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# Determination of seed number in sea level quinoa (*Chenopodium quinoa* Willd.) cultivars

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**Abstract**

The identification of the critical period for seed determination is a central aspect for adequate crop management for higher yields. As important variability in the developmental stages associated to this critical period and its duration has been detected for several crops, results from one species cannot be extrapolated to other. The flowering period, from first anthesis to the end of flowering, appears as the most sensitive to the environment in quinoa; as results from experiments conducted under field conditions in the Argentinean pampas, using sea level quinoa cultivars adapted to temperate environments, suggest. Data from a combination of four cultivars, three densities and 2 years were used to estimate the association (adjusted $R^2$) between crop biomass increment (g m$^{-2}$) and seed number (seeds m$^{-2}$) for four phases: vegetative, reproductive, flowering and seed filling. The strongest association was that detected for the flowering phase ($R^2 = 0.71$), followed by seed filling ($R^2 = 0.64$); associations for the two phases preceding first anthesis were low. Differences in biomass increment during flowering where strongly associated with crop growth rate (g m$^{-2}$ day$^{-1}$) during flowering ($R^2 = 0.86$); crop growth rate was associated to differences in the amount of intercepted photosynthetically active radiation (MJ m$^{-2}$ day$^{-1}$) ($R^2 = 0.79$) and radiation use efficiency ($R^2 = 0.71$). Significant differences between cultivars were detected for reproductive efficiencies (seeds per gram of panicle biomass at the end of flowering or seeds per gram of panicle biomass increment during flowering) in 1 year, but differences in biomass partitioning to the inflorescence during flowering were not significant. A negative association between reproductive efficiencies and panicle biomass was observed, and possible determinants of this association are discussed in the light of available knowledge for the species. As in several other crop species, maximization of growth during the late reproductive period appears as the main target for crop management. As radiation interception efficiencies were usually below 0.95 this variable explained most variation in growth during flowering; but even crops managed for maximum radiation interception could exhibit differences in biomass increment associated to variability in radiation use efficiency and perhaps the duration of flowering.


*Keywords:* Quinoa; *Chenopodium quinoa*; Seed number; Critical period; Yield determination

## 1. Introduction

The period in the crop cycle during which seed number is determined is usually named critical period (Kiniry and Ritchie, 1985; Cantagallo et al., 1997; Egli, 1998; Andrade et al., 1999; Kantolic and Slafer, 2005; Takai et al., 2006). The study of the association of seed number with climate and soil factors during that period (e.g. Fischer, 1985; Chimenti and Hall, 2001; Cantagallo et al., 2004; Prystupa et al., 2004; Poggio et al., 2005; D'Andrea et al., 2006); and the processes involved in it (Vega et al., 2000, 2001a,b; Cantagallo and Hall, 2002) has provided a robust tool for agronomic management (Egli, 1998, and see further references in Andrade et al., 2005).

Crop biomass increment during that critical period often expressed using crop growth rate as its principal determinant integrates the effect of genotypic, climatic and soil factors and is the most important variable controlling seed number (Andrade et al., 2002), under the assumption that seed number determination is mostly source limited (Evans, 1993; Egli, 1998). Factors that affect the rate of crop growth during this critical period and

*Abbreviations:* $BI_{FL}$, biomass increment (g m$^{-2}$) during flowering; $CGR_{FL}$, crop growth rate (g m$^{-2}$ day$^{-1}$) during flowering; SN, seed number (seeds m$^{-2}$); SW, individual seed weight (mg); $PB_{EF}$, panicle biomass at the end of flowering (g m$^{-2}$); $PBI_{FL}$, panicle biomass increment during flowering (g m$^{-2}$); $RE_{EF}$, reproductive efficiency for panicle biomass at the end of flowering (SN $PB_{EF}{}^{-1}$); $RE_{FL}$, reproductive efficiency for panicle biomass increment during flowering (SN $PBI_{FL}{}^{-1}$); PAR, photosynthetically active radiation (MJ m$^{-2}$ day$^{-1}$); IPAR, intercepted PAR (MJ m$^{-2}$ day$^{-1}$); F, fraction of PAR intercepted at midday; FD, daily fractional interception; RUE, radiation use efficiency (g MJ PAR$^{-1}$)

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its duration have been analyzed for their effect on grain number (Andrade et al., 2000; Arisnabarreta and Miralles, 2004; Demotes-Mainard and Jeuffroy, 2004; Gonzalez et al., 2005; D'Andrea et al., 2006). The discovery that variation in photoperiod during the critical period in wheat can affect seed number inspired an elegant hypothesis about possible avenues to increase yield through breeding (Slafer et al., 1996, 2001).

Differences in seed number other than those caused by variation in crop biomass increment during the critical period can also be attributed to the partitioning of biomass to the reproductive structures (D'Andrea et al., 2006) and the efficiency with which reproductive biomass increment during the critical phase is translated into seed number (Charles-Edwards et al., 1986; Abbate et al., 1998; Egli, 1998; Echarte et al., 2000; Vega et al., 2001a).

The part of the crop cycle included in the critical period exhibit marked differences between species (i.e. Egli, 1998), and these differences also lead to differences in the optimal management needed to achieve higher yields. Quinoa (*Chenopodium quinoa* Willd.) is an Andean species of high-nutritive value, associated to its high-protein quality (Ruales and Nair, 1992). It is cultivated as a cereal over a wide range of latitudes and altitudes, from Southern Colombia to Southern Chile (Wilson, 1990), and its particular tolerance to adverse environments characterized by low temperatures, precipitation and even limitations associated to salinity lead to attempts to cultivate it in many areas (Jacobsen, 2003). Knowledge about factors determining differences in seed number in this species is null, and this paper uses data from field experiments, combining different cultivars, densities and years, to generate some understanding of the factors and processes behind differences in seed number. The main objectives are:

1. To analyze the association between seed number and crop biomass increment during different phenological phases, in order to identify the critical phase determining this yield component in quinoa.
2. To analyze the variables determining differences in crop biomass increment during the most sensitive phase, as a guide to crop management for achieving optimal seed yield.
3. To examine the determination of seed number in terms of its three components: crop biomass increment during the critical phase, biomass partitioning to reproductive structures, and seed number per unit reproductive biomass.

## 2. Materials and methods

### 2.1. Experimental design and growing conditions

Sea level quinoa (*Chenopodium quinoa* Willd.) genotypes from Chile or selected from lines of that origin were cultivated in Argentina at the Instituto Nacional de Tecnología Agropecuaria (INTA) Experimental Station in Pergamino (33°56′S, 60°35′W), on a silty clay loam soil (Typic Argiudoll, Soil Taxonomy, U.S. Department of Agriculture) in two consecutive years. Sea level quinoa are traditionally grown at low altitudes in Central and Southern Chile (Tapia et al., 1979) and they low photoperiod sensitivity make them suitable for cultivation in temperate environments (Bertero et al., 1999). Mean soil properties in the first 20 cm of soil depth at sowing were: pH (in water 1:2.5) 5.5; organic matter 28.5 g $kg^{-1}$; organic nitrogen 1.4 g $kg^{-1}$, mineral phosphorus 41 g $kg^{-1}$ and electric conductivity 0.144 dS $m^{-1}$.

Four cultivars: NL-6 (Holland), RU-5 (UK), CO-407 (USA) and Faro (Chile), were sown at two densities in a factorial experiment arranged in a randomized complete block design with four replicates. All these cultivars belong to the sea level quinoa group (G4, Bertero et al., 2004a,b) Sowing dates were 28 August 2003 and 7 September 2004. Plots were hand-planted and thinned to 22 (D1) and 33 (D2) pl $m^{-2}$ in rows 0.45 and 0.30 m apart in 2003 and to 33 (D2) and 66 (D3) pl $m^{-2}$ in rows 0.30 and 0.15 m apart, respectively, in 2004. Densities were changed from 2003 to 2004 because no treatment reached full radiation interception during the first year. Plot size ranged from 12 to 23 $m^2$, depending on the density. Plots received supplementary irrigation and fertilization at sowing (20 kg P and 18 kg N $ha^{-1}$) and two urea applications (totaling 200 kg N $ha^{-1}$) at 30 and 60 days after emergence to minimize nutrient restrictions. Soil nitrate availably at sowing was low (below 15 kg N $ha^{-1}$ as $NO_3^-$ for the 0–60 cm depth) and N doses were decided on the basis of previous reports of maximum yields being achieved between 160 (Jacobsen et al., 1994) and 225 kg N $ha^{-1}$ (Berti et al., 2000). To prevent insect pests and fungal diseases insecticides and fungicides were applied regularly and weeds removed by hand. Mean air temperature (°C) and total radiation values (MJ $m^{-2}$ $day^{-1}$) were obtained from a weather station (Li-COR 1200, Lincoln, NE) located near the experimental field.

### 2.2. Biomass, leaf area index (LAI) and developmental stage

Starting 1 month after emergence, plants were sampled every week (2003) or fortnightly (2004) for measuring above-ground biomass and leaf area index (LAI). Five contiguous plants per plot (those adjacent to the site where radiation interception was measured) were harvested. Biomass was separated into green leaves (main stem and branches), senescent leaves, stem (main stem and branches) and inflorescences when present. Samples were dried in air-forced drying oven at 70 °C to constant weight. Leaf area was measured with a Li-Cor LI-3100 leaf area meter (LI-Cor Inc., Lincoln, Nebraska) and expressed on a per ground area basis.

Developmental stages (recorded when three out of five sampled plants within each plot reached the stage) were determined as: emergence, visible flower bud (Bertero et al., 1999), first anthesis (at least one flower opened), end of anthesis (no more flowers opened determined from observations on the main inflorescence), and physiological maturity (visually determined from examination of seeds on the medium third of the inflorescence). These stages defined four developmental phases: vegetative, reproductive, flowering and seed filling. Biomass increment per phase (g $m^{-2}$) was estimated from linear interpolation between sampling dates when samplings did not coincide with one of the developmental stages considered.

### 2.3. Light interception

Photosynthetically active radiation (PAR) interception was measured with a 0.8-m long ceptometer (Decagon Devices Inc., Pullman, Washington) between 11:30 and 14:00 h on clear days. Six measurements were taken in each replicate. The first and last measurements were made above the canopy to determine incident PAR ($I_o$). The other four measurements were taken at the soil surface placing the sensor below the canopy and moving it parallel to rows at regular intervals while taking the readings to determine transmitted PAR ($I_t$) as indicated by Charles-Edwards and Lawn (1984). The fraction of PAR intercepted at midday ($F$) was calculated as

$$F = \frac{I_o - I_t}{I_o}$$

where $I_o$ and $I_t$ are the means of the measurements above and below the canopy.

Daily fractional interception ($F_D$) was calculated as (Charles-Edwards and Lawn, 1984):

$$F_D = \frac{2F}{1+F}$$

and applied to corresponding daily integrals of PAR to estimate intercepted PAR (IPAR). Daily incident PAR was calculated as the incident total solar radiation measured with a standard weather station 200 m from the plots multiplied by 0.45 (Monteith, 1965). Daily values were summed from emergence for each plot to obtain accumulated IPAR. Radiation use efficiency (RUE, g MJ PAR$^{-1}$) for the flowering period was estimated as the slope of the association between accumulated growth during flowering and accumulated IPAR for the same period; RUE values for each cv. $x$ density combination were estimated as the quotient between accumulated growth and accumulated IPAR for each particular treatment.

### 2.4. Data analysis

Linear regression and analysis of variance were used; data representing fractions were arcsine transformed prior to analysis. $R^2$s are expressed as adjusted $R^2$ (Dike, 1997). The critical period for seed determination was defined as that with the higher $R^2$ from the association between crop growth for a given phenological phase and seed number as in Takai et al. (2006). Daily thermal time increment was estimated as the difference between daily average temperature and Tb (base temperature); for days where average temperatures were lower than a Tb value of 3 °C, this increment was assumed to be zero (Bertero et al., 1999). Reproductive efficiencies were estimated in two ways, as the ratio between seed number and panicle biomass increment during flowering ($RE_{FL}$) or the ratio between seed number and panicle biomass at the end of flowering ($RE_{EF}$). The boundary regression line in Fig. 4 was fitted using the Blossom statistical package available from the US Geological Survey (www.fort.usgs.gov/products/software/blossom.asp) as the equation estimated for the 0.9 quantile (Cade and Noon, 2003). Average differences between observed and predicted seed numbers were expressed as root mean square errors (RMSE) values (Mayer and Butler, 1993). The relative proportion of reproductive biomass shown in Fig. 6 were estimated as the reproductive to total aerial biomass quotient at each sampling date after first anthesis.

## 3. Results and discussion

### 3.1. General results

Average temperatures during crop cycle were similar for both years (18.4 and 18.8 °C for 2003 and 2004, respectively, but radiation was lower in 2004 (7.8 against 9.4 MJ PAR m$^{-2}$ day$^{-1}$). The combined data from both years explored a wide range of biomass and seed number values (Tables 1 and 2). Significant differences between cultivars were detected for most of the variables analyzed, but differences between densities were detected

Table 1
Yield and yield components for the experimental year 2003

| Genotype | Density (pl m$^{-2}$) | Yield (g m$^{-2}$) | Biomass (g m$^{-2}$) | Harvest index | SN seeds (m$^{-2}$) | SW (mg) |
|---|---|---|---|---|---|---|
| NL-6 | D1 | 208.0 | 453.8 | 0.461 | 79001 | 2.649 |
| | D2 | 306.8 | 673.7 | 0.457 | 111764 | 2.736 |
| RU-5 | D1 | 285.6 | 677.6 | 0.422 | 128721 | 2.232 |
| | D2 | 285.3 | 685.8 | 0.422 | 130231 | 2.181 |
| CO-407 | D1 | 347.6 | 865.8 | 0.401 | 121900 | 2.910 |
| | D2 | 372.5 | 876.4 | 0.428 | 130079 | 2.890 |
| Faro | D1 | 436.1 | 1233.2 | 0.357 | 194132 | 2.244 |
| | D2 | 493.5 | 1222.7 | 0.406 | 204975 | 2.406 |
| Mean | | 342.0 | 836.1 | 0.42 | 137688 | 2.531 |
| S.E. | | 31.1 | 88.6 | 0.016 | 13767 | 0.107 |
| G | | $P<0.001$ | $P<0.001$ | $P<0.001$ | $P<0.001$ | $P<0.001$ |
| D | | $P<0.055$ | Ns | $P<0.12$ | Ns | Ns |
| G × D | | Ns | Ns | Ns | Ns | Ns |

D1 (22 pl m$^{-2}$), D2 (33 pl m$^{-2}$), SN (seeds m$^{-2}$) and SW (individual seed weight, mg).

Table 2
Yield and yield components for the experimental year 2004

| Genotype | Density (pl $m^{-2}$) | Yield (g $m^{-2}$) | Biomass (g $m^{-2}$) | Harvest index | SN ($m^{-2}$) | SW (mg) |
|---|---|---|---|---|---|---|
| NL-6 | D2 | 248.2 | 631.8 | 0.397 | 105187 | 2.365 |
| | D3 | 136.1 | 451.8 | 0.300 | 59076 | 2.314 |
| RU-5 | D2 | 84.4 | 283.7 | 0.267 | 41311 | 1.982 |
| | D3 | 99.4 | 390.3 | 0.228 | 49317 | 1.962 |
| CO-407 | D2 | 141.6 | 582.9 | 0.203 | 52234 | 2.620 |
| | D3 | 199.4 | 771.2 | 0.256 | 79504 | 2.548 |
| Faro | D2 | 111.3 | 546.4 | 0.182 | 54286 | 2.045 |
| | D3 | 231.3 | 1090.6 | 0.209 | 107110 | 2.146 |
| Mean | | 156.5 | 593.6 | 0.255 | 68511 | 2.25 |
| S.E. | | 44.0 | 147.5 | 0.030 | 18741 | 0.086 |
| G | | Ns | $P<0.02$ | $P<0.001$ | Ns | $P<0.001$ |
| D | | Ns | Ns | Ns | Ns | Ns |
| G × D | | $P<0.08$ | Ns | $P<0.08$ | $P<0.08$ | Ns |

D2 (33 pl $m^{-2}$), D3 (66 pl $m^{-2}$), SN (seeds $m^{-2}$) and SW (individual seed weight, mg).

only for cultivar NL-6 during 2003; hence the analysis presented here is focused in the comparison between cultivars but not between densities. The 2004 experiment suffered from waterlogging around mid-November, coinciding with the flowering phase in some of the cultivars. This stress prematurely stopped the expansion of leaf area and had several consequences on growth and yield, particularly evident on harvest index values (Table 2), which suffered important reductions compared with 2003. Growth analysis for these experiments is the subject of a separate paper (Ruiz and Bertero, unpublished) and will not be further analyzed in this article.

### 3.2. General patterns of growth

Fig. 1 illustrates the temporal pattern of growth of a sea level quinoa crop using cultivar Faro. Total aerial biomass growth and that of stems (main stem + branches) and reproductive structures (inflorescence parts + seeds) is presented; the example reflects the general pattern exhibited by all cultivars and treatments in these experiments. After an initial period (≈1 month) during which total growth is minimal, stem growth starts shortly after the beginning of the reproductive phase, and stabilizes mid-way into the seed filling phase. Reproductive biomass, on the other hand, is almost negligible before first anthesis and its growth accelerates after that. Growth of non-seed parts of the inflorescence continues during seed filling (average 150 g $m^{-2}$), as calculated from the difference between inflorescence biomass at harvest (seeds plus non-seed parts) minus seeds and that of it at the end of flowering. Active seed filling (beginning of the linear phase of biomass accumulation by the seed) starts shortly after the end of flowering (≈2 days) under the conditions of these experiments, and ≈85% of inflorescence biomass at harvest is in the seeds.

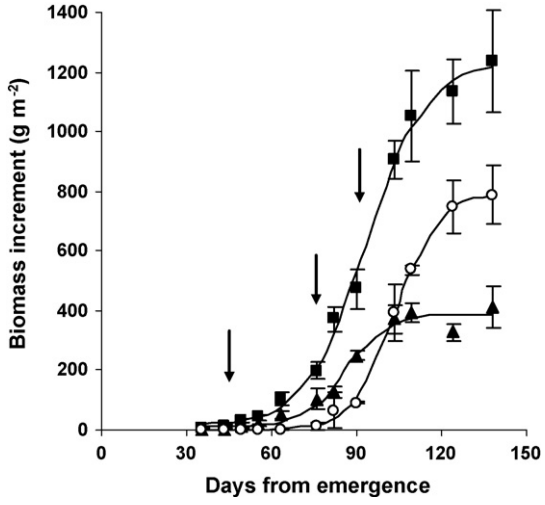


Fig. 1. Temporal evolution of biomass accumulation in different organs in quinoa for cv. Faro in 2003 (density 22 pl $m^{-2}$). Vertical arrows (from left to right) indicate the dates of: visible floral buds, 1st anthesis and end of flowering, respectively. Symbols: (■) total aerial biomass, (▲) stem biomass and (◯) inflorescence biomass. Vertical bars represent ± 1 standard deviation.

### 3.3. Assocciation between seed number and growth per phase

Crop yield exhibited a very strong association with seed number ($R^2 = 0.94$, $P \ll 0.01$, ranges 12.8–566.6 g $m^{-2}$ and 7211–240363 seeds $m^{-2}$, respectively), and confirmed the initial assumption of seed number as the main determinant of yield in quinoa. There was no association between seed number and seed weight ($R^2 = 0.03$). The association between accumulated growth and seed number for the four phases analyzed: vegetative, reproductive, flowering and seed filling, and for the whole growth period is shown in Fig. 2.

Weak associations were found for the two phases preceding first anthesis, and the strongest association was that detected for the flowering period ($R^2 = 0.71$). Association with growth during seed filling was also comparatively high ($R^2 = 0.64$), suggesting that conditions during this period could have an impact on seed number. Alternative interpretations for the association with growth during this last phase are that it reflects a carry over effect of conditions during the previous phase, but the magnitude

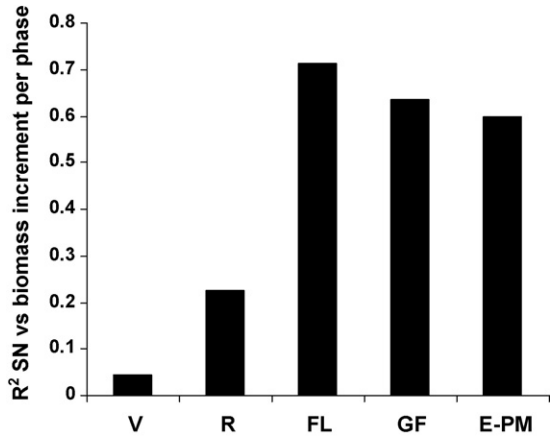


Fig. 2. Degree of association (adjusted $R^2$) between seed number (SN) and biomass increment per phase ($g\,m^{-2}$) for different periods: V (vegetative), R (reproductive), FL (flowering), GF (grain filling) and E-PM (emergence-physiological maturity).

of the association of growth between both phases ($R^2 = 0.38$) is indicative of a partial independence of growth for these two periods. Consideration of a longer period did not result in a marked improvement of the strength of the association, as in the case of the emergence-physiological maturity ($R^2 = 0.60$) and anthesis-maturity ($R^2 = 0.74$) periods. Evidence of changes in seed set in response to source-sink manipulations conducted at the end of flowering, however, suggest that the relevance of this last phase for seed number determination needs to be explored (Bertero, unpublished). An alternative analysis, in which the association between biomass increment for periods of fixed durations in thermal time units (150 °Cd) centered around 1st anthesis, and seed number was explored, exhibited a similar response pattern.

This temporal pattern could have arisen from changes in crop capacity to capture resources more than from a sudden change in the intrinsic sensitivity to environmental conditions of the processes determining seed number. If more resources are captured during a particular phase, then a higher proportion of total growth is explained by growth during that period and will have a higher relative impact on seed number. The analysis of the temporal evolution of average interception efficiencies and the proportion of total biomass accumulated per period does not match that of $R^2$s shown in Fig. 2 (data not shown). In conclusion, Fig. 2 strongly supports the notion of the flowering period as the one with the highest sensitivity to environmental factors. In Section 3.4, processes occurring during this phase are analyzed in more detail.

### *3.4. Components of the association between seed number and growth during the flowering period*

Fig. 3A shows the association between seed number and biomass increment for the flowering period. Variation in crop biomass increment during flowering ($BI_{FL}$) can arise from differences in crop growth rate and duration, and crop growth rate ($CGR_{FL}$) can be further decomposed into radiation use efficiency (RUE, g MJ $PAR^{-1}$) and mean (daily) radiation intercepted. There was a strong association between accumulated

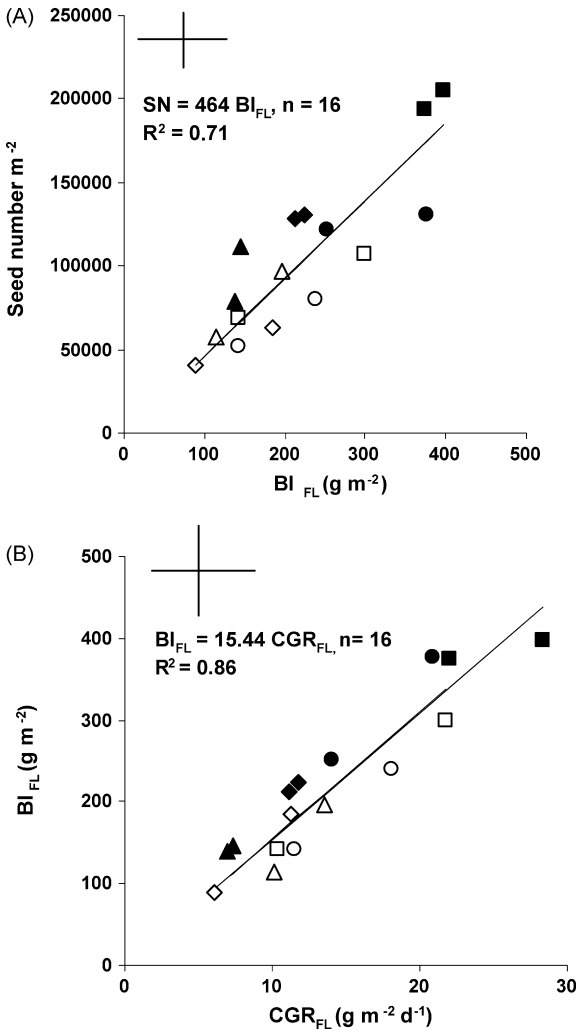


Fig. 3. Association between average SN and crop growth ($BI_{FL}$) (A) and of crop growth with crop growth rate ($CGR_{FL}$), during flowering (B). Symbols: cv. NL-6 (▲), cv. RU-5 (♦), cv. CO-407 (●) and Faro (■); year 2003 (closed symbols), year 2004 (open symbols). Each data point represents the mean of a cultivar x density treatment in each year. Regressions were forced through the origin as intercepts were not significantly different from zero. Bars indicate standard deviations.

growth ($BI_{FL}$) and $CGR_{FL}$ (Fig. 3B, $R^2 = 0.86$), the association with phase duration was non-significant ($R^2 = 0.01$). $CGR_{FL}$ exhibited a positive association with both average total intercepted radiation ($R^2 = 0.79$) and RUE ($R^2 = 0.71$, range 1.33–3.35 g MJ $PAR^{-1}$)). The slope of the association between $BI_{FL}$ and accumulated intercepted radiation (RUE) during that period was 2.33 g MJ $PAR^{-1}$. Average intercepted radiation was positively associated with average interception efficiency ($R^2 = 0.69$) and less with average incident radiation ($R^2 = 0.35$), discarding the impact of an important effect of the temporal pattern of radiation experienced during these experiments.

### *3.5. Biomass partitioning to panicles, reproductive efficiency and harvest index*

$BI_{FL}$ is just one of three factors determining seed number, the other two are partitioning of that growth to the inflores-

cence (panicle) and reproductive efficiency ($RE_{FL}$, seeds per gram panicle biomass increment during flowering). The first two variables are determined at the end of flowering, but the last one can also be affected by conditions during later phases, if differences in seed set or seed abortion are involved in final seed number. Mean biomass partitioning to the panicle during flowering (panicle biomass increment/crop biomass increment) ranged from 0.29 to 0.40, but differences between cultivars were not significant, and average partitions during 2003 were moderately higher than during 2004 (0.38 against 0.34, respectively). Adding this variable to the association between seed number and $BI_{FL}$ in a multiple regression did not result in an improvement of the association.

Significant differences between cultivars were detected for $RE_{FL}$ during 2003, however ($P<0.01$), but not during 2004. For 2003, average $RE_{FL}$s ranged from a maximum of 2250 seeds $g^{-1}$ in NL-6 to 1031 seeds $g^{-1}$ in CO-407; values during 2004 were on average 14% lower and a narrower range of variation was observed (1058–1411 seeds $g^{-1}$) although cultivar rankings were maintained. Average panicle biomass at the end of flowering ($PB_{EF}$) and average panicle biomass increment during flowering ($PBI_{FL}$) were strongly associated ($PB_{EF} = 1.13 \times PBI_{FL}$, $R^2 = 0.95$, $n = 16$), and the association between $PB_{EF}$ and reproductive efficiency ($RE_{EF}$, calculated on the basis of $PB_{EF}$) is shown in Fig. 4.

There is a negative association between reproductive efficiency and panicle biomass, expressed as an upper boundary in efficiency that declines as panicle biomass increases. Variation within a given panicle biomass value is also very high, covering almost the whole range of values under the upper boundary. A negative association between RE and inflorescence biomass at the end of the critical period was reported for other crops (Abbate et al., 1998; Vega et al., 2001a). It could be hypothesized that values near the boundary of the response represent maximum attainable efficiencies, while the distance between this upper limit and REs observed under a given circumstance is associated to a stress factor. The experiments reported here were not designed to test this hypothesis, but the fact that RE values during 2004 were on average 26% lower than those of 2003, probably as a consequence of a water excess stress during flowering, is consistent with it and deserves further exploration. An alternative interpretation to the dispersion of values at low panicle biomass was offered by Vega et al. (2001a) as a result of high unstability of RE, but the fact that most data were concentrated near the upper limit in a more recent experiment where no abiotic stresses were identified suggest that is not the case of quinoa (unpublished results).

Several of the associations explored here could be used to predict seed number early in development from growth during flowering, and if the physiological basis of RE determination is understood, a more powerful tool would be available. The association between seed number (SN) and biomass increment during flowering is presented in the following equation:

$$SN = 464 \times BI_{FL}\ (R^2 = 0.71, n = 16, P < 0.01, \text{intercept not significantly different from 0}) \quad (1)$$

Although only $CGR_{FL}$ played a significant rôle in variation of accumulated growth in these experiments, the approach based on total growth was considered a more meaningful expression of factors controlling SN. This equation is similar to the approach used to model SN in several species (Ritchie and Otter, 1985; Andrade et al., 2002; Gerik et al., 2004). The association of SN with $CGR_{FL}$, on the other side, was weaker than that with total growth ($R^2 = 0.54$).

Given the strong association between crop and panicle growth during flowering ($R^2 = 0.89$, Fig. 5), the lack of significant differences in partitioning between cultivars, and the strong association between panicle growth during flowering and panicle biomass at the end of flowering, Eq. (1) can be replaced by the association between SN and panicle biomass at the end of flowering (Eq. (2)), a similar approach to that used to estimate SN in wheat and barley (Bindraban et al., 1998; Moreno-Sotomayor

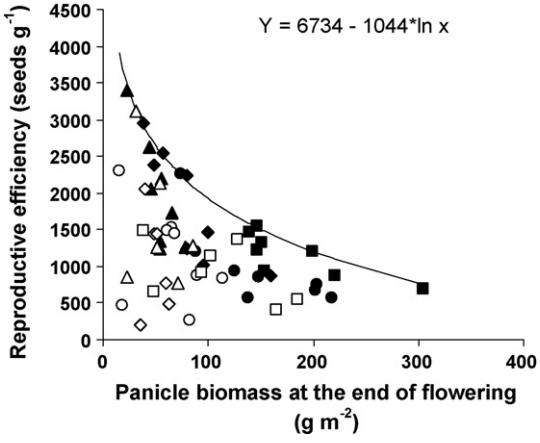


Fig. 4. Asocciation between reproductive efficiency ($RE_{EF}$, seeds per panicle biomass at the end of flowering) and panicle biomass at the end of flowering. The equation was fitted to the 0.9 quartile using the Blossom statistical package. Symbols as in Fig. 3.

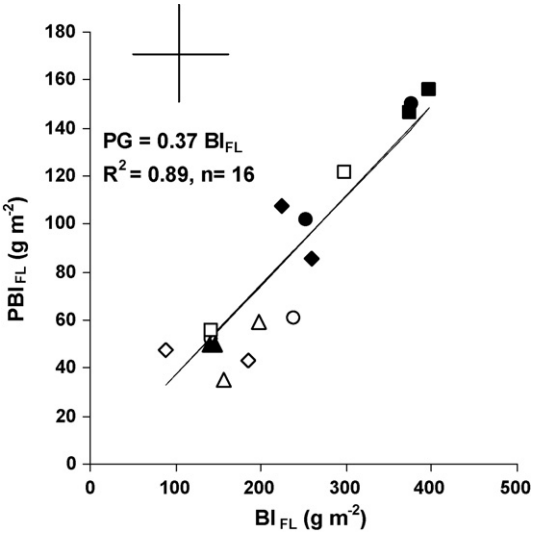


Fig. 5. Association between panicle ($PBI_{FL}$, g $m^{-2}$) and crop ($BI_{FL}$ g $m^{-2}$) biomass increment during flowering. Symbols as in Fig. 3.

and Weiss, 2004; Prystupa et al., 2004):

$$SN = 1044 \times PB_{EF}\ (R^2 = 0.60, n = 16, P < 0.01,\ \text{intercept not significantly different from 0}) \quad (2)$$

A preliminary test of the predictive capacity of these equations was done by applying them to the prediction of seed number in an experiment conducted at the Faculty of Agronomy of the University of Buenos Aires during 2005, involving shading treatments at different phenological phases in cv. NL-6 (unpublished). Average yield in that experiment was 350 g m$^{-2}$. Seed number were reasonably well predicted by these equations (RMSE values were 15 and 18% of average seed number for Eqs. (1) and (2), respectively).

From the results of these experiments it is clear that crop growth during flowering is the most critical, but not the only factor determining seed number. The relevance of this other factors can be seen in an example. NL-6 and Faro are two cultivars with contrasting values of seed number and growth during flowering, as seen in results from the experiment conducted during 2003 at a density of 33 plants m$^{-2}$ (D2) using the variables described before:

$$SN_{NL\text{-}6} = 146\ g\ m^{-2} \times 0.33\ g\ g^{-1} \times 2250\ seeds\ g^{-1} = 111760\ seed\ m^{-2},$$

$$SN_{Faro} = 398\ g\ m^{-2} \times 0.39\ g\ g^{-1} \times 1313\ seeds\ g^{-1} = 204970\ seed\ m^{-2}$$

where variables, from left to right are: $BI_{FL}$, partitioning to panicles and $RE_{FL}$, respectively. Crop growth during flowering was 2.73 times higher for Faro compared with NL-6, but the advantage in SN was lower (1.83 times) as a consequence of differences in reproductive efficiency. This higher $RE_{FL}$ for NL-6 was insufficient to compensate for the lower growth and partitioning to panicles, but was associated with a higher harvest index (0.46 and 0.40, respectively). The temporal pattern of biomass partitioning to reproductive structures after first anthesis (a potential explanation to differences in harvest index in this example) is presented in Fig. 6. Although the proportion of biomass in reproductive structures is lower in NL-6 than Faro at the start of the period, it increases at a higher rate and reaches 80% of total biomass at maturity, while the final value for Faro is only 70%.

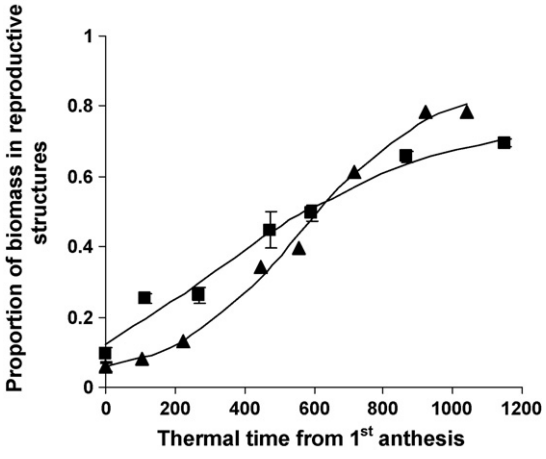


Fig. 6. Temporal evolution of the proportion of total aerial biomass assigned to reproductive structures as a function of thermal time (Tb 3 °C) from first anthesis in cvs. NL-6 (▲) and Faro (■) during 2003 at a density of 33 pl m$^{-2}$. Vertical bars represent ± 1 standard deviation.

When analyzed for the whole set of treatments, however, no association was found between biomass partitioning and harvest index, and only a weak association with $RE_{FL}$ ($R^2 = 0.26$), but it does not diminish its potential relevance for the specific comparison discussed before.

## 4. Conclusions

As in several other species (e.g. wheat, barley, rice, maize and sorghum) the period of active inflorescence growth before seed filling appears as the most sensitive to environmental conditions in terms of determination of seed number in sea level quinoa cultivars. Panicle growth before first anthesis was low for the cultivars studied in these experiments, but exponential growth of reproductive structures started shortly after first anthesis (Fig. 1). If examined in terms of phenology, quinoa compares better to species like soybean, where the critical period starts after first anthesis (Egli, 1998; Kantolic and Slafer, 2005), than to species like wheat or barley, where most variation in SN is resolved before flowering (Gonzalez et al., 2005; Arisnabarreta and Miralles, 2006). An important component of this difference is the timing of inflorescence growth in these species. This indirect approach to the identification of the critical period in quinoa needs to be confirmed by direct manipulation of environmental conditions during plant growth using shading treatments (Kiniry and Ritchie, 1985; Cantagallo and Hall, 2002; Gonzalez et al., 2005); these experiments are being performed under field conditions at the Faculty of Agronomy of the University of Buenos Aires.

Partly because of the plant densities used, but also because of environmental conditions during 2004 (water excess around flowering) crops never reached full radiation interception, hence most variability in growth was associated to variation in interception efficiency, which was in turn mostly caused by differences in leaf area index. Higher light extinction coefficients ($k$) at the highest densities during 2004 did not compensate for reduced leaf area index (data not shown). The obvious crop management indication arising from these results are that maximum crop growth rate (associated with interception efficiencies higher than 0.95) should be reached before, and sustained during the flowering period. If crops are managed in this way, then only variation in the duration of the flowering period and radiation use efficiency would remain as factors causing differences in crop growth during that phase. As higher radiation is usually correlated with higher temperatures in most environments, exploring an environment with higher radiation as a strategy to increase growth is not suggested as this would be detrimental to growth if duration of flowering is reduced. Although exploring a fairly narrow range of variation a positive and significant association of crop growth rate with radiation use efficiency was detected.

Associations of growth with duration of flowering were not significant in our experiments however, partly because of the narrow range of environmental conditions explored and of minor variation for duration of flowering between the cultivars tested. This does not discard an impact of duration if a wider range of conditions and cultivars are explored, as evidence from other crops indicates (Miralles and Richards, 2000; Gonzalez et al., 2005).

Partitioning of growth to the panicle was fairly stable, as arising from the strong, linear association between crop and panicle growth during the flowering phase (Fig. 5). An increase in reproductive partitioning has been the main target of breeding for higher yield in the last decades (Bingham, 1969; Slafer et al., 1990; Luque et al., 2006), and an important part of growth during flowering is partitioned to the stem (Fig. 1) suggesting that partitioning to the panicle could be targeted for improvement in quinoa. All this leaves reproductive efficiency as the last potential modulator of variation in SN. The fact that significant differences between cultivars were detected, and its potential relevance as a factor to be manipulated through breeding makes it an interesting trait to explore.

REs in our work are the opposite of $a_G$, or minimum assimilate flux requirement, in the model initially proposed by Charles-Edwards et al. (1986) and analyzed in detail by Egli (1998), who used it as a simple way to explain differences between species and also within species in the response of seed number to assimilate availability. A positive association between $a_G$ and seed growth rate was proposed as an explanation to some of the differences between cvs. and species, but this explanation does not seems to hold for quinoa where, although with a similar tendency, a very weak association was found ($R^2 = 0.29$). A second factor, morphological limitations (caused by inflorescence structure) to higher seed number, was analyzed in detail by Vega et al. (2001a) in maize and sunflower. Morphological limitations arise when the potential size of the inflorescence is determined before the critical period conditioning the number of seed that can be established (i.e. the size of the capitulum in sunflower) but this limitation does not seem to apply to quinoa, a species with a modular inflorescence where new branches and sub-branches can be added (Bertero et al., 1996). A saturation response of seed number to changes in inflorescence biomass was detected in maize only when allometric relationships were used to estimate the association between kernel number and crop growth rate at an individual plant level however, but not when using average plant data (Andrade et al., 1999). Variability in the temporal distribution of flower production, affecting competition between reproductive structures (either during flower formation or seed setting) could also be proposed as factors affecting reproductive efficiency, as in the case for the positive association between increasing flowering synchrony and pod or seed set in soybean and maize (Cárcova et al., 2000; Egli and Bruening, 2002; Egli, 2005).

Finally, a reduced capacity to translate an increase in growth into more seeds is a common trait of species and cultivars not intensively selected for high yields (e.g. Echarte et al., 2000 and examples in Evans, 1993), as is the case with the present situation of quinoa breeding (Jacobsen and Mujica, 2002; Bertero et al., 2004b).

Could variation in RE have a rôle additional to that of the other two components of the equation in improving seed yield? Egli (1998) using the equation of Charles-Edwards et al. (1986) argues that not, as a higher seed number under constant assimilate availability will result in compensations between seed number and seed size with no change in yield, but his reasoning does not takes into account the temporal component of growth, neither positive feedbacks of higher seed number on crop growth during seed filling (Reynolds et al., 2005 and examples in Evans, 1993) and even perhaps nitrogen uptake (Mi et al., 2000). The positive association between seed number, yield and RE (for a constant assimilate supply) reported by Abbate et al. (1998) is encouraging in that sense.

Finally, the equations derived from the analysis of the association between seed number and its determinants (Eqs. (1) and (2)) can be proposed as a useful instrument for the modelization of growth and yield of this species under non-limiting conditions, following the example of other crops. Models predicting the duration of different phenological phases and leaf appearance in response to temperature and photoperiod have been developed and validated for quinoa (Bertero et al., 1999; Bertero, 2001) and it would be possible to derive the required parameters for duration to and of flowering. Several of the parameters needed for the prediction of biomass accumulation are already available (Bertero et al., 2004a).

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