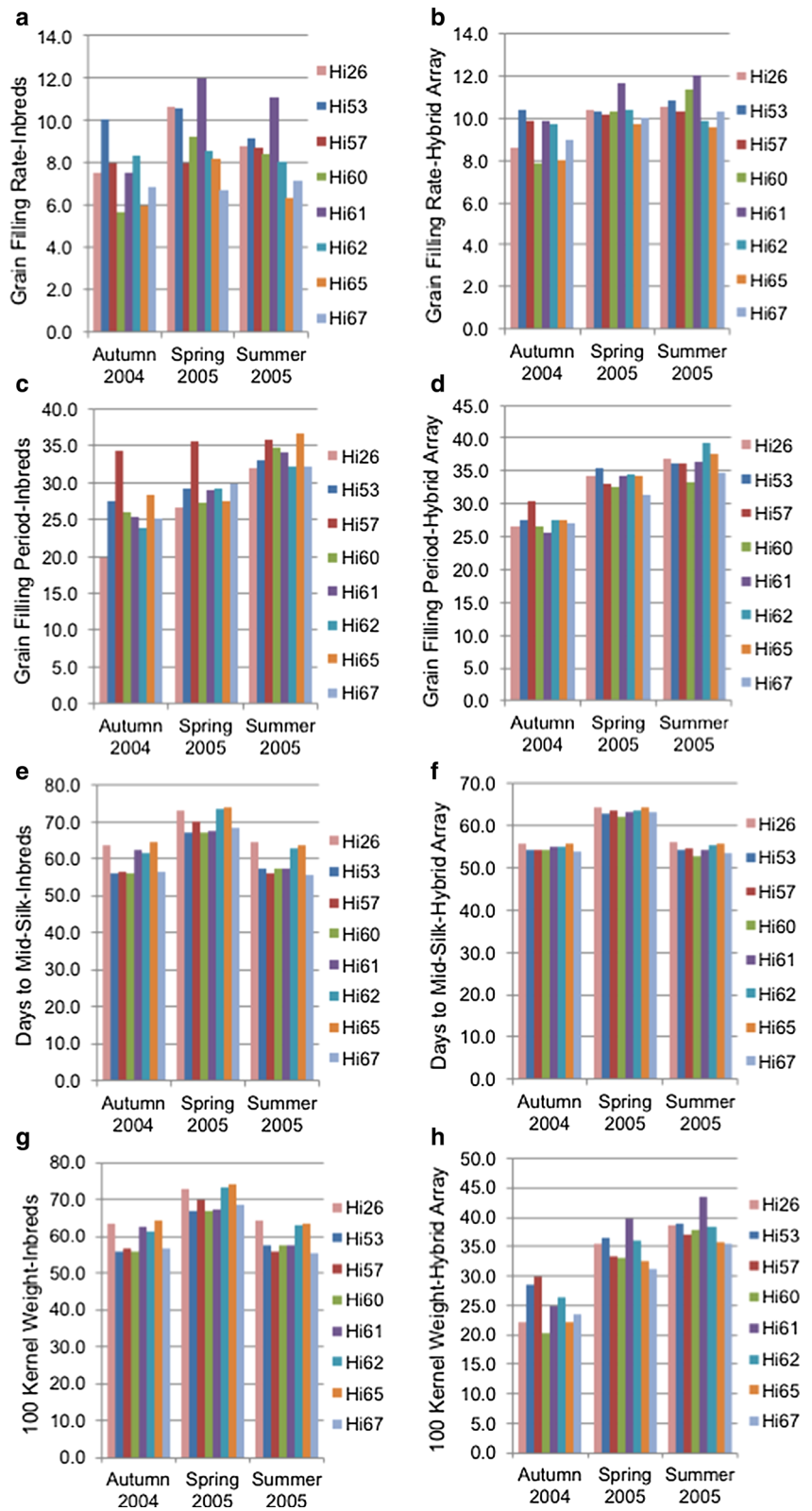


Table 4 Mean trait values for inbreds and diallel hybrids across three seasons

Entries	Grain filling rate	Grain filling period	Days to mid-silk	100 kernel weight
Inbreds (N = 8)				
Hi26	9.00	26.2	67.0	23.7
Hi53	9.96	30.0	60.2	29.8
Hi57	8.22	35.3	60.8	29.0
Hi60	7.79	29.3	60.2	22.8
Hi61	10.21	29.5	62.5	30.5
Hi62	8.32	28.4	66.0	23.6
Hi65	6.86	30.8	67.3	20.8
Hi67	6.92	29.0	60.2	20.1
Min	6.86	26.2	60.2	20.1
Max	10.21	35.3	67.3	30.5
Inbred seasonal means				
Autumn	7.51c ^a	26.3d	59.6c	19.6e
Spring	9.24b	29.3c	70.1a	26.9cd
Summer	8.48bc	33.9b	59.4c	28.6c
Overall inbred means	8.41	29.8	63.0	25.0
LSD (5%)	1.89	7.7	2.8	2.8
Diallel hybrids (N = 28)				
Hi26 × Hi53	10.46	32.6	58.5	34.2
Hi26 × Hi57	11.05	32.4	57.7	35.7
Hi26 × Hi60	8.62	34.4	58.8	29.8
Hi26 × Hi61	10.73	31.0	58.2	34.0
Hi26 × Hi62	9.68	32.8	59.5	31.8
Hi26 × Hi65	8.41	33.6	60.7	28.6
Hi26 × Hi67	9.93	31.2	58.5	30.8
Hi53 × Hi57	10.11	34.9	56.0	35.2
Hi53 × Hi60	11.05	28.5	55.2	31.3
Hi53 × Hi61	11.42	33.4	60.2	38.3
Hi53 × Hi62	10.64	35.0	57.3	36.9
Hi53 × Hi65	9.99	34.5	56.8	34.1
Hi53 × Hi67	10.03	32.7	56.3	32.9
Hi57 × Hi60	10.14	30.3	56.5	30.7
Hi57 × Hi61	11.41	33.9	55.8	38.7
Hi57 × Hi62	9.25	35.3	59.2	32.7
Hi57 × Hi65	8.94	34.7	58.8	30.9
Hi57 × Hi67	9.90	30.8	57.5	30.3
Hi60 × Hi61	11.14	31.9	55.7	36.2
Hi60 × Hi62	10.04	32.8	57.0	33.2
Hi60 × Hi65	8.47	28.8	56.8	25.0
Hi60 × Hi67	9.61	28.9	54.7	28.1
Hi61 × Hi62	12.02	32.5	57.5	38.9
Hi61 × Hi65	10.14	32.3	58.7	33.5
Hi61 × Hi67	11.36	29.5	55.8	33.7
Hi62 × Hi65	9.42	35.6	59.8	33.1
Hi62 × Hi67	9.09	31.9	56.3	29.1

Table 4 continued

Entries	Grain filling rate	Grain filling period	Days to mid-silk	100 kernel weight
Hi65 × Hi67	8.31	32.3	58.7	26.9
Diallel hybrids (N = 28)				
Min	8.31	28.5	54.7	25.0
Max	12.02	35.6	60.7	38.9
Hybrid seasonal means				
Autumn	9.17b	27.3cd	54.8d	24.9d
Spring	10.38a	33.7b	63.4b	34.9b
Summer	10.60a	36.3a	54.6d	38.2a
Overall hybrid means	10.05	32.4	57.6	32.7
Mean MPH (%)	19.7	9.1	− 8.6	30.7
LSD (5%)	1.65	6.4	2.9	3.6
Overall mean across entries	9.68	31.86	58.8	30.96
CV (%)	7.81	9.42	2.3	4.95

^aMeans with a common letter are not significantly different (Students *t*, $\alpha = 0.05$, $t = 1.97149$)

ANOVA for GCA and SCA

Additive gene effects predominated GFR for the combined analysis across the three Waimanalo, Hawaii seasons compared to the other traits (Table 5) revealed by the high ratio of GCA to SCA mean squares (4.83). Magnitudes of GCA and SCA effects were inconsistent across seasons as indicated by the significant GCA and SCA by season interactions.

Additive and non-additive gene effects were not statistically significant for GFP (Table 5). The ratio of GCA to SCA mean squares was 2.15 indicating slightly greater contributions of additive genetic

effects. The importance of additive gene effects was also evident for DTS and KW. Magnitudes of individual GCA effects for GFP, DTS and KW were inconsistent among the seasons as shown by the significant GCA and SCA by season interactions (Table 5).

GCA and SCA effects

The significance of combining ability effects indicates breeding opportunities to alter GFR, GFP, DTS and KW in tropical HFS inbred germplasm. Large seeded Hi61 followed by Hi53 had the greatest potential to

Table 5 ANOVA for GCA and SCA effects showing the mean squares for traits

Source	df	Grain filling rate	Grain filling period	Days to mid-silk	100 kernel weight
GCA	7	9.69	22.56	33.88*	120.76
SCA	28	2.01*	10.52	17.55**	35.11**
GCA × season	14	5.13**	16.14**	8.11**	66.33**
SCA × season	56	0.98**	7.47*	4.53**	9.41**
Error	102 ^a	0.29	4.51	0.29	1.18
Ratio: MS _{GCA} /MS _{SCA}		4.83	2.15	1.93	3.44

^aPooled error adjusted to account for some missing data

*, ** Statistically significant at the 5% and 1% levels, respectively

increase GFR and KW as shown by their outstanding GCA effects compared to other inbreds (Table 6). Inbred Hi65 had the lowest tendency to contribute to increasing GFR due to its low GCA value. Inbred Hi57 could be used to extend GFP as shown by its high and positive GCA effect and, as opposed to Hi60 and Hi67 which could reduce GFP because of their low GCA values (Table 6). Breeding for early maturing hybrids could be achieved by utilizing Hi60 due to its lowest GCA effect for DTS.

Discussion

Information on the performance of tropical inbreds their diallel hybrids, interaction with seasons, determination of their GCA and SCA effects for GFR and GFP in Hawaii are limited. This study uniquely represented variation in germplasm and seasons in Hawaii that occur in maize production. Unlike prior temperate studies (Cross 1975; Ottaviano and Camussi 1981; Katsantonis et al. 1986) where evaluation of genotypes was restricted to similar environmental conditions such as identical durations of light during the summer year after year, this study clearly demonstrated the variation, behavior and genetics of GFR and GFP in tropical germplasm under various seasonal conditions exclusively unique to Hawaii. The inbred parents of the diallel were superior in performance and genetically diverse with both tropical and temperate origins (Brewbaker 1997; Brewbaker and Josue 2007). The seasons during kernel development ranged from superior in Hawaii's summer to severely yield-limiting in low-light autumn. Differences in PAR and temperature during the kernel filling period in this study clearly accounted for some seasonal interactions in GFR, GFP, DTS and KW. These seasonal differences ranged from those favorable to the worst of short day for Hawaii maize production.

Grain filling performance of the inbred and hybrid entries varied significantly, consistent with Fahrner (1991) in prior Hawaii trials. Inbreds Hi61 and Hi53 had the greatest potential to increase GFR based on their high and positive GCA effects compared to other inbreds. Under the growing conditions of this study, Hi60 of temperate origin (Mo17) was among the inbreds that had a low GCA for GFR (-0.19). Studies by Katsantonis et al. (1986) and Ottaviano and Camussi (1981) showed that Mo17 increased GFR

under long day conditions of temperate trials, with summer sunlight conditions estimated to be 50% greater than possible compared to Hawaii. Mid-parent heterosis for GFR was generally positive for all hybrids but varied among the three seasons, leading to significant interactions of seasons with both inbreds and hybrids.

Performance in GFP also varied significantly among diallel entries. Mid-parent heterosis was evident in the reduced filling periods of many hybrids. Heterosis was especially significant for hybrids between tropical inbreds and the ill-adapted temperate inbred Hi60 (Mo17), and maximized for its hybrid with Thailand inbred Hi57. A very low and non-significant variation occurred among replications, reflecting the high homogeneity of soils at the Waimanalo Research Station, Hawaii. Highly significant interactions occurred between genotypes and seasons for GFP as it was for GFR. This was again accounted for by the significant differences in PAR and temperature among autumn, spring and summer trials at Waimanalo, Hawaii.

Diallel analyses of GFR values were interpreted critically from the trials, for which the ratio of GCA to SCA mean squares was 4.83. This predominance of additive gene effects for GFR in the Hawaii germplasm paralleled with temperate maize (Cross 1975; Ottaviano and Camussi 1981; Katsantonis et al. 1986; Wang et al. 1999). Significant seasonal interactions with GCA and SCA effects were evident in this study. These types of interactions however are not unusual in Hawaii. Jong et al. (1982) studied yield variations that doubled from winter to summer over a 41-month study at Waimanalo, Hawaii. The differences in PAR and temperature which coincided with the kernel filling period among the trials in this study clearly accounted for the significance of interactions of GCA and SCA with seasons.

The prevalence of additive gene effects for GFR in this set of HFS inbred germplasm suggests that GFR can be altered by breeding methods that exploit additive gene effects. In Hawaii this may properly focus on maintaining GFR and extending GFP, or increasing GFR and reducing GFP. In northern latitude environments where early frost is a concern, breeding may seek largely to accelerate kernel filling. The SCA effects were also large enough to encourage appropriate hybrid breeding methods. However, the highly significant interactions of season with genotype

Table 6 GCA (bold), SCA for the traits and their standard error estimates

Trait	Hi26	Hi53	Hi57	Hi60	Hi61	Hi62	Hi65	Hi67
Hi26								
Grain filling rate	– 0.02	0.12	1.04	– 0.59	– 0.06	0.05	– 0.20	0.59
Grain filling period	– 0.47	0.69	– 0.35	3.34	0.04	0.73	1.30	0.81
Days to mid-silk	1.25	– 0.83	– 1.65	– 0.16	– 1.55	– 1.43	– 1.03	– 0.65
100 kernel weight	– 0.48	1.13	3.05	0.99	0.09	0.78	0.62	2.37
Hi53								
Grain filling rate		0.48	– 0.17	0.73	– 0.05	0.26	0.48	0.16
Grain filling period		0.36	0.71	– 1.97	1.02	1.56	1.09	1.16
Days to mid-silk		– 0.64	– 1.01	– 1.02	1.84	– 1.17	– 2.02	– 0.38
100 kernel weight		1.78	0.36	– 0.09	1.06	2.37	2.43	1.67
Hi57								
Grain filling rate			0.01	0.53	0.43	– 0.30	0.17	0.53
Grain filling period			1.21	– 1.44	0.54	0.91	0.40	– 1.13
Days to mid-silk			– 0.45	– 0.21	– 1.60	0.02	– 0.71	0.30
100 kernel weight			1.00	0.18	2.09	– 0.04	0.84	0.50
Hi60								
Grain filling rate				– 0.19	0.41	0.49	0.01	0.52
Grain filling period				– 0.94	1.24	1.21	– 1.88	– 0.45
Days to mid-silk				– 1.06	– 1.11	– 1.00	– 1.60	– 1.21
100 kernel weight				– 1.40	2.67	2.77	– 1.17	1.27
Hi61								
Grain filling rate					0.86	0.92	0.21	0.78
Grain filling period					– 0.25	0.27	0.11	– 0.68
Days to mid-silk					– 0.18	– 1.51	– 1.11	– 1.22
100 kernel weight					2.68	2.96	1.07	1.38
Hi62								
Grain filling rate						– 0.03	0.56	– 0.04
Grain filling period						0.46	1.86	0.46
Days to mid-silk						0.71	– 1.12	– 1.73
100 kernel weight						0.30	3.18	0.31
Hi65								
Grain filling rate							– 0.73	0.08
Grain filling period							0.50	0.67
Days to mid-silk							1.19	– 0.46
100 kernel weight							– 1.87	0.82
Hi67								
Grain filling rate								– 0.38
Grain filling period								– 0.86
Days to mid-silk								– 0.83
100 kernel weight								– 2.00
Traits		SE estimates						
		σ_{gi}^a		σ_{sij}^b		$\sigma_{(gi-gj)}^c$		$\sigma_{(sii-sjj)}^d$
Grain filling rate		0.16		0.49		0.14		0.34
Grain filling period		0.63		1.92		0.55		1.34

Table 6 continued

Traits	SE estimates			
	σ_{gi}^a	σ_{sij}^b	$\sigma_{(gi-gj)}^c$	$\sigma_{(sii-sjj)}^d$
Days to mid-silk	0.16	0.49	0.14	0.34
100 kernel weight	0.32	0.98	0.28	0.69

^aStandard error for GCA of each parent

^bStandard error for SCA

^cStandard error difference of GCA between 2 parents

^dStandard error difference of SCA between 2 parents

under low-light stress mandate breeding in multiple planting dates throughout the year. This has been a traditional practice in Hawaii (Brewbaker 2003).

Diallel analyses of GFP values were also interpreted critically, for which the modified ratio of GCA to SCA mean squares was 2.15, indicating larger contribution of additive gene effects. Seasonal interactions were significant for GCA and SCA effects. The inconsistency of genetic effects for GFP over seasons in Hawaii coincided with the findings of prior studies with temperate maize germplasm (Cross 1975; Ottaviano and Camussi 1981; Katsantonis et al. 1986; Wang et al. 1999). The GCA to SCA ratios for GFP in this study was typically much less than for GFR. Breeding to extend GFP in Hawaii using the current germplasm would clearly be challenging but however can be compensated by evaluation of multiple trials throughout the year. The identification of markers and QTLs associated with grain filling traits appear to be promising (Liu et al. 2011), and might be a logical approach to supplement future breeding by marker assisted selection. In Hawaii where climate permits breeding and year-round corn production, focus is on improved yield that might exploit long GFP especially during seasons of short days and low incident light values (Jong et al. 1982). It is evident that breeding for high grain yield in the tropics such as in Hawaii has led, and will continue to lead, to increased grain-fill periods.

Tropical and temperate maize genetic systems obviously affect GFR and GFP. Variation in inbred and hybrid performance was observed for both traits in Hawaii, and inbred values generally served as a good predictor of hybrid performance. Genetic control of GFR largely involved additive gene effects. Grain

filling periods interacted significantly with seasonal variations in light and temperature. Despite interest to extend or attenuate grain filling periods in the short-day tropics, breeding to alter GFP may be challenging. The effects of low-light winters in Hawaii on GFR and GFP mandate that breeding and evaluation occur by shuttle-breeding throughout the year.

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References

- Brewbaker JL (1985) The tropical environment for maize cultivation. In: Brandolini A, Salamini F (eds) Breeding strategies for maize production improvement in the tropics. FAO/UN and Inst. Agron. L'Oltremare, Firenze, pp 47–77
- Brewbaker JL (1997) Registration of 13 tropically adapted parental inbred lines of maize resistant to maize mosaic virus. *Crop Sci* 37:631–632
- Brewbaker JL (2003) Corn production in the tropics: the Hawaii experience. College of Tropical Agriculture and Human Resources, University of Hawaii, Honolulu
- Brewbaker JL (2004) Quantitative genetics on a spreadsheet, 2nd edn. University of Hawaii, Honolulu
- Brewbaker JL, Josue AD (2007) Registration of 27 maize parental inbred lines resistant to maize mosaic virus. *Crop Sci* 47:459–461
- Carter MW, Poneleit CG (1973) Black layer maturity and filling period variation among inbred lines of corn. *Crop Sci* 13:436–439
- Cross HZ (1975) Diallel analysis of duration and rate of grain filling of seven inbred lines of corn. *Crop Sci* 15:532–535
- Daynard TB (1972) Relationships among black layer formation, grain moisture, and heat unit accumulation in corn. *Agron J* 64:716–719
- Daynard TB, Duncan WG (1969) The black layer and grain maturity in corn. *Crop Sci* 9:473–476

- Daynard TB, Kannenberg LW (1976) Relationships between length of the actual and effective grain filling periods and the grain yield of corn. *Can J Plant Sci* 56:237–242
- Fahrner LK (1991) Dry matter accumulation and grain filling period in tropical maize. MS Thesis, University of Hawaii
- Gasura E, Setimela PS, Tarekegne A, Icishahayo D, Edema R, Gibson PT, Okori P (2014) Variability of grain filling traits in early maturing CIMMYT tropical maize inbred lines. *Crop Sci* 54:530–536
- Griffing B (1956) Concept of general and specific combining ability in relation to diallel crossing systems. *Aust J Biol Sci* 9:463–493
- Johnson DR, Tanner JW (1972) Calculation of the rate and duration of grain filling in corn (*Zea mays* L.). *Crop Sci* 12:485–486
- Jones DB, Peterson ML, Geng S (1979) Association between grain filling rate and duration and yield components in rice. *Crop Sci* 19:641–643
- Jong SK, Brewbaker JL, Lee CH (1982) Effects of solar radiation on the performance of maize in 41 successive monthly plantings in Hawaii. *Crop Sci* 22:13–18
- Josue AD (2007) Quantitative genetic analysis of grain filling rate and grain filling period in tropical maize (*Zea mays* L.). Dissertation, University of Hawaii
- Katsantonis N, Gagianas A, Sfakianakis J, Fotiadis N (1986) Inheritance of duration and rate of grain filling and their relationship to grain yield in maize. *Plant Breed* 96:115–121
- Liu ZH, Ji HQ, Cui ZT, Wu X, Duan LJ, Feng XX, Tang JH (2011) QTL detected for grain filling rate in maize using a RIL population. *Mol Breed* 27:25–36
- Maddoni GA, Otegui ME, Bonhomme R (1998) Grain yield components in maize II. Postsilking growth and kernel weight. *Field Crop Res* 56:257–264
- McCann JC (2005) *Maize and grace: Africa's encounter with a new world crop, 1500–2000*. Harvard University Press, Cambridge
- Ottaviano E, Camussi A (1981) Phenotypic and genetic relationships between yield components in maize. *Euphytica* 30:601–609
- Poneleit CG, Egli DB (1979) Kernel growth rate and duration in maize as affected by plant density and genotype. *Crop Sci* 19:385–388
- Reddy VM, Daynard TB (1983) Endosperm characteristics associated with rate of grain filling and kernel size in corn. *Maydica* 28:339–355
- Sala RG, Andrade FH, Westgate ME (2007) Maize kernel moisture at physiological maturity as affected by the source sink relationship during grain filling. *Crop Sci* 47:711–716
- Salvador RJ, Pearce RB (1995) Proposed standard system of nomenclature for maize grain filling events and concepts. *Maydica* 40:141–146
- SAS (1996) SAS Institute Inc. Cary, NC
- Wang G, Kang MS, Moreno O (1999) Genetic analysis of grain filling rate and duration in maize. *Field Crop Res* 61:211–222