

# Kernel Number Determination in Argentinean Maize Hybrids Released between 1965 and 1993

L. Echarte,\* F. H. Andrade, C. R. C. Vega, and M. Tollenaar

## ABSTRACT

Grain yield and the stability of harvest index are greater in newer than in older Argentinean maize (*Zea mays* L.) hybrids. The objective of this study was to elucidate mechanisms underlying the superior yield and harvest index stability of newer Argentinean maize hybrids using the relationship between kernel number per plant ( $KN_p$ ) and plant growth rate during the period bracketing silking (PGR<sub>s</sub>). Three experiments were performed at Balcarce, Argentina, during two growing seasons (1998–2000). Maize was grown under a wide range of plant densities (from 2 up to 30 plants  $m^{-2}$ ) to generate contrasting availability of resources per plant. Growth of individual plants during the period bracketing silking was estimated through a nondestructive method on the basis of relationships between actual shoot dry matter and morphometric variables, including stem and ear diameters and ear length. Detasseling and silk pollination synchronization treatments were imposed in one experiment to also modify available resources per kernel and kernel sink strength. Newer hybrids set more kernels per unit PGR, than older hybrids as is indicated by (i) the lower threshold PGR, for kernel set and (ii) greater potential kernel number at high availability of resources per plant, for newer than for older hybrids. At low and intermediate PGR<sub>s</sub>, the greater kernel set per unit PGR, in newer vs. older hybrids was attributable to greater partitioning of dry matter to the topmost ear during the period bracketing silking, whereas number of kernels set per unit of ear growth rate did not differ. In contrast, kernel set per unit of ear growth rate was greater in newer than in older hybrids when PGR<sub>s</sub> was high. Results of this study indicate that genetic yield improvement in maize is attributable, in part, to increased partitioning of dry matter to the ear during the critical period bracketing silking.

YIELD OF NEWER ARGENTINEAN MAIZE HYBRIDS is greater than that of older ones (Echarte et al., 2000). Genetic yield improvement in North American maize hybrids has been associated with increased dry matter accumulation and not with the proportion of above-ground dry matter that is partitioned to the grain (i.e., harvest index), which has remained relatively stable (Tollenaar and Lee, 2003). Contrarily, harvest index has increased from older to newer Argentinean hybrids (Echarte and Andrade, 2003). More recently released hybrids are more tolerant to high plant-density stress than older hybrids (Russell, 1984; Castleberry et al., 1984; Tollenaar et al., 1992; Duvick, 1997; Tollenaar and Wu, 1999; Echarte et al., 2000; Tollenaar and Lee, 2002). This is associated with higher stability in harvest index of newer hybrids (Echarte and Andrade, 2003), which

may result from their ability to maintain a high kernel number per plant ( $KN_p$ ) as resource availability per plant decreases (Echarte and Andrade, 2003).

Grain yield improvement is highly associated with kernel number (e.g., Andrade et al., 1996; Echarte et al., 2000; Tollenaar et al., 2000). Kernel number per plant is associated with plant growth rate during the critical period bracketing silking (PGR<sub>s</sub>) (Aluko and Fischer, 1988; Tollenaar et al., 1992; Andrade et al., 1999; Vega et al., 2001a). In maize, the  $KN_p$ -PGR<sub>s</sub> relationship has been described by two successive curves to account for the first and second ear in prolific, or a single curve in nonprolific plants (Tollenaar et al., 1992; Andrade et al., 1999; Vega et al., 2001b). A particular feature of the  $KN_p$ -PGR<sub>s</sub> relationship is the significant PGR<sub>s</sub> threshold for kernel set which probably reflects the abrupt decreases in dry matter partitioning to the ear when resources per plant are low (Edmeades and Daynard, 1979; Tollenaar et al., 1992; Andrade et al., 1999; Vega et al., 2001a). The lower threshold of biomass per plant measured at physiological maturity for yield observed in newer hybrids (Echarte and Andrade, 2003) could be associated with a lower PGR<sub>s</sub> threshold for grain set in comparison with older hybrids. On the other hand, the greater  $KN_p$  response to increases in resource availability per plant of newer hybrids (Echarte and Andrade, 2003) could be the result of a greater potential kernel number in the topmost ear. Greater kernel number per plant in newer hybrids was related to a greater kernel set in the topmost ear and not to a greater prolificacy (Echarte and Andrade, 2003).

Differences among hybrids in the number of kernels set per unit of PGR<sub>s</sub> (i.e.,  $KN_p/PGR_s$ ) may be attributable to either or both dry matter partitioning to the ear and number of kernels set per unit of ear growth rate during the period bracketing silking (Andrade et al., 1999; Vega et al., 2001b). First, increased partitioning to the ear could increase  $KN_p/PGR_s$ . Genetic reduction in tassel size or tassel removal have generally favored dry matter partitioning to the ear and increased kernel set (Fischer and Palmer, 1984; Bolaños and Edmeades, 1993; Edmeades et al., 1993). Tassel size has declined linearly in U.S. hybrids from the 1930s to the 1990s (Tollenaar et al., 2000). A lower response of kernel number to tassel removal would be anticipated in newer than in older hybrids if the reduction in tassel size has been associated with a concomitant reduction in tassel dominance over the ear. Second, an increase in the number of kernels set per unit of ear growth rate during the critical period bracketing silking (EGR<sub>s</sub>) could in-

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**Abbreviations:** EGR<sub>s</sub>, growth rate of the topmost ear during the critical period bracketing silking;  $KN_p$ , kernel number per plant;  $KN_1$ , kernel number of the topmost ear; PGR<sub>s</sub>, plant growth rate during the critical period bracketing silking; EGR<sub>s</sub>.

**Table 1. Year of hybrid release, hybrid type and endosperm type, and relative maturity.**

Hybrid	Year of release	Hybrid type and endosperm type	Relative maturity†
DKF880	1965	Double cross, flint	120
M400	1978	Double cross, flint	128
DK4F36	1982	Double cross, flint	127
DK4F37	1985	Double cross, flint	124
DK664	1993	Single cross, semi-dent	116
DK752	1993	Single cross, semi-dent	125

† Relative maturity 120 = FAO 600; Relative maturity 130 = FAO 700.

crease  $KN_p/PGR_s$ . Differences in the assimilate requirement per kernel (Edmeades and Daynard, 1979; Edmeades et al., 1993; Vega et al., 2001a) may account for differences in number of kernels set per unit of  $EGR_s$  among hybrids. In addition, differences in kernel number per unit  $EGR_s$  may be associated with differences in synchronization of fertilization of florets within the ear (Cárcova et al., 2000; Cárcova and Otegui, 2001). Therefore, if kernel number set per unit of ear growth is a mechanism underlying a greater  $KN_p/PGR_s$  of newer hybrids, (i) a lower minimum assimilate requirement per kernel and/or (ii) a lower kernel set response to improved synchronism in silk pollination would be expected in newer than in older hybrids.

In this work, we examine the mechanisms that underlie the differences in kernel number per unit of  $PGR_s$  between newer and older Argentinean maize hybrids. The  $PGR_s$  threshold for kernel set and the response of  $KN_p$  to  $PGR_s$  increments could be involved in the high kernel number and HI stability of new Argentinean maize hybrids. We tested the hypothesis that the  $PGR_s$  threshold for grain set is lower and the kernel response to  $PGR_s$  increments is greater in newer than in older hybrids by examining the response of  $KN_p$  to resource availability of individual plants rather than that of plot means. In addition, we examined whether dry matter partitioning to the ear and kernel number set per unit of ear growth rate during the critical period bracketing silking is associated with the greater kernel number per unit of  $PGR_s$  of a newer versus an older maize hybrid.

## MATERIALS AND METHODS

### Site and Crop Management

Maize was grown at Balcarce, Argentina (37°45' S, 58°18' W; elevation 130 m), during the 1998–1999 (Exp. 1) and the 1999–2000 growing seasons (Exp. 2 and 3). Crops were fertilized with 35 kg P ha<sup>-1</sup> before sowing, and with 150 kg N ha<sup>-1</sup> at V6 (Ritchie and Hanway, 1982). Soil water to 1-m depth was kept over 50% of maximum available water by sprinkler irrigation. Weeds and insects were effectively controlled.

### Plant Material and Experimental Design

The maize hybrids Morgan 400, DeKalb 4F36, DeKalb 664, and DeKalb 752 (Exp. 1) and DeKalb F880 and DeKalb 752 (Exp. 2 and 3) were sown on 6 Oct. 1998 (Exp. 1) and 8 Oct. 1999 (Exp. 2 and 3). Each of these hybrids was among the three topmost cultivated hybrids in the Argentinean Pampas for at least 5 yr after their release (Table 1). In all three experiments, plant density was used as the source of experimental variation for  $KN_p$  and  $PGR_s$ . Plots were oversown and

thinned to the desired plant densities at V3. The experimental design was a split-plot randomized complete-block design with three replications, with plant densities as main plots and hybrids as subplot. Plant densities at harvest were 2, 4, 8, 16, and 30 plants m<sup>-2</sup> in Exp. 1 and 2, and 8 and 16 plants m<sup>-2</sup> in Exp. 3. Subplots comprised five 7-m-long rows at low plant densities and six to seven 7-m rows at intermediate (8 plants m<sup>-2</sup>) and higher plant densities (16–30 plants m<sup>-2</sup>). The distance between rows was 0.7 m in all cases. In Exp. 3, treatments of detasseling and artificial synchronous pollination were applied at random within each experimental unit. Detasseling was performed by hand ( $n \cong 80$  plants per hybrid) when the tassel was still surrounded by four leaves (i.e.,  $6.5 \pm 0.14$  d before silking, average for both hybrids and plant densities). In plants chosen for artificial synchronous pollination ( $n \cong 80$  plants per hybrid), both the uppermost and the second ears were bagged before silk emergence. Both ears were pollinated 5 d after first silks emerged from the husks of the uppermost ear. In border rows, additional plants were sown 2 wk after 8 Oct. 2000 to assure adequate pollen availability during the whole period.

### Measurements

Shoot biomass of tagged plants was quantified at approximately 10 d before and 15 d after silking (henceforth this period is referred to as the critical period bracketing silking) through a combination of destructive and nondestructive sampling following methodologies described by Vega et al. (2001a, 2001b) (see below). At maturity,  $KN_p$  was determined in the topmost ( $KN_1$ ) and second ear. In Exp. 1 and 2, anthesis and silking dates were recorded for each experimental unit as the dates when 50% of the plants presented visible anthers on the main branch and at least one emerged silk from the husks, respectively. In Exp. 3, silking and anthesis dates were recorded for each individual plant ( $n \cong 240$  plants per hybrid).

### Destructive Sampling

Morphometric variables, i.e., basal stem diameter and diameter and length of the topmost ear, were measured on a density-dependent number of plants (three plants per replicate at low plant density and six to eight plants per replicate at high plant densities). Diameter of the stem and the ear were measured on the widest section. Immediately after measurements, plants were harvested, leaving borders of at least 1 m between successive harvests. Plants were separated into leaf blade, stem plus sheath, and ears and oven dried at 65°C until constant weight. Allometric relationships were established between morphometric variables and dry weights of shoot and female reproductive structures. Reproductive structures included kernels and rachis of the topmost ear. Models fitted to shoot dry weight are summarized in Table 2 and models fitted to dry weight of the topmost ear are summarized in Table 3.

### Nondestructive Sampling

Before silking, a density-dependent number of consecutive plants were tagged within each subplot, i.e., six plants at the lowest plant density and up to 30 plants at the highest plant density. Shoot and reproductive biomass were assessed for each tagged plant using the allometric relationships shown in Tables 2 and 3. In all cases, sample areas were bordered by at least three (low plant densities) or four (intermediate and higher plant densities) guard rows; and by at least 1 m within the row. The radiation profile along the stem was measured 2 wk after flowering for control and detasseled plants in Exp.

**Table 2. Relationships between shoot biomass and morphometric variables at the beginning ( $S_0$ ) and at the end ( $S_1$ ) of the critical period for kernel set (sd = stem diameter; ed = uppermost ear diameter; el = uppermost ear length). All models were significant at  $P < 0.05$ .**

Exp.	Hybrids	Shoot biomass g plant <sup>-1</sup>	R <sup>2</sup>
1	M400	$S_0 = -58.1 + 53.6 \times sd$	0.83
		$S_1 = -5.0 + 16.4 \times sd^{2.0} + 2.4 \times (ed \times el)^{0.6}$	0.93
	DK4F36	$S_0 = -45.7 + 45.2 \times sd$	0.89
		$S_1 = -0.4 + 21.4 \times sd^{1.8} + 0.1 \times (ed \times el)^{1.3}$	0.94
	DK664	$S_0 = 0.3 + 9.0 \times sd^{1.8}$	0.91
		$S_1 = 8.7 + 8.0 \times sd^{2.4} + 0.7 \times (ed \times el)^{0.9}$	0.98
2	DK752	$S_0 = -31.0 + 27.8 \times sd^{1.3}$	0.92
		$S_1 = -9.2 + 23.3 \times sd^{1.5} + 0.2 \times (ed \times el)^{1.3}$	0.98
	DKF880	$S_0 = -52.8 + 52.6 \times sd$	0.83
		$S_1 = [2.7 + 0.007 \times (sd \times ed)^2 + 0.2 \times el + 1.8 \times sd]^2$	0.96
3	DK752	$S_0 = -54.3 + 46.4 \times sd$	0.89
		$S_1 = (3.34 + 0.6 \times sd \times ed + 0.003 \times el)^2$	0.95
	DKF880	$S_0 = -52.8 + 52.6 \times sd$	0.83
		$S_1 = (2.0 + 1.95 \times sd + 0.47 \times ed + 0.15 \times el)^2$	0.81
	DK752	$S_0 = -54.3 + 46.4 \times sd$	0.89
		$S_1 = 9.0 - 28.8 \times ed + 3.6 \times (sd \times el) + 17 \times (sd \times ed) - 0.89 \times (el \times sd)^2$	0.90

**Table 3. Relationships between dry matter of the uppermost ear (E) and morphometric variables at the end of the critical period for kernel set (ed = uppermost ear diameter; el = uppermost ear length). All models were significant at  $P < 0.05$ .**

Exp.	Hybrids	Ear dry matter at the end of critical period G ear <sup>-1</sup>	R <sup>2</sup>
2	DKF880	$E = (-0.13 + 0.41 \times ed + 0.02 \times el \times ed)^2$	0.94
	DK752	$E = (-0.91 + 0.82 \times ed + 0.001 \times ed \times el)^2$	0.96
3	DKF880	$E = (-0.35 + 0.55 \times ed + 0.07 \times el)^2$	0.82
	DK752	$E = [-0.24 + 0.06 \times (ed \times el) - 0.0001 \times (ed \times el)^2]^2$	0.85

3 with a quantum sensor (model LI-190SA, LI-COR, Lincoln, NE).

**Data Analysis**

Growth rate during the critical period for kernel set was estimated as the ratio between accumulated biomass in shoots or topmost ear and the duration of the period. We assumed a linear relationship between biomass accumulation per plant and days during the period bracketing silking, and female reproductive biomass to be negligible at 10 d before silking (beginning of the critical period bracketing silking).

The relationship between  $KN_1$  and  $PGR_s$  was investigated using a nonlinear model (Model 1; Jandel Scientific, 1991). This model was chosen because it includes parameters with biologically meaning parameters (Echarte and Andrade, 2003).

$$KN_1 = a_1 \times \{1 - \exp[-(PGR_s - x_0)/b_1]\} \text{ if } PGR_s \geq x_0 \quad [\text{Model 1}]$$

$$KN_1 = 0 \text{ if } PGR_s < x_0$$

Parameter  $a_1$  quantifies the potential number of kernels set in the topmost ear, and  $b_1$  is a measure of the curvilinearity of the  $KN_1$ - $PGR_s$  relationship. A large  $b_1$  value indicates that the curve approaches a straight line. The parameter  $x_0$  (g plant<sup>-1</sup> d<sup>-1</sup>) represents the  $PGR_s$  threshold for kernel set in the uppermost ear. Free iteration of parameters yielded large errors in the estimation of the  $PGR_s$  threshold for kernel set (parameter  $x_0$ ). Particularly, the model did not adequately estimate  $KN_1$  for plants with  $KN_1 < 200$  kernels. Most of the residuals of the model were negative for these plants, especially for older hybrids. This was mainly explained by a large variability in  $KN_1$  close to the threshold and to the high initial slope of the  $KN_1$ - $PGR_s$  relationship. Therefore, parameter  $x_0$  was set in the model as an input based on  $PGR_s$  data from nonbarren plants bearing nubbins. The threshold of  $PGR_s$  for kernel set was estimated as the average  $PGR_s$  of plants that set 1 to 100 kernels. Selection of a narrow range of  $KN_1$ , i.e.,  $1 > KN_1 >$

50 did not provide enough number of plants to perform valid statistic comparisons among hybrids. Barren plants were not included in the calculation of the  $PGR_s$  threshold because they underestimate its value.

Additionally, we fitted the relationship between  $KN_1$  and the ear growth rate at the period bracketing silking ( $EGR_s$ ) using Model 2 (Vega et al., 2001a); which presented a greater  $R^2$  than Model 1.

$$KN_1 = [a_2 \times (EGR_s - x_t)]/[1 + b_2 \times (EGR_s - x_t)] \text{ if } EGR_s \geq x_t \quad [\text{Model 2}]$$

$$KN_1 = 0 \text{ if } EGR_s < x_t$$

Parameters  $a_2$  and  $b_2$  represent the initial slope and the curvilinearity of the  $KN_1$ - $EGR_s$  relationship, respectively. Low  $b_2$  indicates that the curve approaches to a straight line. Parameter  $x_t$  quantifies the  $EGR_s$  threshold to set kernels.

The minimum assimilate requirement per kernel (mg kernel<sup>-1</sup> d<sup>-1</sup>) was estimated as the mean  $EGR_s KN_1^{-1}$  for the interval of  $EGR_s$  at maximum kernel set per unit of  $EGR_s \pm 10\%$ . The maximum kernel set per unit of  $EGR_s$  was obtained from the hyperbolic  $KN_1$ - $EGR_s$  relationship (Vega et al., 2001a).

Data were processed by  $t$  test of parameters and  $t$  tests were used to assess differences between hybrids in  $KN_p$  and  $EGR_s$  for intervals of  $EGR_s$  or  $PGR_s$ .

**RESULTS AND DISCUSSION**

**Relationship between  $KN_p$  and  $PGR_s$**

The relationship between  $KN_p$  and  $PGR_s$  was curvilinear (Fig. 1) with a  $PGR_s$  threshold for kernel set ( $x_0$ ). A trend toward a plateau for kernel number of the topmost ear ( $KN_1$ ) at high values of  $PGR_s$  indicates morphogenetic limitations in reproductive plasticity. There was not a clear trend in mean  $PGR_s$  at each plant density with year of hybrid release (data not shown).

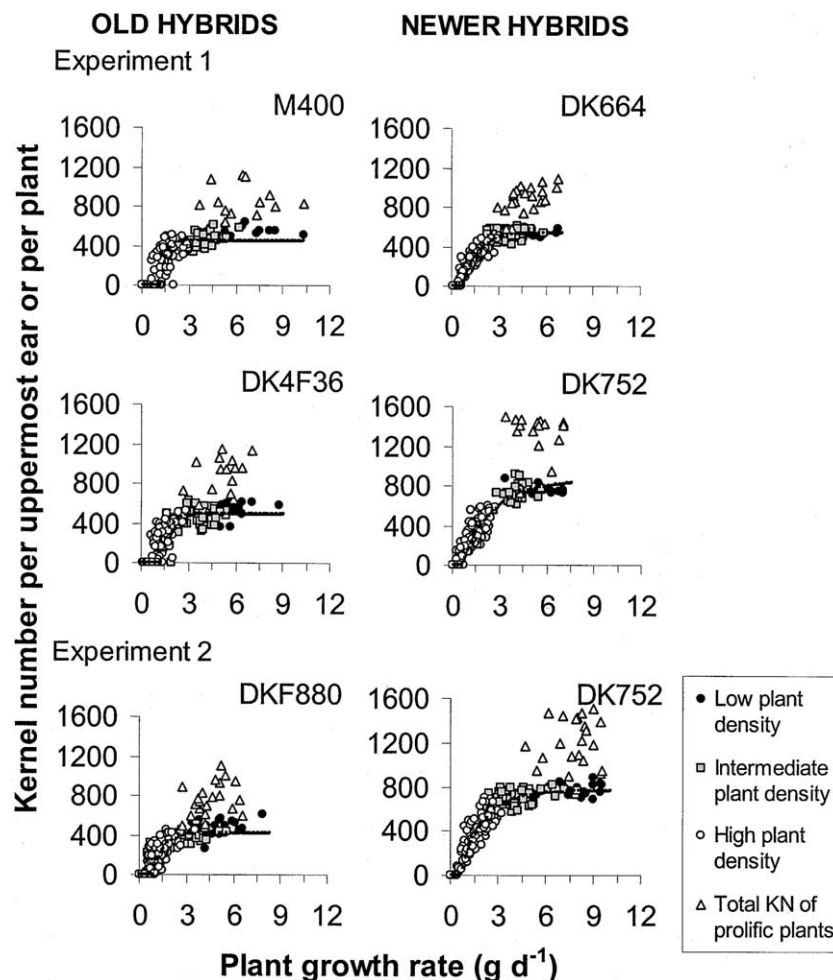


Fig. 1. Relationship between kernel number per uppermost ear ( $KN_1$ ) or per plant ( $KN_p$ ) and plant growth rate during a period bracketing silking ( $PGR_s$ ) in five maize hybrids released in Argentina in different decades. Curves are the fitted Model 1. Triangles represent kernel number of prolific plants (kernel number of the topmost and second ear). Other symbols represent  $KN_1$  at low (2–4 plants  $m^{-2}$ ; solid circles); intermediate (8 plants  $m^{-2}$ ; squares), and high plant densities (16–30 plants  $m^{-2}$ ; white circles).

At fixed ranges of  $PGR_s$ ,  $KN_1$  was greater in newer than in older hybrids ( $P < 0.05$ ). For example, the newer hybrid DK752 set 26 and 51% more kernels than the older hybrid M400 at low (i.e.,  $0.5 < PGR_s < 1.5 \text{ g d}^{-1}$ ) and at high (i.e.,  $5 < PGR_s < 6 \text{ g d}^{-1}$ ) resource availability per plant, respectively. This supports contentions of a previous analysis based on mean plot data which concluded that greater  $KN_p$  set per unit of  $PGR_s$  would primarily underlie the greater  $KN_p$  of newer hybrids (Echarte et al., 2000). The threshold  $PGR_s$  ( $x_0$ ) was higher for older hybrids ( $0.82 \text{ g plant}^{-1} \text{ d}^{-1} > x_0 > 1.18 \text{ g plant}^{-1} \text{ d}^{-1}$ ) than for newer hybrids ( $0.52 \text{ g plant}^{-1} \text{ d}^{-1} > x_0 > 0.65 \text{ g plant}^{-1} \text{ d}^{-1}$ ) (Table 4). In accordance to these values of  $PGR_s$  thresholds for kernel set, more than half of the plants of the old hybrids and only 16% of the plants of the newer hybrids were sterile at  $PGR_s$  from 0.5 to  $1 \text{ g d}^{-1}$  (Fig. 2a). Those proportions decreased to 17 and 0% for older and newer hybrids at  $PGR_s$  from 1 to  $1.5 \text{ g d}^{-1}$  (Fig. 2b). In addition, supporting a lower threshold  $PGR_s$  for kernel set in newer than in older hybrids, the  $PGR_s$  values below which half of the plants were sterile were lower in newer ( $0.82$  and  $0.91 \text{ g d}^{-1}$  for DK664 and DK752, respectively) than in

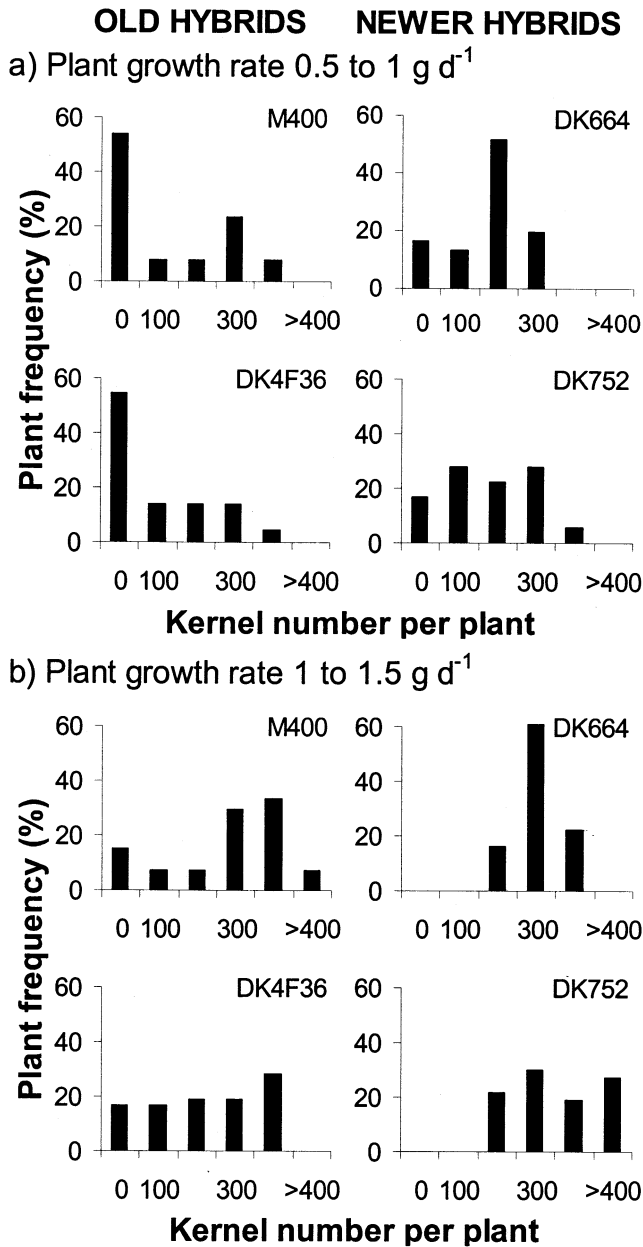
older hybrids ( $1.58$  and  $1.43 \text{ g d}^{-1}$  for M400 and DK4F36, respectively). The degree of curvilinearity of the  $KN_1$ – $PGR_s$  relationship was lower in newer than in older hybrids (parameter  $b_1$ , Table 4—a large  $b_1$  value indicates that the curve approaches a straight line). Consequently,  $KN_1$  continues to increase to greater  $PGR_s$  in newer than in older hybrids (Fig. 1). A comparison of the newer hybrid DK752 with the older hybrid DKF880 showed that both number of kernel rows (20 vs. 14) and number of kernels per row (37 vs. 28) were greater in the newer hybrid at intermediate plant density (i.e., 8 plants  $m^{-2}$ ).

Results showing that the  $PGR_s$  threshold for kernel set and the initial slope of the  $KN_1$ – $PGR_s$  relationship were lower for newer than for older hybrids (Fig. 1) do not agree with results of previous reports (Tollenaar et al., 1992; Maddonni et al., 2000; Luque, 2000). We believe that the difference is attributable to the employed method that was based on analysis of whole-plot means in the previous reports. In contrast, a wide range of values for  $PGR_s$  and  $KN_p$  were obtained in our experimental approach that used data for individual plants rather than plot means, allowing for a more precise

**Table 4.** Threshold of  $PGR_s$  for kernel set ( $x_0$ ) estimated as the mean  $PGR_s$  of plants that set 1 to 100 kernels, potential kernel number ( $a_1$ ) and degree of curvilinearity ( $b_1$ ), and  $R^2$  of Model 1 ( $KN_1 = a_1 \times \{1 - \exp[-(PGR_s - x_0)/b_1]\}$  if  $PGR_s \geq x_0$  and  $KN_1 = 0$  if  $PGR_s < x_0$ ) fitted to the  $KN_1$ - $PGR_s$  relationship of five Argentinean maize hybrids.

Exp.	Hybrid	Year of release	$x_0$	$a_1$	$b_1$	$R^2$
			$g\ plant^{-1}\ d^{-1}$	kernel no.		
1	M400	1978	1.18 ab†	448 d	0.31 d†	0.63
	DK4F36	1982	1.17 a	493 c	0.48 c	0.67
	DK664	1993	0.65 bc	541 b	0.97 b	0.87
	DK752	1993	0.52 c	852 a	2.00 a	0.87
2	DKF880	1965	0.82 a	420 b	0.67 b	0.64
	DK752	1993	0.58 b	769 a	1.45 a	0.90

† Means within a column and within an experiment followed by the same letter are not significantly different at  $P < 0.05$ .



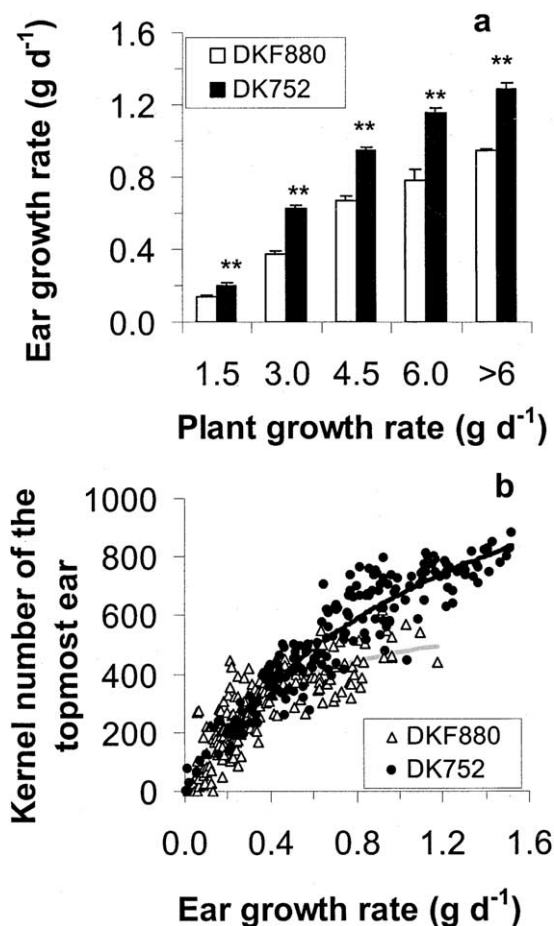
**Fig. 2.** Frequency distributions (%) of kernel number per plant for plants growing at (a)  $0.5 > PGR_s > 1\ g\ d^{-1}$  and (b) at  $1 > PGR_s > 1.5\ g\ d^{-1}$  in four maize hybrids released in Argentina at different decades (data from Exp. 1).

estimate of  $x_0$ . There was a significant positive association between  $x_0$  and barrenness at high plant density ( $r = 0.94$ ,  $P < 0.05$ ), and a negative correlation between  $x_0$  and the number of kernels set per unit of  $PGR_s$  at low  $PGR_s$  ( $r = 0.95$ ,  $P < 0.05$ ). The threshold of  $PGR_s$  for kernel set strongly correlated with the threshold in terms of biomass per plant measured at physiological maturity reported by Echarte and Andrade (2003) ( $r = 0.87$ ,  $P < 0.05$ ). This supports the contention that the improved tolerance of newer hybrids to high plant density is attributable, in part, to a lower  $PGR_s$  threshold for kernel set. High  $x_0$  in maize probably relates to rather abrupt decreases in dry matter partitioning to the ear when resources per plant diminish during the critical period for kernel set (Edmeades and Daynard, 1979; Tollenaar et al., 1992; Andrade et al., 1999; Vega et al., 2001a). Such a high susceptibility of the female reproductive structure to low resource availability per plant may reflect the dominance of the tassel (Fischer and Palmer, 1984; Doebley et al., 1997).

The lower  $x_0$  of newer hybrids, which is associated with an improved performance under stress or high plant density, could result from indirect selection under progressively higher plant densities in breeding programs and from hybrid evaluation across a wide range of environments, including low-yield environments (Troyer, 1996; Reeder, 1997; Tollenaar and Lee, 2002; Fasoula and Fasoula, 2002). Similarly, genotype selection for tolerance to mid-season drought stress may lead to changes that also contribute to increased tolerance to nitrogen and high plant density stress (Bänziger et al., 2002). Andrade et al. (2002) showed that the  $KN_1$ - $PGR_s$  relationship is not influenced by the nature of the cause for variation in  $PGR_s$  (e.g., plant density, nitrogen, water). Therefore, it is likely that kernel set in older and newer hybrids will follow the specific  $KN_1$ - $PGR_s$  relationships irrespective of the nature of the environmental stress. Greater dry matter partitioning to the ear at the period bracketing silking, and/or greater kernel number set per unit of ear growth rate (Andrade et al., 2000; Vega et al., 2001a), could underlie the decrease in  $x_0$  and the increase in kernel set per unit  $PGR_s$  with year of release.

#### Partitioning of Dry Matter to the Topmost Ear and Kernel Set per Unit EGR<sub>s</sub>

Growth rate of the topmost ear during the period bracketing silking was greater in the newer hybrid



**Fig. 3.** (a) Mean growth rate of the topmost ear ( $EGR_s$ ) vs. 1.5-g  $d^{-1}$  intervals of plant growth rate during a period bracketing silking ( $PGR_s$ ) and (b) relationship between kernel number of the topmost ear and ear growth rate during a period bracketing silking of an older (DKF880) and a newer hybrid (DK752) in Exp. 2. In Fig. 3a values at the x axis indicate the upper limit of the interval. Bars indicate standard error. Number of individuals ranged from 10 to 100 depending on the range of  $PGR_s$ . \*\* indicates significant differences between hybrids within the range of  $PGR_s$  at  $P < 0.05$ . In Fig. 3b, curves are the fitted Model 2 for DKF880 (gray line) and DK752 (black line). Fitted regression were: DKF880,  $KN_1 = 1807 \pm 189.9 \times (EGR_s - 0.028 \pm 0.01) / [1 + 2.77 \pm 0.464 \times (EGR_s - 0.028 \pm 0.01)]$  if  $EGR_s \geq x_i$  and  $KN_1 = 0$  if  $EGR_s < x_i$ ,  $R^2 = 0.78$ ; DK752,  $KN_1 = 1191 \pm 89.5 \times (EGR_s - 0.007 \pm 0.019) / [1 + 0.76 \pm 0.12 \times (EGR_s - 0.007 \pm 0.019)]$ ,  $x_i$  and  $KN_1 = 0$  if  $EGR_s < x_i$ ,  $R^2 = 0.89$ . The regressions were significant at  $P < 0.05$ .

DK752 than in the older hybrid DKF880, across the whole range of  $PGR_s$  ( $P < 0.05$ , Fig. 3a). In addition, mean  $PGR_s$  at each plant density was greater in the newer hybrid DK752 than in the older hybrid (data not shown). As a consequence, the proportion of plants with very low  $EGR_s$  (i.e.,  $EGR_s < 0.2$  g  $d^{-1}$ ) was greater in the older hybrid (55%) than in the newer hybrid (23%) at high plant densities (16–30 plants  $m^{-2}$ ). When expressed at equal  $EGR_s$ , however,  $KN_1$  did not differ between the two hybrids at low and intermediate  $EGR_s$  (i.e.,  $EGR_s$  from 0 to 0.5 g  $d^{-1}$ , Fig. 3b). Therefore, these results support the contention that the greater number of kernels set per unit of  $PGR_s$  of the newer hybrid was not influenced by a greater kernel number set per unit

of ear growth rate at relatively low resource availability per plant. In contrast, the number of kernels set per unit of ear growth rate was greater in the newer hybrid ( $P < 0.05$ , Fig. 3b) at low plant densities (i.e.,  $EGR_s \geq 0.6$  g  $d^{-1}$ ) because of a larger reproductive plasticity, i.e., potential kernel number per ear (Table 4). In addition, at high resource availability per plant, Vega et al. (2001a) showed that  $EGR_s$  of the topmost ear is lower in prolific plants compared with nonprolific plants of the same hybrid. This suggests that a low mean  $EGR_s$  of the topmost ear at high resource availability per plant could be associated, in part, with a high prolificacy (i.e., large proportion of prolific plants). However, since prolificacy did not present a clear trend with the year of hybrid release (Echarte et al., 2000; Echarte and Andrade, 2003) and since at intermediate and low plant densities the oldest and the newer hybrids presented the same proportion of prolific plants (27.5 and 28.3% for DKF880 and DK752, respectively), a lower growth rate of the topmost ear in the older hybrid is not associated with a greater prolificacy.

Tassel removal, performed to diminish the dominance of the tassel over the ear, increased  $KN_1$  in the older hybrid only when  $PGR_s$  ranged from 1.5 to 3 g  $plant^{-1} d^{-1}$  ( $p < 0.05$ ; Fig. 4a). Out of this  $PGR_s$  range,  $KN_1$  increases in the older hybrid in relation to the control were less significant ( $p > 0.5$ ). Synchronization of pollination, performed to diminish competition for assimilates among kernels within the ear, did not affect  $KN_1$  ( $p > 0.05$ , data not shown). In Exp. 3,  $PGR_s$  varied between 0 and 4 g  $plant^{-1} d^{-1}$  and kernel number per plant varied between 0 and 680 for DKF880 and between 0 and 850 for DK752. Within each hybrid, mean  $PGR_s$  did not differ among treatments, i.e.,  $PGR_s$  was  $1.55 \pm 0.09$ ,  $1.56 \pm 0.13$  and  $1.41 \pm 0.09$  g  $plant^{-1} d^{-1}$  for DKF880 and  $2.14 \pm 0.09$ ,  $2.28 \pm 0.09$  and  $2.17 \pm 0.11$  g  $plant^{-1} d^{-1}$  for DK752, for detasseled, synchronous pollination and control treatments, respectively. Tassel removal resulted in greater  $EGR$  at  $PGR$ s lower than 3 g  $plant^{-1} d^{-1}$  in DKF880 and did not have any effect on DK752. For the old hybrid, the increase in the growth rate of the topmost ear associated with tassel removal for  $PGR_s$  between 1.5 to 3 g  $plant^{-1} d^{-1}$  (Fig. 4b) was associated with an increase in  $KN_1$  per unit of plant growth rate (Fig. 4a). There were no differences ( $P > 0.05$ ) in the radiation profile of detasseled and control plants in both hybrids (data not shown). The positive effect of tassel removal on kernel number in the older hybrid and the lack of response in the newer hybrid may be associated with differences in tassel size between the two hybrids. Although tassel size was not measured in this study, this trait has been shown to decline from older to newer U.S. hybrids (e.g., Tollenaar et al., 2000).

Improved synchronization in floret fertilization can reduce competition among kernels within the ear and reduced competition may result in an increase in kernel set (Cárcova et al., 2000; Cárcova and Otegui, 2001). Increased synchronization of fertilization, however, did not improve kernel set per unit  $EGR_s$  in either hybrid ( $P > 0.05$ , data not shown). In addition, the minimum assimilate requirement per kernel (Edmeades and Day-

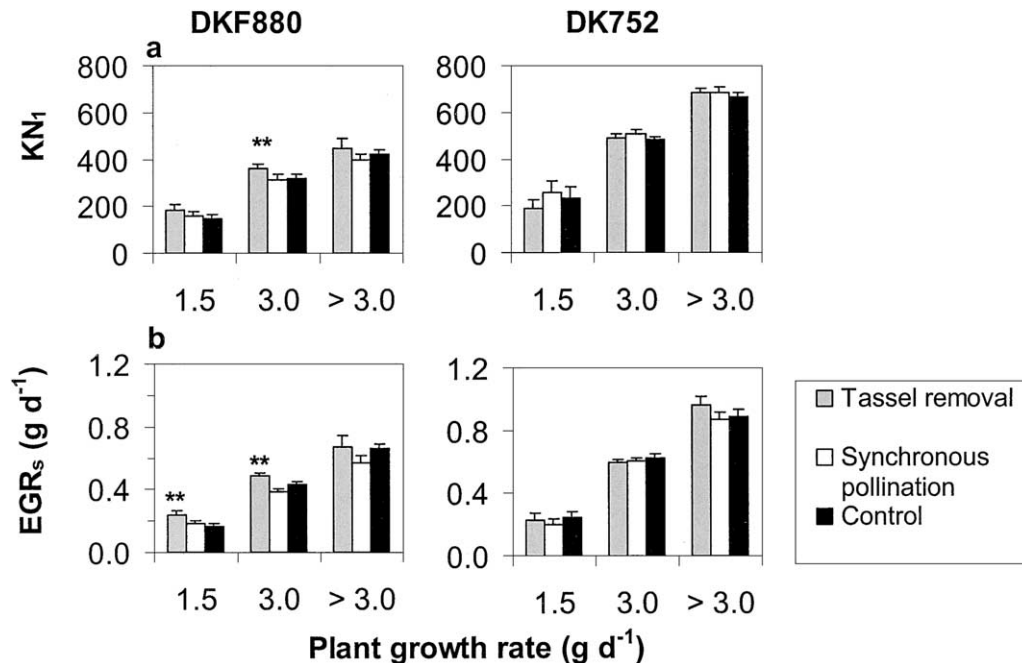


Fig. 4. (a) Mean kernel number per uppermost ear (KN<sub>1</sub>) vs. 1.5-g d<sup>-1</sup> intervals of plant growth rates during a period bracketing silking (PGR<sub>s</sub>), (b) mean growth rate of the topmost ear (EGR<sub>s</sub>) vs. 1.5-g d<sup>-1</sup> intervals of PGR<sub>s</sub> in an older (DKF880) and newer hybrid (DK752) in Exp. 3. Values at the x axis indicate the upper limit of the interval unless indicated otherwise. Bars indicate the standard error. Number of individuals in each treatment ranged from 10 to 40 depending on the range of PGR<sub>s</sub>. \*\* indicates significantly different from the control at  $P < 0.05$ .

nard, 1979; Edmeades et al., 1993; Vega et al., 2001a) was not significantly different between hybrids ( $P > 0.05$ ,  $1.28 \pm 0.44$  and  $0.90 \pm 0.073$  mg kernel<sup>-1</sup> d<sup>-1</sup> for DKF880 and DK752, respectively). Therefore, the similar KN<sub>1</sub> response to synchronous pollination at low PGR<sub>s</sub> and the lack of significant differences in minimum assimilate requirement per kernel between hybrids are in accordance with the similar KN<sub>1</sub> per unit of EGR<sub>s</sub> up to EGR<sub>s</sub> about  $0.5$  g d<sup>-1</sup> (Fig. 3b).

## CONCLUSIONS

Results reported in the current study elucidate the mechanisms involved in differences in kernel set between older and newer hybrids. Newer Argentinean hybrids set more kernels per unit PGR<sub>s</sub> than older Argentinean hybrids as indicated by (i) the lower threshold PGR<sub>s</sub> for kernel set and (ii) the greater potential kernel number at high availability of resources per plant, for newer than for older hybrids. At low and intermediate PGR<sub>s</sub>, the greater kernel set per unit of PGR<sub>s</sub> in newer vs. older hybrids was attributable to a greater partitioning of dry matter to the topmost ear during the period bracketing silking, whereas the number of kernels set per unit of ear growth did not differ. The KN<sub>1</sub> response to detasseling in the older hybrid, but not in the newer one, and the lack of KN<sub>1</sub> response to synchronous pollination in both hybrids supported this contention. In contrast, kernel set per unit of ear growth rate was greater in newer than in older hybrids when PGR<sub>s</sub> was high. The greater partitioning of dry matter to the ear in newer hybrids will contribute to greater yield stability and harvest index stability relative to those of older hybrids when resources per plant decreases. In addition,

the greater partitioning of dry matter to the ear and the greater number of kernel set per unit of ear growth rate at high PGR<sub>s</sub> for newer than for older hybrids will contribute to greater yield in newer than in older hybrids grown at low plant density or in case of an irregular plant stand (i.e., gaps in the row). The failure of others (cf., Tollenaar et al., 1992; Maddonni et al., 2000; Luque, 2000) to show a significant difference between older and newer hybrids in the threshold PGR<sub>s</sub> for kernel set may be attributable to the use of analyses based on plot means, which do not allow precise estimations of the KN<sub>P</sub>-PGR<sub>s</sub> relationship at low PGR<sub>s</sub>. Results of this study indicate that genetic yield improvement in maize is attributable, in part, to increased partitioning of dry matter to the ear during the critical period bracketing silking.

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