

Scientific article

Responses of garlic to environmental and agronomic factors.**I. Dynamics of dry matter production and leaf growth****Respuesta del ajo a los factores ambientales y agronómicos. I. Dinámica de la producción de materia seca y crecimiento de la hoja**José Alberto Saluzzo^{1*}; Paul Christiaan Struik²; Jorge Rattin¹¹ Cátedra de Horticultura, Universidad Nacional de Mar del Plata, Balcarce, Buenos Aires, R. Argentina.² Centre for Crop Systems Analysis, Department of Plant Sciences, Wageningen University & Research, Wageningen, The Netherlands.*E-mail: saluzzoj@yahoo.com / jrattin@mdp.edu.ar**Abstract**

The timing of planting (PD) and the size of garlic (*Allium sativum* L.) cloves (CS) are known to affect crop growth and its response to environmental conditions. This study aimed to understand the effects of environmental factors on garlic dry matter production in Balcarce (37°45'50" S and 58°17'20" W), Argentina. Experiments were conducted over two growing seasons, with treatments combining four PDs and two CS levels. No significant effects of PD or CS were found on the rate of total dry matter yield increase per degree day accumulated (DDA). Similarly, the relationship between bulb dry matter yield (BDMY) and DDA was consistent across all PDs, with BDMY tending to decrease with delayed planting. BDMY was slightly higher in Year 2. In Year 2, small seed cloves resulted in an earlier onset of bulbing with the latest PD. Bulb fresh matter yield (BFMY) increased steadily at early PDs, but slowed down with the latest PD. Leaf dry matter yield increase per DDA was higher in earlier PDs, particularly in Year 1, with no clear CS effect. While leaf area (LA) expansion per DDA showed no differences among PDs, it tended to decrease as PD was delayed. Overall, PD, CS, and the year of study significantly influenced leaf dry weight and LA. The onset of bulbing, defined by the bulb base diameter-to-pseudostem diameter ratio, was significantly modified by PD and CS. A strong relationship was found between LA and BFMY, which was also affected by PD and CS. The most notable effect of the year was on the rate of bulb and leaf growth, expansion, and the bulbing ratio. Environmental conditions, primarily temperature, combined with agronomic practices (PD and CS), affected these crop traits. These findings are expected to contribute to improve garlic production in southeastern Buenos Aires province.

Keywords: *Allium sativum* L.; Bulbing index; Planting date; Seed clove size; Specific leaf area.**Resumen**

Se sabe que el momento de siembra (FP) y el tamaño de los dientes de ajo (*Allium sativum* L.) (TD) afectan el crecimiento del cultivo y su respuesta a las condiciones ambientales. Este estudio tuvo como objetivo comprender los efectos de los factores ambientales en la producción de materia seca de ajo en Balcarce (37°45'50" S y 58°17'20" O), Argentina. Se realizaron experimentos durante dos temporadas de cultivo, con tratamientos que combinaron cuatro FP y dos niveles de TD. No se observaron efectos significativos de FP ni TD en la tasa de aumento del rendimiento total de materia seca por grado-día acumulado (GDA). De igual manera, la relación entre el rendimiento de materia seca de bulbos (RMSB) y el GDA fue consistente en todos las FP, con una tendencia a disminuir con el retraso de la siembra. El RMSB fue ligeramente mayor en el segundo año. En el segundo año, los dientes de semilla pequeños iniciaron más temprano la formación de bulbos con las FP más tardías. El rendimiento de materia fresca del bulbo (RMFB) aumentó de forma constante con las FP tempranas, pero se desaceleró con las FP más tardías. El aumento del rendimiento de materia seca foliar por GDA fue mayor en las FP más tempranas, especialmente en el primer año, sin un efecto claro en el TD. Si bien la expansión del área foliar (AF) por GDA no mostró diferencias entre las FP, tendió a disminuir a medida que se retrasó la FP. En general, la FP, el TD y el año de estudio influyeron significativamente en el peso seco de la hoja y el AF. El inicio de la bulbificación, definido por la relación entre el diámetro de la base del bulbo y el diámetro del pseudotallo, se vio significativamente modificado por la FP y el TD. Se observó una fuerte relación entre el AF y el RMFB, que también fue afectada por la FP y el TD. El efecto más notable del año se observó en la tasa de crecimiento y expansión de bulbos y hojas, así como en la tasa de bulbificación. Las condiciones ambientales, principalmente temperatura, combinadas con prácticas agronómicas (FP y TD), influyeron en estas características del cultivo. Se espera que estos hallazgos contribuyan a mejorar la producción de ajo en el sureste de la provincia de Buenos Aires.

Palabras clave: *Allium sativum* L.; Área foliar específica; Fecha de plantación; Índice de bulbificación; Tamaño de diente semilla.

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Introduction

Temperature is the most important factor affecting the development of garlic. Bulb growth and development are affected by temperatures during pre-planting storage, by temperature and photoperiod after planting, and by low red: far-red ratio of the light (Mann, 1952; Mann and Mingués, 1958; Ledesma *et al.*, 1983; Brewster, 1994; Micolis *et al.*, 2000; Atif *et al.*, 2019). Response of plant growth to these environmental factors may depend on genotype. The red variety of garlic in this article is one of six types grown in Argentina (Burba, 2013) and requires the highest cold and photoperiod induction for bulb formation.

There is some evidence on the effect of radiation on dry matter allocation in bulbs. The rate of initiation, differentiation and development of garlic cv. Bangladesh cloves increased with increasing light transmission through shade covers and were maximal in unshaded plots at Wye, UK (Rahim and Fordham, 1990). Light quality (i.e., low red: far-red ratio) is an important factor in stimulating bulbing (Brewster, 1994). Radiation and soil temperature are important for the induction of clove primordia (Mann and Mingués, 1958; Rahim and Fordham, 1990), but agronomic factors may be involved as well, such as seed clove size (Rahim and Fordham, 1990).

Dry matter production of garlic has been analyzed in response to nutrients (Bowalda, 1986; Bertoni *et al.*, 1992; Saluzzo, 2003), location (Ledesma *et al.*, 1997; Rizzali *et al.*, 2002; Portela, 2004) and planting date (Ledesma *et al.*, 1983; Stahlschmidt *et al.*, 1997; Portela, 2004; Alsup-Egbers *et al.*, 2020; Sultana *et al.*, 2022). In some of these studies, plant growth was related to the dynamics of leaf and bulb formation, and their interrelations, but information about the effects of environmental factors on garlic growth is scarce (Rahim and Fordham, 1994; Rizzali *et al.*, 2002; Portela, 2004).

Portela (2004) and Rizzali *et al.* (2002) studied the same ecophysiological garlic type III (white type) under field conditions (Burba, 2013). Portela (2004) analyzed the effects of temperature and day length on five phases: i) sprouting, ii) initial vegetative growth, iii) rapid vegetative growth, iv) rapid bulb growth and v) final bulb growth. These stages were determined on the basis of linear regression between dry matter production during plant growth and chronological time during crop

growth. Temperature interacted with photoperiod for the linear growth of the bulbs and for the bud differentiation moment on the axils of leaves that tended to be delayed at later planting dates. The minimum temperature at the appearance of cloves was determinant. Bulbing ratio (bulb diameter: pseudostem diameter) was defined but it was considered not suitable for defining different steps of the garlic crop ontogeny.

Rizzali *et al.* (2002) defined plant growth based on the partitioning of dry matter during the growing season. They distinguished four different partitioning phases: predominant growth of leaves in the first phase, pseudostem formation in the second, and cloves in the last two phases (beginning of rapid clove growth up to beginning of dry matter loss from blades and pseudostems, and from the last event to senescence). However, in spite of working with two planting dates, they did not establish relationships between environmental factors and growth stages. The effect of radiation was analyzed to assess a model of radiation interception and dry matter partitioning. Bulbing ratio was used to define the length of the bulb growth period.

Based on the findings mentioned above, garlic growth can be described on the basis of identifying different growth stages, assessing dry matter allocation during these phases, and analyzing the effect of environmental factors on each of these phases. However, more knowledge on yield formation in garlic can be obtained by analyzing the effects of different growing conditions during different phases, especially yield formation in relation to degree days accumulated (Bowalda, 1986). Bulbing set can be determined on the basis of changes for dry matter production and allocation during crop growth since dry matter production responds to short-term fluctuations of irradiance as photosynthesis does (Krug, 1997). Bulbing ratio may be useful to define the stage of development of the plant under field conditions (Mann, 1952; Mann and Mingués, 1958; Ledesma *et al.*, 1983, 1997; Rizzali *et al.*, 2002). However, some authors consider the use of this ratio to define the beginning of bulb growth to be controversial (Mondal *et al.*, 1986; Portela, 2004).

Leaf dry matter production and leaf expansion are also important to the analysis of crop growth (Hsiao *et al.*, 2019; Sultana *et al.*, 2022). Dry matter allocation responds to temperature and photoperiod in the white type garlic (Portela, 2004),

but information about the effects of radiation on dry matter allocation is scarce (Rahim and Fordham, 1990; 1994). The end of leaf (blade + sheaths) growth is reached after the beginning of the rapid bulb growth period (Ledesma *et al.*, 1997; Rizzali *et al.*, 2002; Portela, 2004). Loomis *et al.* (1976) (cited in Van Delden *et al.*, 2001) have determined that the decrease in foliar expansion responds to the decrease in photoassimilates caused by the increase in competition with other sinks. In garlic, allocation of dry matter to the canopy decreases because of the formation of axillary bud primordia as a new sink for assimilates (Rizzali *et al.*, 2002).

The size of seed cloves is directly related to final yield because clove size modifies leaf area of plants as a result of the amount of reserves available at the beginning of crop growth (Stahlschmidt *et al.*, 1997; Castellanos *et al.*, 2004). Large clove size increases yield and improves the fraction of bulbs suitable for ware (Burba *et al.*, 1987; Aljaro, 1992; Stahlschmidt *et al.*, 1997; Castellanos *et al.*, 2004; Jiménez-Vázquez *et al.*, 2014; Desta *et al.*, 2021). Research indicates that achieving a high percentage of marketable bulbs requires a specific clove size (Castellanos *et al.*, 2004; Portela *et al.*, 2005); specifically, for cultivars within the same ecophysiological group as the garlic used in our study, Portela *et al.* (2005) identified an optimal clove weight between 1.3 and 2.8 g. Moreover, the effect of clove weight on bulb weight distribution was analysed in relation to planting date. In the white garlic type cv. Blanco, clove weight of 2.5 g yielded a lower percentage of bulb class of 6 cm diameter (14.5%) than the clove weight of 9.8 g (63.7%) at an early planting date (Stahlschmidt *et al.*, 1997). However, this difference of nearly 50% of that bulb size turned to be only 14% at the last planting date. The change in bulb size distribution, influenced by changes in the size of the planted clove, was associated with a high ratio between harvested bulb weight and seed clove weight. To properly assess the interaction between planting date and clove size, it is necessary to analyse crop yield not only in terms of dry matter production but also in terms of bulb size distribution.

Finally, the moment of harvest determines the end of crop growth, but there is no agreement on which plant or bulb characteristics define that moment. Harvest index has been used to define the appropriate moment but also thickness and number of cataphylls of bulbs and chemical compounds (Aljaro and Cañoles, 1997; Ledesma

et al., 1997). The decline of the green leaf area and the yellowing of the plant are also important features of this phase in hardneck garlic. However, the harvest time does not always coincide with the plant's maturity stage, as it depends on the intended use of the crop. The presence of at least 40% of green leaves is preferable for fresh-market garlic, but not for garlic intended for seed.

The combined effects of planting date and clove size can modify crop growth given its exposure to different environmental conditions and the vigour of the seed clove. The effects of these agronomic factors on the garlic crop were studied during two growing cycles. In the first part of our study, dry matter production during crop growth will be analysed. This part will pay attention to the growth processes of bulbs and shoots as affected by thermal conditions.

This work aimed to gain insight into the effects of environmental factors on dry matter production in garlic grown in the southeastern region of Buenos Aires. Data were collected to quantitatively analyse dry matter production of red garlic type in response to planting date, clove size, and growing season through growth variables such as bulb dry matter production, leaf dry weight, and leaf area.

Materials and Methods

Conditions and cultural practice

The experiments were conducted in Balcarce, Buenos Aires, Argentina, during the growing seasons of 1994 (Year 1) and 1995 (Year 2). Agrometeorological data were obtained from a meteorological station located nearby the experimental site (Figure 1). In the upper 40 cm, the soil contained 4.6% organic matter, 33.5 mg kg⁻¹ P₂O₅ and 6.9 mg kg⁻¹ NO₃ in Year 1 and 4.5% organic matter, 31.6 mg kg⁻¹ P₂O₅, and 5.3 mg kg⁻¹ NO₃ in Year 2.

Seed cloves of the red garlic type cv. Colorado Mendoza (bolting or hardneck type) were stored under controlled conditions with diffuse light at 12–14 °C until the last planting date was planted. The day before planting, from the bulbs with a fresh weight above 45 g, the cloves were treated with a nematicide (fenamiphos 0.15 l/100 L H₂O) and fungicides (benomyl and mancozeb, both 0.2 kg/100 L H₂O). The cloves were hand-planted at 0.10 m between cloves within the row, and 0.75 m between rows, resulting in a plant density of 13.3

plants m^{-2} ; cloves were placed on the top of the soil and then covered by making ridges. Fertilizer was side-dressed. At planting, $92 \text{ kg ha}^{-1} \text{ P}_2\text{O}_5$ and $36 \text{ kg ha}^{-1} \text{ N}$ were applied as ammonium phosphate (18-46-0); before bulb initiation, $74 \text{ kg ha}^{-1} \text{ N}$ as urea (46-0-0) was applied. The experiment was

surrounded by two buffer rows. Weed control and cutting of the floral stalks were done by hand. No damage from pests or diseases was observed in either experimental year. Water was applied by overhead irrigation whenever required.

Treatments and experimental design

The treatments consisted of the combinations of two levels of clove size: $2.47 \pm 0.43 \text{ g}$ and $3.86 \pm 0.58 \text{ g}$ in Year 1 and $1.82 \pm 0.69 \text{ g}$ and $4.27 \pm 1.14 \text{ g}$ in Year 2, and four levels of planting date: 19 May, 8 June, 5 July and 29 July in Year 1, and 17 May, 7 June, 4 July and 26 July in Year 2. The experiments were laid out as completely randomized block designs with four replicates, with $6 \text{ m} \times 3 \text{ m}$ plots.

Measurements

The date of 50% of plants emerged was assessed by frequent counting of successive plants in the central rows. From the first week of October onwards (i.e., starting 1 month before the critical photoperiod), five periodic harvests were carried out with intervals of 15–20 days. These intermediate harvests included eight plants surrounded by guard plants. The sample was divided into bulb (bulb plus neck) and shoot (pseudostem plus leaf blades). Bulbing index (Bi; base diameter: pseudostem diameter ratio) was determined for each of 5 plants (Mann, 1952). Before drying the foliage, three representative plants were used to assess leaf area using a Li 3100 area meter (Li-Cor, Ne, USA) and their corresponding dry weights to assess specific leaf area. Fresh weights were recorded for bulbs, and dry weights were recorded for bulbs and shoots. Dry weight was determined after drying in an oven at 70°C until constant weight. Thereafter, the leaf weight ratio (LWR) was determined as the quotient between shoot (pseudostem plus leaf blade weight) and total plant weight. Dry matter concentrations for bulbs were calculated from dry and fresh weights. Final harvests took place on 09/12 in Year 1 and 14/12 in Year 2.

Data processing

Crop growth data were related to temperature accumulated during the growing season because temperature was considered the

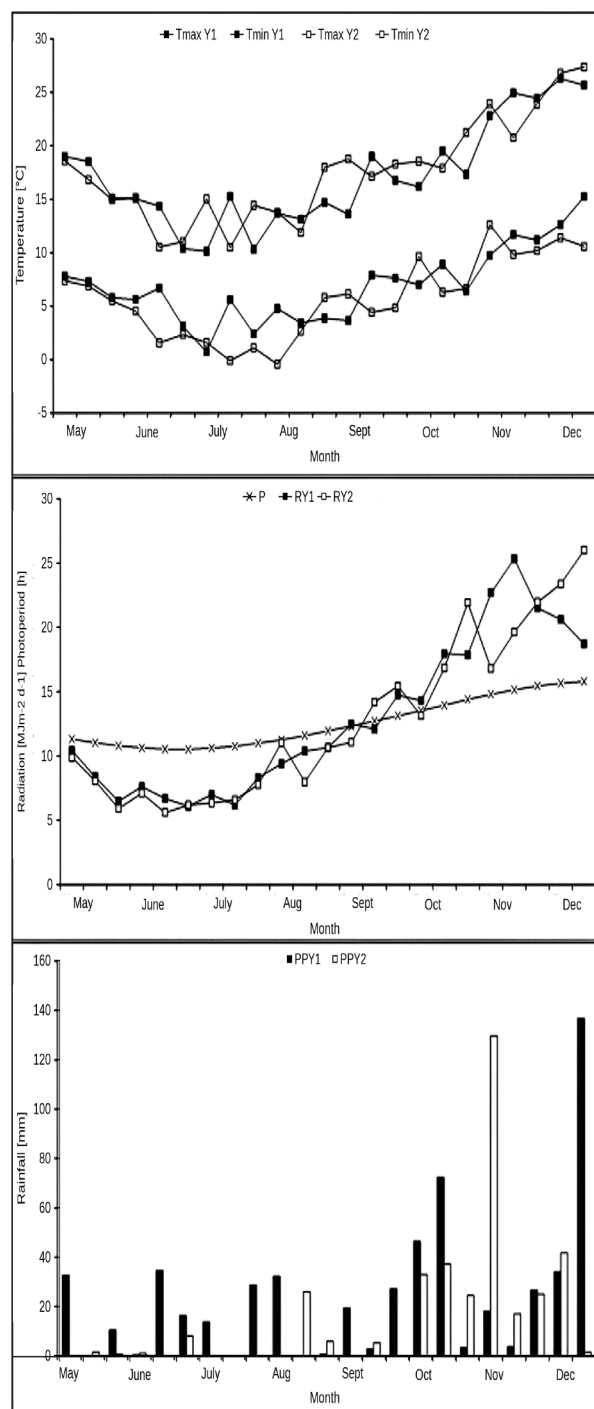


Fig. 1 Meteorological data of temperature (T , $^\circ\text{C}$), radiation intensity (R , $\text{MJ m}^{-2} \text{ d}^{-1}$), daylength (D , h) and rainfall (pp , mm) during the experiment in Balcarce, Buenos Aires Province. Data are means of 10 days from May (May), June (June), August (Aug), September (Sept), October (Oct), November (Nov) and December (Dec) for Year 1 (Y1) and Year 2 (Y2) growing cycles; rainfall is the sum of a 10-day period.

major environmental variable controlling crop performance, including growth, morphogenesis and plant quality (Bowalda, 1986; Krug, 1997). The base temperature for calculating degree days accumulated (DDA) was 0 °C (Espagnacq *et al.*, 1987). Crop growth variables, total dry matter, tuber dry and fresh weight, leaf area and dry weight were related to DDA.

Regression analysis was done with the models $y=a_1x + b$, $y=a_1x^2 + a_2x + b$, and $y=b.e^{ax}$. The highest coefficient of determination (R^2) was the selection criterion to choose the combination of explanatory factors. A linear model for factorial experiments (Model I) was used with four repetitions for the ANOVA using InfoStat (2020), considering growing season (year effect), planting date (four levels), and clove size (two levels) as fixed factors (Steel and Torrie 1985). Data were checked for normality and homogeneity of the variance. For all variables, the requirements for the statistical assumptions were met.

Results

Total dry weight

In both years, the rates from the first to the fourth PD showed similar relationships between total dry matter yield (TDMY) and degree days accumulated (DDA) (Table 1; Figure 2). The rates were slightly higher in Year 1 when the temperature was more conducive to dry matter accumulation than in Year 2, but the confidence intervals were similar. The rate of TDMY increase was not significantly modified by clove size in both growing cycles. The lowest DDA before dry matter production was for the last PD that varied between 543 °C day (in Year 1) and 556 °C day (in Year 2).

A quadratic linear relationship was found between TDMY at harvest and crop growth duration (CGD) from emergence to harvest (Figure 3). That equation showed that any

Table 1: Effects of planting date and clove size on the yield rate ($\text{kg ha}^{-1} (\text{°Cd})^{-1}$) of total dry weight, bulb fresh weight and bulb dry weight of garlic during two growth cycles (Year 1 and Year 2). Values $a_1(a;b)$ $b(a;b)$ 95% confidence interval; values $\pm$ standard error; (R^2) determination coefficient.

Planting date		Total dry weight $y = a_1x + b$ $\text{kg ha}^{-1} (\text{°Cd})^{-1}$	Bulb fresh weight $y = a_1x + b$ $\text{kg ha}^{-1} (\text{°Cd})^{-1}$	Bulb dry weight $y = a_1x + b$ $\text{kg ha}^{-1} (\text{°Cd})^{-1}$
Year 1	19/05	$a_1: 4.2 (2.5; 6.0)$ $b: -4481 (-7310; -1651) (0.95)$	$a_1: 9.3 (7.3; 11.2)$ $b: -10300 (-13495; -7106) (0.99)$	$a_1: 2.9 (1.8; 4.1)$ $b: -3651 (-5461; -1841) (0.97)$
	08/06	$a_1: 4.1 (2.5; 5.7)$ $b: -3403 (-5673; -1134) (0.95)$	$a_1: 9.0 (7.1; 10.8)$ $b: -7769 (-10323; -5016) (0.99)$	$a_1: 2.9 (1.8; 4.0)$ $b: -2872 (-4441; -1302) (0.96)$
	05/07	$a_1: 4.0 (3.2; 4.9)$ $b: -2602 (-3642; -1562) (0.99)$	$a_1: 9.2 (6.9; 11.5)$ $b: -6280 (-9123; -3437) (0.98)$	$a_1: 2.9 (1.7; 4.0)$ $b: -2296 (-3711; -3711) (0.97)$
	29/07	$a_1: 3.6 (3.1; 4.0)$ $b: -1954 (-2481; 1428) (0.99)$	$a_1: 8.1 (6.0; 10.1)$ $b: -4548 (-6772; -2325) (0.63)$	$a_1: 2.6 (1.3; 3.9)$ $b: -1781 (-3229; -333 0) (0.95)$
Year 2	17/05	$a_1: 3.6 (2.9; 4.3)$ $b: -3628 (-4748; -2507) (0.99)$	$a_1: 7.3 (5.6; 9.0)$ $b: -7368 (-10066; -4669) (0.98)$	$a_1: 2.9 (1.0; 4.8)$ $b: -3374 (-6394; -353) (0.85)$
	07/06	$a_1: 3.8 (2.8; 4.9)$ $b: -3147 (-4580; -1704) (0.98)$	$a_1: 7.2 (5.3; 9.2)$ $b: -5833 (-8520; -3146) (0.98)$	$a_1: 3.0 (0.8; 5.1)$ $b: -2827 (-5770; 115) (0.83)$
	04/07	$a_1: 3.4 (2.5; 4.4)$ $b: -2240 (-3397; -1083) (0.98)$	$a_1: 6.8 (4.9; 8.8)$ $b: -4319 (-6639; -1999) (0.98)$	$a_1: 2.7 (0.9; 4.6)$ $b: -2115 (-4398; 168) (0.84)$
	26/07	$a_1: 3.1 (1.9; 4.3)$ $b: -1724 (-3048; -401) (0.96)$	$a_1: 5.7 (4.0; 7.4)$ $b: -2877 (-4748; -1007) (0.97)$	$a_1: 2.5 (0.6; 4.3)$ $b: -1634 (-3726; 457) (0.81)$
Clove size				
Year 1	Large	$a_1: 4.3 (3.1; 5.4)$ $b: -3240 (-4825; -1656) (0.98)$	$a_1: 9.3 (7.2; 11.4)$ $b: -7433 (-10242; -4623) (0.98)$	$a_1: 3.0 (1.8; 4.1)$ $b: -2741 (-4306; -1176) (0.96)$
	Small	$a_1: 3.7 (2.7; 4.7)$ $b: -2885 (-4212; -1557) (0.98)$	$a_1: 8.5 (6.4; 10.5)$ $b: -6869 (-9574; -4163) (0.98)$	$a_1: 2.7 (1.6; 3.8)$ $b: -2504 (-3931; -1077) (0.96)$
Year 2	Large	$a_1: 3.8 (2.9; 4.6)$ $b: -2772 (-3865; -1679) (0.98)$	$a_1: 7.4 (5.4; 9.3)$ $b: -5310 (-7904; -2716) (0.98)$	$a_1: 3.0 (0.9; 5.0)$ $b: -2615 (-5298; 69) (0.88)$
	Small	$a_1: 3.2 (2.1; 4.3)$ $b: -2522 (-3989; -1056) (0.97)$	$a_1: 6.1 (4.5; 7.8)$ $b: -4679 (-6853; -2504) (0.98)$	$a_1: 2.6 (0.7; 4.4)$ $b: -2302 (-4775; 172) (0.86)$

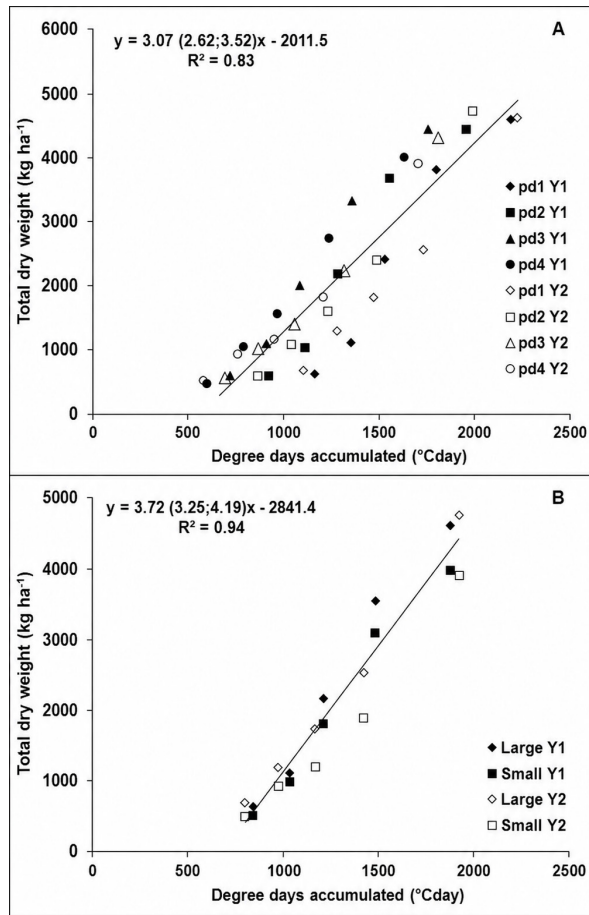


Figure 2: Total dry weight (kg ha^{-1}) of garlic for two growing cycles, Year 1 (Y1) and Year 2 (Y2), against degree days accumulated ($^{\circ}\text{C day}$). A: four planting dates (pd) at each year (Y1 and Y2) and B: two clove sizes (large and small) at each year (Y1 and Y2). $y = a_1$ (95% confidence interval) $x + b$.

advancement of PD, and therefore a longer CGD, would not contribute significantly to an increase in TDMY.

Bulb growth

Bulb growth as defined by bulbing index (Bi) started later in Year 1 than in Year 2 (Table 2). There were differences between years for Bi, which was significantly advanced in the second growing cycle. $\text{Bi}=2$ was reached between 14.7 h (Year 2) and 15.2 h (Year 1) photoperiod in both years. The effects of PD and year interacted for Bi, as well as CS interacted at the second sampling date; analysing these effects in each year also allowed for the evaluation of the CS effect. Regarding the first growth cycle, at the two first sampling dates, Bi was similar among PDs and CSs, but at the third sampling date, Bi increased as PD was further delayed. In Year 2, at the first two sampling dates, the trend of advancing bulbing as PD was

Table 2: Bulbing index (Bi) (bulb diameter: pseudostem diameter ratio) of garlic for years 1 and 2 at three times of the growing cycle (date and photoperiod), four planting dates (PD) and two clove sizes (CS, large and small). Means ($n=5$); ANOVA Fisher - Snedecor test: * ($p<0.05$), ** ($P<0.01$), ns (not significant).

			Samples		
			20 Oct (14.1 h)	03 Nov (14.7 h)	18 Nov (15.2 h)
Year	PD	CS	Bi		
Year 1	19/05	Large	1.52	1.71	2.26
		Small	1.52	1.71	2.18
	08/06	Large	1.49	1.64	2.62
		Small	1.52	1.63	2.47
	05/07	Large	1.51	1.75	2.42
		Small	1.46	1.67	2.63
	29/07	Large	1.47	1.74	2.53
		Small	1.54	1.69	2.56
	Mean		1.50	1.69	2.46
				20 Oct (14.1 h)	02 Nov (14.7 h)
Year 2	17/05	Large	1.55	1.72	2.73
		Small	1.58	1.75	2.77
	07/06	Large	1.53	1.70	2.55
		Small	1.62	1.88	2.78
	07/06	Large	1.77	1.90	2.68
		Small	1.75	1.89	2.80
	26/07	Large	1.67	1.85	2.61
		Small	1.67	1.95	2.75
	Mean		1.64	1.83	2.71
	Analysis of Variance				
Planting date (pd)			**	**	**
Clove size (cs)			ns	ns	ns
Year (y)			**	**	**
pd × y			**	**	**
cs x y		ns		**	ns

delayed, was evident; meanwhile, the CS effects were apparent at the second and third sampling dates when the smallest CS showed higher Bi. A combined effect of both factors modified Bi before and after Bi rose to 2.

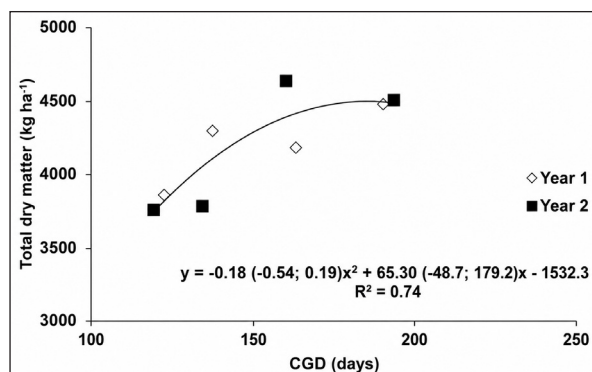


Figure 3: Total dry matter production at harvest – crop growing duration (CGD) relationship of garlic for two growing cycles Year 1 and Year 2. Data are means of two clove sizes, $y = a_2$ (95% confidence interval) $x^2 + a_1$ (95% confidence interval) $x + b$.

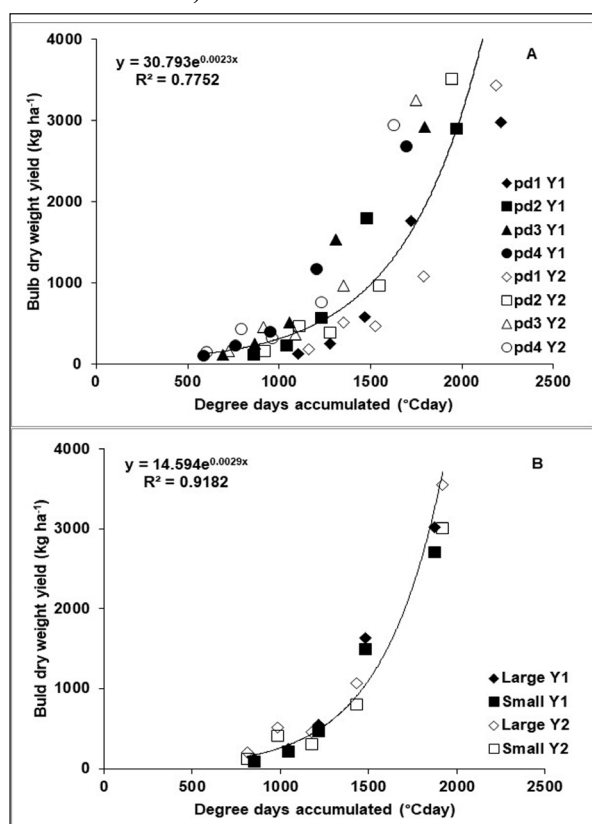


Figure 4: Bulb dry weight (kg ha^{-1}) of garlic for two growing cycles, Year 1 (Y1) and Year 2 (Y2), against degree days accumulated ($^{\circ}\text{C day}$). A: four planting dates (pd) at each year (Y1 and Y2) and B: two clove sizes (large and small) at each year (Y1 and Y2).

Bulb growth against DDA was analysed using a linear regression for bulb dry matter yield (BDMY) (Figure 4; Table 1). The rate of increase of BDMY was similar for all four PDs, and no differences were found between growing cycles (Table 1). CS had no significant effects on BDMY rate; the results were similar, and the large CS did not have the highest rate in both years. However, the rate tended to decrease from the first to the last

PD in both growing cycles. A higher coefficient of determination was found in Year 1 because of less variation in the initial dry weight of bulbs; unlike in Year 2, from the onset of bulb growth, dry matter accumulation was steadily increasing (Figure 4). This different performance of the dynamics of dry matter allocation in garlic organs may be partially explained in terms of Bi behaviour (Table 2).

Bulb fresh matter yield (BFMY) was analysed using a linear regression that had a higher coefficient of determination than that for BDMY in both years. For BFMY, there was a trend to a lower weight accumulated over DDA as planting date was delayed in both years, being slightly higher in Year 1 in spite of no significant differences among PDs, between CSs and between years (Table 1; Figure 5). Regarding the CS effect, no differences in the performance of the rates of BFMY increase

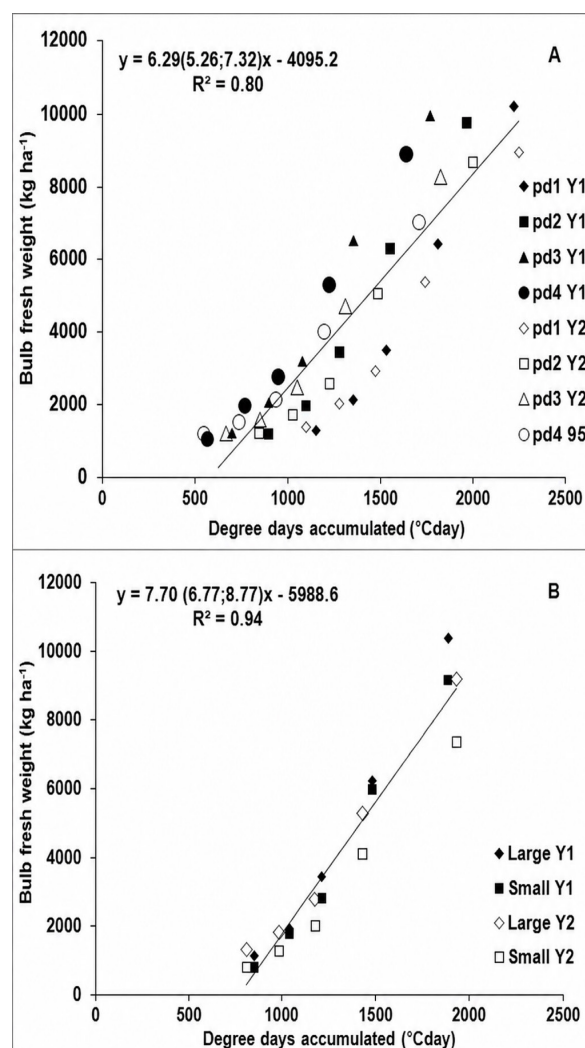


Figure 5: Bulb fresh weight (kg ha^{-1}) of garlic for two growing cycles, Year 1 (Y1) and Year 2 (Y2), against degree days accumulated ($^{\circ}\text{C day}$). A: four planting dates (pd) at each year (Y1 and Y2) and B: two clove sizes (large and small) at each year (Y1 and Y2). $y = a_1$ (95% confidence interval) $x + b$.

were evident in both growing cycles.

Leaf growth

Maximum values of dry matter accumulated per hectare in leaves (blade + sheath) (LDM) were reached close to 18 November of Year 1 and 17 November of Year 2, and they tended to be higher at earlier PD in both years (Table 3). At the end

of crop growth, LDM decreased as a result of dry mass redistribution from the leaves to the bulbs in association with leaf senescence.

A second-degree polynomial was used to analyse leaf dry matter yield (LDMY) response to compare the coefficients of each PD and CS. In both years, the linear coefficient rate of LDMY against DDA decreased from the first to the last PD but differences were not significant (Table 4).

Table 3: Leaf area per plant and leaf dry weight per plant of garlic of the three last samples of the growing cycle (03 Nov, 18 Nov and 10 Dec in Year 1; 02 Nov, 17 Nov and 15 Dec in Year 2) for four planting dates (PD) and two clove sizes (CS). Means (n=4); ANOVA Fisher - Snedecor test : *(p<0.05), ** (p<0.01), ns (not significant).

			Leaf area (cm ² plant ⁻¹)			Leaf dry weight (g plant ⁻¹)		
			03 Nov	18 Nov	10 Dec	03 Nov	18 Nov	10 Dec
Year	PD	CS						
Year 1	19 May	Large	728.1	787.3	649.4	14.7	14.9	10.7
		Small	629.8	681.4	541.8	11.0	11.7	8.0
	08 Jun	Large	688.6	756.2	603.5	12.6	12.6	9.2
		Small	605.2	672.7	565.7	10.1	12.4	7.9
	05 July	Large	663.8	758.0	607.3	11.1	13.2	9.7
		Small	563.1	648.1	544.1	8.7	9.4	7.5
	29 Jul	Large	610.0	741.5	604.8	9.4	11.7	9.6
		Small	532.4	604.0	504.6	7.1	9.8	6.5
	Mean		627.6	706.2	577.	10.6	11.9	8.6
				02 Nov	17 Nov	15 Dec	02 Nov	17 Nov
Year 2	17/05	Large	453.4	557.1	263.3	9.7	10.4	6.5
		Small	369.7	435.7	242.9	7.6	8.6	5.7
	07/06	Large	378.4	569.9	369.1	8.8	11.4	7.8
		Small	279.8	426.2	278.9	5.6	7.9	5.8
	07/06	Large	344.5	482.4	272.9	8.3	9.1	6.2
		Small	263.2	364.9	208.0	4.9	7.2	4.7
	26/07	Large	329.4	434.6	256.3	6.7	8.4	5.1
		Small	225.1	344.9	228.4	3.2	5.7	4.2
	Mean		330.4	451.9	264.9	6.8	8.6	5.8
	Analysis of Variance							
Planting date (pd)			**	**	**	**	**	**
Clove size (cs)			**	**	**	**	**	**
Year (y)			**	**	**	**	**	**
pd × y			ns	ns	ns	ns	**	**
cs x y			ns	ns	ns	ns	**	*

Table 4: Effects of planting date and clove size on rate of change with degree day accumulated ($^{\circ}\text{Cd}$) of leaf dry matter yield ($\text{kg ha}^{-1} (^{\circ}\text{Cd})^{-1}$) and leaf area per plant ($\text{cm}^2 \text{ plant}^{-1} (^{\circ}\text{Cd})^{-1}$) of garlic during two growth cycles (Year 1 and Year 2). Values (a; b) 95% confidence interval; values $\pm$ standard error (R^2) determination coefficient.

Planting date		Leaf dry weight		Leaf area	
		$y = a_2x^2 + a_1x + b$		$y = a_2x^2 + a_1x + b$	
		$\text{kg ha}^{-1} (\text{°Cd})^{-1}$	$\text{kg ha}^{-1} (\text{°Cd})^{-1}$	$\text{cm}^2 \text{ plant}^{-1} (\text{°Cd})^{-1}$	$\text{cm}^2 \text{ plant}^{-1} (\text{°Cd})^{-1}$
		a_1	$a_2 (*10^{-3})$	a_1	$a_2 (*10^{-3})$
Year 1	19/05	12.7 (−3.5; 28.9)	−3.4 ± 1.0 (0.92)	4.3 (1.7; 6.9)	−1.1 ± 0.2 (0.98)
	08/06	9.3 (−1.8; 20.5)	−2.8 ± 0.9 (0.93)	3.7 (1.9; 5.4)	−1.1 ± 0.1 (0.98)
	05/07	7.1 (1.0; 13.2)	−2.3 ± 0.6 (0.97)	3.1 (2.6; 3.6)	−1.1 ± 0.04 (0.99)
	29/07	5.5 (1.9; 9.0)	−2.0 ± 0.3 (0.98)	3.0 (2.2; 3.7)	−1.1 ± 0.07 (0.99)
Year 2	17/05	7.2 (1.4; 12.9)	−1.9 ± 0.4 (0.96)	2.7 (−0.1; 5.5)	−0.8 ± 0.2 (0.90)
	07/06	5.9 (−0.5; 12.0)	−1.7 ± 0.5 (0.95)	2.1 (−0.1; 4.8)	−0.7 ± 0.2 (0.88)
	04/07	4.3 (−0.1; 8.7)	−1.4 ± 0.4 (0.95)	1.7 (−0.4; 3.8)	−0.6 ± 0.2 (0.88)
	26/07	3.0 (−0.3; 6.3)	−1.0 ± 0.3 (0.95)	1.4 (−0.3; 3.2)	−0.6 ± 0.2 (0.87)
Clove size					
Year 1	Large	9.1 (−0.6; 18.8)	−2.8 ± 0.8 (0.95)	3.7 (2.6; 4.8)	−1.2 ± 0.09 (0.99)
	Small	7.7 (1.1; 14.3)	−2.4 ± 0.5 (0.96)	3.3 (2.5; 4.2)	−1.1 ± 0.07 (0.99)
Year 2	Large	6.0 (0.1; 11.9)	−1.9 ± 0.5 (0.95)	2.2 (−0.4; 4.7)	−0.7 ± 0.2 (0.88)
	Small	3.9 (0.3; 7.5)	−1.2 ± 0.3 (0.96)	1.7 (−0.3; 3.7)	−0.6 ± 0.2 (0.89)

In relation to CS, there were no differences between years. The quadratic coefficient tended to decrease from the first to the last PD in both growing cycles, and they were nearly halved in the second growing season. Analysing leaf dry weight (LDM), the three last sampling dates during crop growth (Table 3) showed that there were significant differences among PDs, CSs and years and PD and CS interacted with year at the last two sampling dates. LDM decreased as a consequence of delaying planting time and reducing clove seed size during both growing cycles. At the fourth sampling date (18 November Year 1; 17 November Year 2), LDM reached the maximum values for most PDs in both years, with Year 1 exceeding Year 2.

We also analysed the leaf weight ratio (LWR = leaf weight / total plant weight) during the crop growing cycle for both years. At the same sampling date for LDM the ratio showed higher values in Year 1 than in Year 2 for four out of

the five sampling dates. These values were 0.69, 0.72, 0.72, 0.51 and 0.32 g.g^{-1} from the first to the last sampling dates in Year 1, and 0.56, 0.44, 0.69, 0.54 and 0.23 g.g^{-1} from the first to the last sampling dates in Year 2. These differences led to more pronounced dry matter partitioning to cloves in Year 2.

In relation to leaf (blade) area (LA), early PDs tended to have higher rates (linear coefficient) in both years than the late PDs, and there were no differences between CSs in either year (Table 4). The quadratic coefficients showed similar values among PDs at each growing cycle, being slightly less negative in the second one. As was pointed out for LDM, at the fourth sampling date (18 November Year 1; 17 November Year 2) maximum values were reached and differences between years were apparent, and so were the differences among PDs and CSs, with LA being larger in Year 1 (Table 3). LA decreased as the clove seed size diminished for both growing cycles and when the

planting time was delayed in Year 2; in Year 1, PD did not affect LA in any of the three analysed sampling dates.

There was a close association between leaf area (LA) per plant and BFMY at two harvest dates in both growing cycles. This relation was stronger for the 3rd sampling than at harvest and was affected by both agronomic factors (Table 5). The LA - BDMY relationship was analysed too. This relation was significant at the 3rd sampling and tended to be absent at the final harvest.

Discussion

Bulbing and leaf growth

The year effect was evident: cooler conditions in Year 2 advanced bulb growth and compensated for the photoperiod requirement (Figure 1). Moreover, the Bi was sensitive enough to detect PD and CS differences in Year 2. There is evidence that low temperature between planting and bulb initiation advances the beginning of bulb growth and may

Table 5: Correlation coefficients (r) between bulb dry or fresh weight yield and leaf area at two times of the growing cycle of garlic. For planting dates, data were pooled over two clove sizes and two years. For clove size data were pooled over four planting dates and two years. Pearson correlation: * (p<0.05), ** (p<0.01); sample size n=16.

Planting date		Bulb dry weight	Bulb fresh weight
		r	r
At beginning of bulb growth	1st	0.75**	0.81**
	2nd	0.77**	0.78**
	3rd	0.71**	0.77**
	4th	0.73**	0.70**
At harvest	1st	-0.49*	0.62*
	2nd	-0.42	0.66**
	3rd	-0.10	0.59*
	4th	0.03	0.79*
Clove size			
At beginning of bulb growth	Large	0.66**	0.73**
	Small	0.81**	0.81**
At harvest	Large	-0.37*	0.54**
	Small	-0.15	0.70**

compensate for the requirement of photoperiod (Mann and Mingués, 1958; Ledesma *et al.*, 1983; Brewster, 1994). It was found that bolting-type garlic plants (or hardnecks) needed 30–40 days under 0–4 °C or 50–60 days under 10 °C at the four-leaf stage for vernalization (Atif *et al.*, 2020). Moreover, it has been shown for *Allium* species, including garlic, that bulb initiation and development are influenced by carbohydrates as well (Atif *et al.*, 2020). Soil temperature could play a role because the months previous to the analysed period had lower minimum temperature in Year 2 than in Year 1 (Figure 1) (Rahim and Fordham, 1990).

Thermal conditions at the beginning of bulbing were close to optimal for bulb growth; mean temperature was over 18 °C during both years (Krug, 1997). For the same garlic type, in the Andean region of Famatina, it was found that bulbing occurred at 12.6/12.7 h of photoperiod under 19/20 °C, nearly 10 days before and after Bi=2 in two growing cycles (Saluzzo *et al.*, 2009). Hence, it seems that the bulbing index (base diameter: pseudostem diameter ratio) could enable the identification of the same phenological moment in both growing regions. Considering that a more intense low temperature induction may require a lower threshold of photoperiod, it can be surmised that the cold requirement has been satisfied earlier in Famatina than in Balcarce. For the coldest months, June, July, and August temperature ranged between –0.4 and +3.2 °C and between 0.1 and 2.4 °C in two years in Famatina; in Balcarce, they ranged between 3.0 and 5.2 °C and between 0.9 and 2.8 °C in Year 1 and 2, respectively. Moreover, thermal amplitude (thermoperiod) and radiation intensity may have played a role in dry matter production as well. Therefore, temperature is the main factor to drive bulbing in the red type garlic, and the threshold of the photoperiod for this garlic type mainly depends on the interaction with temperature.

Rates of bulb dry weight increases (in kg ha⁻¹ (°C day)⁻¹) were similar to those found for white garlic type by Portela (2004). This author found differences between years, and higher rates were associated to higher temperature during the fastest growth bulb period. The red garlic type used in our experiment showed similar behaviour in terms of response to temperature in spite of a longer photoperiod to induce bulbing (white type required nearly 12.5 h vs nearly 14.5 h for the red

type). Garlic bulb development is induced by the day length and under an ideal growth temperature, which may vary among garlic cultivars (Atif, 2019) and may be affected by the effect of planting date in interaction with the cultivar effect (Sultana *et al.*, 2022).

Concerning the onset of bulbing, the process of dry matter allocation to the bulbs was more advanced in Year 2 than in Year 1 and was affected by PD. Changes at the beginning of dry matter allocation in bulbs are the result of temperature and daylength interactions affecting clove initials and floral stalk differentiation (Brewster, 1994). This is because of the overlapping thermal requirements of axillary buds (cloves) and floral stalk differentiation at the beginning of the spring months. Then long photoperiod stimulates inflorescence differentiation because of lower thermal requirement for their development than for bulbing, while sufficiently low temperature prevents bulbing. Moreover, under optimal growth conditions, bolting in garlic has no detrimental effect on bulbing and yield; competition for assimilates is important but not so much (Wu *et al.*, 2016). Therefore, the red garlic type showed a sharp increase in dry matter allocation to the floral stalk under less favourable soil fertility (Saluzzo and Villafaña, 2008).

In general, PD and Year effects interacted on Bi under the current experimental conditions. Differences between years and, only in Year 2, differences among PDs as well as CSs for Bi at the 2nd sampling date (02 November) were found. That moment was close and before the onset of bulb growth and highlighted the fact that in Year 2, the bulbing index exhibited higher values at later planting and with smaller clove seed size. Concerning the year effect on bulb induction, there is evidence that maturity and bulb formation may be delayed if plants receive little exposure to low temperature (Man and Minges, 1958; Ledesma *et al.*, 1983, Wu *et al.*, 2016); this is in line with the difference between years in our experiments. The thermal stimulus is crucial for bulbing, and it was effective on plants of different sizes caused by the effects of the agronomic factors PD and CS. It is suggested that the effectiveness of that stimulus is associated with carbohydrate production, and it is probable that small plants achieved critical carbohydrate concentration and bulbing stimulus earlier than large plants, as was shown for garlic (Kahane *et al.*, 1997) and for onion (Mettananda

and Fordham, 1999). Added to that effect, plant size differences caused by CS at each PD (more stressed in Year 2) could be associated with plant age in determining the response to the long day stimulus as was shown for garlic (Atif *et al.*, 2020) and for onion, too (Sobeih and Wright, 1986). Moreover, critical carbohydrate concentration and plant age factors may be associated in the first two samples in Year 2 where a higher proportion of dry matter allocated to bulb would have increased as PD delays along with small CS.

LA and bulb weight are closely related (Stahlschmidt *et al.*, 1997; Olivera *et al.*, 2018). However, when bulb weight was defined in terms of either dry or fresh weight, LA / BFMY relation was more responsive than LA / BDMY. A similar result was found working with different levels of N fertilization with a red garlic type (Saluzzo, 2003). Changes of ambient conditions during bulb growth modified more intensely LA / BDMY when both growing cycles were pooled and may alter dry matter allocation in bulbs without modifying LA growth as was shown in other vegetable crops (Krug, 1997). Environmental factors, radiation, and temperature, may account for this differential response of plant development and growth. Moreover, a high LA at harvest does not guarantee a high BDMY, because BDMY is more sensitive to short-term changes of environmental factors as it is more connected to the photosynthesis process. Also, crops from small cloves were less sensitive to ambient factors, and BFMY of those crops were more closely correlated to LA as was shown for red garlic type by Stahlschmidt *et al.*, (1997). Sanitary controls, irrigation, and soil nutrient management are vital to preserve a healthy leaf growth and, consequently, to achieve adequate bulb yield and quality (Khokhar, 2023).

Leaf dry matter and leaf area

Leaf dry matter per plant decreased as planting date was delayed in both growing cycles (Year 1 and 2) as was also shown by other authors (Sultana *et al.*, 2022). Taking into account the linear relations to explain LDMY behaviour (Table 4), the quadratic coefficient was nearly halved in Year 2 compared with Year 1. This higher rate at the last growing phase in Year 1 may be related not only to the end of leaf dry matter accumulation but also to a stronger competition for assimilates with bulb growth that became the dominant sink. Moreover,

BDMY rate was similar in both years, but the LDMY rate tended to be higher in Year 1 as well as LA rate along with higher LDM and LA per plant, because of warmer conditions. The behaviour of these growth traits along with LWR contributed to explaining a higher specific leaf area in the first growing cycle with no detrimental effect on bulb growth (Figure 6). Finally, LA did not match LDM performance as related to DDA because it was less sensitive to changes in environmental conditions induced by different PDs. It was shown that LA was modified significantly by planting time only in Year 2. Differences between years in this experiment showed a steadier allocation of dry matter to bulbs because of a more conducive temperature for plant growth, along with photoperiod increase in Year 1 than in Year 2. In this last growing cycle, this early competition for assimilates to bulb growth negatively affected leaf growth. Figure 6 shows LA and LDM (sheath + blade) linear relationship, in which the linear regression coefficient means specific leaf area ($\text{cm}^2 \text{g}^{-1}$ leaf) and was significantly lower in the second growing cycle, where expansion and dry matter allocation to aerial organs were negatively affected by the stronger sink of bulbs. That is because of cooler environmental conditions in Year 2 that brought about an earlier formation of axillary bud primordia as new sinks for assimilates (Rizzali *et al.*, 2002). In terms of initial soil nutrients availability, no shortage may be considered because this year organic matter, soil nitrates and phosphates were similar to those of Year 1.

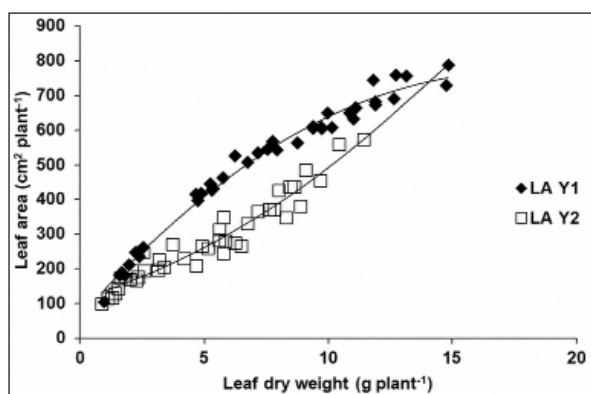


Figure 6: Relation between leaf area (LA) and leaf dry weight of garlic from the beginning up to the harvest time for four planting dates and two clove sizes in two years, Year 1 (Y1) and Year 2 (Y2). Year 1: $LA = -2.33 (-2.84; -1.81) x^2 + 80.89 (72.94; 88.85) x + 62.10$; $R^2: 0.98$, $n=40$. Year 2: $LA = 1.68 (0.38; 2.61) x^2 + 21.03 (6.21; 35.86) x + 113.4$; $R^2: 0.93$,

Conclusions

The combined effects of the agronomic factors planting date and clove size modify the garlic red type growth processes of bulbs and shoots that are mainly affected by thermal conditions in the southeast region of Buenos Aires province. Leaf growth and leaf area should be considered as the most important crop characteristics because they are closely related to the final garlic yield, highlighting the importance of optimizing planting date and seed clove size. Early planting date and large seed clove size will promote a high bulb quality at harvest, given the fact that the rate of bulb dry weight and fresh weight increases tend to decrease, as well as leaf development and expansion when planting dates are delayed. Bulbing induction was accelerated under cooler conditions of the growing cycle and by using small seed clove size in late plantings; bulbing index (bulb swelling) was an integrating variable that reflects the level of development in a specific environment.

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