

# Critical period of weed interference on total polyphenol content in quinoa

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## ABSTRACT

There is limited information about the critical period of weed interference (CPWI) in quinoa (*Chenopodium quinoa* Willd.) and the effect produced by the weed-crop interaction in secondary metabolite accumulation. The objective of the present study was to determine the CPWI and its effect on total polyphenol content in quinoa. The experiments were conducted during two consecutive seasons using a randomized complete block design with 16 treatments consisting of 8 weed growth periods and 8 weed-free growth periods in which weed population and biomass were evaluated; productive parameters, yield components, and total polyphenols were determined in the quinoa crop. Grain number per plant affected yield because of weed interference ( $P < 0.05$ ), which decreased from 4312 to 162 grains plant<sup>-1</sup> in weed growth periods and increased from 181 to 5110 grains plant<sup>-1</sup> in weed-free growth periods. Total polyphenol content was affected by stress from weed interference ( $P < 0.05$ ), which increased from 2.2 gallic acid equivalents (GAE) g<sup>-1</sup> to 3.6 mg GAE g<sup>-1</sup> in weed growth periods and decreased from 3.6 GAE g<sup>-1</sup> to 1.9 mg GAE g<sup>-1</sup> in weed-free growth periods, while the population remained constant ( $P > 0.05$ ). The CPWI was determined between the phenological stages of two true leaves to flowering; therefore, the quinoa crop must remain weed-free between these two phenological stages to rule out production losses greater than 5%.

**Key words:** *Chenopodium quinoa*, critical period of weed interference, stress from weed interference, total polyphenols, weed control in quinoa.

## INTRODUCTION

Quinoa (*Chenopodium quinoa* Willd.) is a pseudocereal belonging to the *Amaranthaceae* family with high nutritional value originating from the Andes region. It is grown in several countries of South America where the cultivated area has increased in the last 20 years and is destined to food for humans (Abugoch, 2009). The quinoa grain also contains high amounts of protein and exhibits an optimum content of amino acids, minerals, vitamins, polyphenols, and flavonoids (Abugoch, 2009; Repo-Carrasco-Valencia et al., 2010). Important amounts of bioactive components such as polyphenols (Alvarez-Jubete et al., 2010), mainly phenolic acids such as caffeic, ferulic, *p*-coumaric, *p*-hydroxybenzoic, vinyl, gallic, cinnamic acids (Pasko et al., 2008; Repo-Carrasco-Valencia et al., 2010), and flavonoids have also been found in its grains. All these bioactive compounds have been reported to provide benefits for human health (Dini et al., 2010).

Although quinoa is a traditionally low-yield crop in the regions of origin, increased demand for its abovementioned benefits has led to increased production. Among the pending problems to be studied are those related to weeds because production decreases as density and duration of weed interference increase. Another factor that interferes with yield is the relationship between weed emergence and the pressure exerted on the crop (Fahad et al., 2014). Yield losses caused

by ineffective weed control are usually higher than losses caused by pests and diseases (Oerke, 2006). In this way weeds are a limiting factor in the quinoa crop; they directly affect yield because they compete for factors such as water, nutrients, and light. The intensity and duration of weed interference are factors that determine the extent of yield losses (Swanton et al., 2015).

The critical period of weed interference (CPWI) can be defined as the stage in the growth cycle of any crop during which weeds must be controlled to prevent unrecoverable yield losses. Knowledge of crop CPWI also contributes to minimize yield losses that this crop can exhibit due to weed infestation (Safdar et al., 2016); determining CPWI is an indispensable tool to propose effective weed management strategies in any crop production system (Tursun et al., 2016). Information about CPWI in quinoa and its effect on yield is very limited; therefore, findings in other crops encourage research on the effect of weeds on quinoa and define when weed management is more efficient. However, weed interference not only affects crop yield but also alters the amount of plant secondary metabolites, which accomplish important functions within the plants (Olivoto et al., 2016). Provoked stress in the plants also increases polyphenol concentration in the tissues but reduces biomass production (De Abreu and Mazzafera, 2005).

Manual weed control methods in quinoa are an important factor that directly interferes with yield; the effect of weed-crop interaction on total polyphenol content in the grain such as stress indicators are still unknown. Therefore, the objective of the present study was to determine the CPWI in quinoa and its effect on total polyphenol content.

## MATERIALS AND METHODS

### Plant material and experimental design

The experiments were conducted during two consecutive seasons in the El Nogal Experimental Station (36°34' S and 74°06' W) in Ñuble Region, Chile. A randomized complete block experimental design was used with four replicates. The experimental unit was 3 × 2 m and consisted of four rows of quinoa 'Regalona' plants with 0.5 m spacing. A modified version of the methodology proposed by Karkanis et al. (2012) was used; each block contained 16 treatments at 0, 15, 30, 45, 60, 75, 90, and 105 d after emergence consisting of 8 weed growth periods and 8 weed-free growth periods (Table 1). This allowed determining the beginning and the end of the period needed to eliminate weeds.

### Agronomic management

The soil was prepared using one pass with a moldboard plow, two passes with a disc harrow plow, and two passes with a vibro-cultivator in each season. Continuous manual sowing was performed in mid-October at a 12 kg ha<sup>-1</sup> seeding rate and 0.5 m row spacing. The soil at the experimental site exhibited 6.8 neutral pH, 8.6 mg kg<sup>-1</sup> available N, 6 mg P kg<sup>-1</sup>, and 626.0 mg K kg<sup>-1</sup> according to soil analysis; therefore, fertilization was uniformly applied as 100 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> and 50 kg K<sub>2</sub>O ha<sup>-1</sup> before sowing together with 160 kg N ha<sup>-1</sup> of which 50% was applied at the 4 true leaf stage and 50% at the beginning of branching. Furrow irrigation was used to facilitate homogeneous water flow, and mechanical/manual weed removal was carried out with a hand hoe in accordance with the treatments at biweekly intervals.

### Determination of critical period of weed interference (CPWI)

Quinoa population and growth were evaluated at 0, 15, 30, 45, 60, 75, 90, and 105 d after emergence. Measurements were taken in two linear meters in the two central rows of each plot; plant height (PH) was measured from the root collar

**Table 1. Description of treatments in weed and weed-free growth periods in the 2015-2016 and 2016-2017 seasons.**

Treatment	Weed growth periods	Treatment	Weed-free growth periods
1	0 d with weeds	9	0 d weed-free
2	15 d with weeds	10	15 d weed-free
3	30 d with weeds	11	30 d weed-free
4	45 d with weeds	12	45 d weed-free
5	60 d with weeds	13	60 d weed-free
6	75 d with weeds	14	75 d weed-free
7	90 d with weeds	15	90 d weed-free
8	105 d with weeds	16	105 d weed-free

to the apex of the panicle in the first seven plants from the selected section. Each treatment was associated with the crop phenological stages proposed by Mujica and Canahua (1989). Likewise, yield per unit area ( $\text{kg ha}^{-1}$ ) was determined from the two linear meters of the two central rows of each plot. To estimate grain number per plant and biomass per plant (g), the mean of the seven first plants in the two linear meters of the two central rows of each plot was considered. The 1000 grains weight (g) variable was determined by recording the weight of 1000 quinoa seeds without considering the perigonium, and this was accomplished by separating the grains from the chaff.

### **Determination of total polyphenols in quinoa seeds**

Quinoa samples (20 g) from each of the 16 treatments that included both weed and weed-free growth periods were ground in a grinder (1093, Cyclotec, Barcelona, Spain). Extracts were prepared according to a modified version of the method described by Fischer et al. (2013). The supernatant was filtered, placed in amber glass jars, and stored at 4 °C until analyzed. A modified version of the method described by Miranda et al. (2010) was used to determine total polyphenols. Results were expressed as mg gallic acid equivalents (GAE)  $\text{g}^{-1}$  and all measurements were performed in triplicate.

### **Weed evaluations**

Quinoa weed population and DM were evaluated by sampling according to a modified version of the method proposed by Stagnari and Pisante (2011); a quadrant with  $0.5 \times 0.5$  m was traced 1 m inside between two central rows of each experimental unit and weed population and DM were extracted with weed growth periods for weeding at 0, 15, 30, 45, 60, 75, 90, and 105 d after emergence. Weeds were cut at soil level, placed in paper bags, and oven-dried (FP 115, Binder, Tuttlingen, Germany) at 65 °C for 72 h.

### **Statistical analysis**

Data was subjected to ANOVA at  $P < 0.05$  after verifying the assumptions of normal distribution and homogeneity of variances. These assumptions were not fulfilled in yield data (data not shown and necessary to calculate the CPWI) and transformations were performed with the  $\ln(x+1)$  function. Means were compared by the LSD test at the 95% significance level.

The CPWI was determined by non-linear regressions adjusted to the Gompertz and simple logistic models. The Gompertz equation was used to model the effect of weed growth periods on relative grain yield, while the logistic equation was used to model the effect of weed-free growth periods on relative grain yield (Singh et al., 2014).

The Gompertz equation  $Y = a * \exp(-b * \exp(-X_0 * X))$  expresses  $Y$  as yield,  $a$  the asymptote,  $b$  the exponential rate,  $X_0$  the inflection point, and  $X$  days after emergence. On the other hand, the logistic equation  $Y = a / (1 + b * \exp(-X_0 * X))$  expresses  $Y$  as yield,  $a$  the lowest asymptote,  $b$  yield rate reduction measurement,  $X_0$  duration of weed interference when maximum yield could be reduced by half over 'a', and  $X$  days after emergence.

The statistical analysis and the curves to estimate the critical period of weed interference were performed with the INFostat statistical program (Di Rienzo et al., 2017). To compare the results of the variables between the first and second season, a combined experiment analysis was performed in which the interaction between year and treatment for all the variables was nonsignificant ( $P > 0.05$ ); therefore, data were grouped and analyzed together.

## **RESULTS AND DISCUSSION**

### **Weed population and biomass**

Weed density was adjusted to a quadratic polynomial model related to days after emergence (DAE) with a significant ( $P < 0.05$ ) coefficient of determination and a highly significant ( $R^2 = 0.81$ ) relationship. The lowest weed density (0 plants  $\text{m}^{-2}$ ) was observed in the treatments that were weed-free during the whole experiment (105 d weed-free and 0 d with weeds). After 60 DAE, weed density reached its maximum value and decreased until 105 DAE (Figure 1), while weed DM at this point continued to increase (Figure 2). The decrease in density could be caused by intra- and interspecific competition in which the environmental load capacity was unable to sustain the whole initial population; the remaining plants continued to accumulate biomass, which allowed them to continue their vegetative cycle until harvest. In a similar study with peas, Singh et al. (2016) reported increased weed density until 60 DAE and then observed a downward trend

Figure 1. Weed density response to increase in days of duration of weed interference after emergence in the quinoa 'Regalona' crop adjusted to a quadratic polynomial model  $Y = a_0 + (a_1 X) + (a_2 X^2)$ .

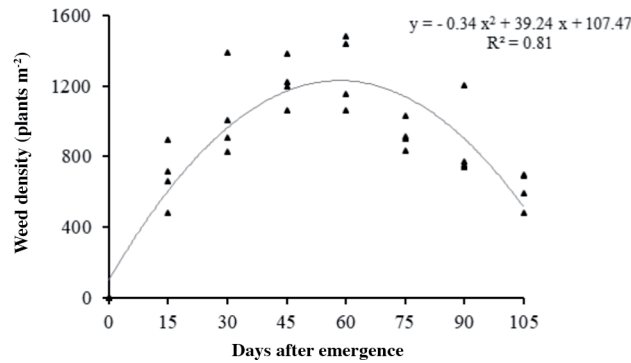
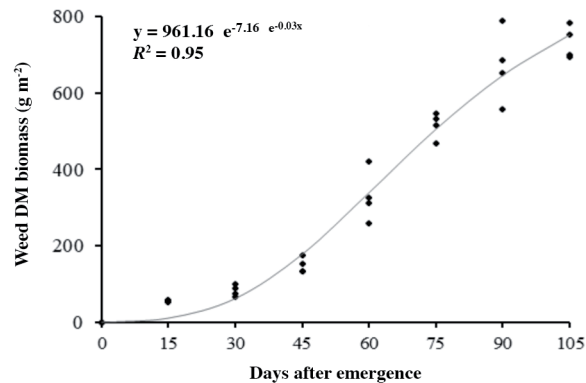


Figure 2. Dry matter biomass response to increase in days of duration of weed interference after emergence. The Gompertz equation adjusted to DM biomass was used.



and concluded that most weeds emerge up to 60 DAE. Weed density was also affected by environmental conditions in both seasons (Table 2); weeds faced high precipitation in the early stages, which could cause their higher germination and emergence and result in higher weed density (Tursun et al., 2012). There was also an increase in temperature during the two seasons (Table 2), and this increase can accelerate weed seed germination and increase competition between different species (Giménez et al., 2013).

The weed DM biomass growth curve was adjusted to the Gompertz model, which showed a highly significant ( $R^2 = 0.95$ ) relationship in which the coefficients to determine model parameters  $a$ ,  $b$ , and  $X_0$  were adjusted to the model ( $P < 0.05$ ) (Figure 2). Maximum weed biomass was recorded in the treatments that had weeds during the whole experiment, that is, 105 d with weeds and 0 d weed-free; therefore, weed DM biomass was directly influenced by the increase in the duration of periods of weed interference and increased until harvest. This could be caused by the shade of the highest weeds and of the crop on the germination of new weed populations. These results concur with data reported by Tursun et al. (2016), who detected increased weed biomass until harvest in three types of corn. On the other hand, Stagnari and Pisante (2011)

Table 2. Meteorological data for mean temperature and precipitation at the experimental site for two seasons.

Month	Mean temperature		Precipitation	
	2015-2016	2016-2017	2015-2016	2016-2017
	°C		mm mo <sup>-1</sup>	
October	12.06	13.27	98.60	65.00
November	15.13	16.65	7.40	11.60
December	18.24	17.86	0.00	31.50
January	20.50	21.50	7.30	3.50
February	19.95	20.40	0.00	13.00

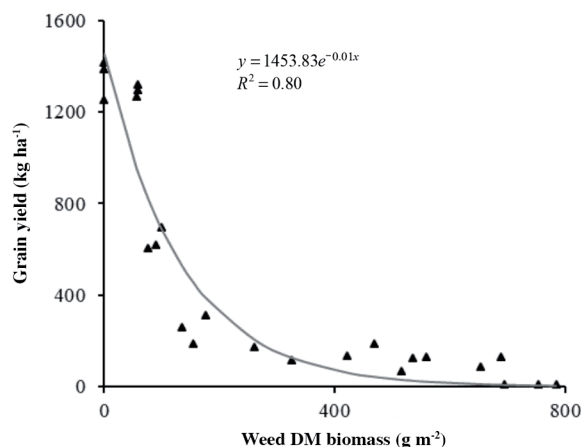
determined that weed biomass increased as the duration of weed infestation increased in a bean crop. Ahymadvand et al. (2009) also found increased weed DM biomass until harvest in a potato crop and reported that weed biomass was higher in low sowing densities; they concluded that open spaces with low sowing densities could increase weed biomass and that early canopy closure at high sowing densities impede weed growth.

The relationship between quinoa grain yield and weed biomass was adjusted to an exponential model with a significant ( $P < 0.05$ ) coefficient of determination and highly significant ( $R^2 = 0.80$ ) relationship. Figure 3 illustrates that grain yield decreased as weed DM biomass increased; therefore, maximum grain yield was obtained in the treatments that remained weed-free during the whole experiment (0 d with weeds and 105 d weed-free). On the other hand, the lowest grain yields were obtained in the treatments with high weed biomass, that is, the treatments that had weeds during the whole experiment (105 d with weeds and 0 d weed-free). This implies that higher weed biomass maintained until later stages can damage crop production because of the weed-crop competition for water, light, and nutrients, which directly affected quinoa yield. Similar studies in other crops have demonstrated yield reduction due to weed interference. Qasem (2009) determined that mean cauliflower yield decreased and was directly affected by weed interference during the crop season. Meanwhile, Singh et al. (2014) reported a negative linear relationship between grain yield and weed DM biomass in rice where treatments with higher weed DM biomass caused a loss of up to 100% of crop yield compared with treatments with lower weed biomass; therefore, weeds would constitute an important restrictive biotic factor that influences yield.

### Critical period of weed interference (CPWI)

The quinoa population remained constant ( $P > 0.05$ ) in all periods, whereas PH varied ( $P < 0.05$ ) in the weed (Table 3) and weed-free (Table 4) growth periods. Thus, quinoa PH increased as the duration of weed interference decreased, that is, as the number of days increased in which treatments remained weed-free. In the weed growth periods (Table 3), PH did not show much difference between treatments ( $P > 0.05$ ) up to 30 DAE; then at 45 DAE, treatments with only 0 and 15 d with weeds exhibited higher values (68.3 and 64.1 cm, respectively) than the mean of the other treatments. At 60 DAE,

**Figure 3. Grain yield response to increase in weed DM biomass in the quinoa ‘Regalona’ crop adjusted to an exponential model  $Y = a \cdot \exp(b X)$ .**



**Table 3. Plant height response to weed growth periods expressed as days after emergence (DAE) of the quinoa ‘Regalona’ crop.**

Treatments	0 DAE	15 DAE	30 DAE	45 DAE	60 DAE	75 DAE	90 DAE	105 DAE
0 d with weeds	1.8a	6.1a	15.2a	68.3a	80.5a	81.1a	83.3a	83.9a
15 d with weeds	1.8a	5.3b	15.0a	64.1a	75.1b	76.3a	77.1a	79.4b
30 d with weeds	1.9a	5.2b	12.9b	42.5b	46.1c	47.7b	49.3b	49.5c
45 d with weeds	1.9a	5.2b	12.7b	40.5bc	43.3cd	45.0bc	45.2bc	45.7cd
60 d with weeds	1.8a	5.3b	12.6b	38.8bc	42.7cde	44.1bc	44.2bcd	45.0de
75 d with weeds	1.9a	5.3b	12.6b	38.0c	39.3de	41.0c	43.2cd	44.7de
90 d with weeds	1.8a	5.3b	12.6b	36.8c	38.8e	40.3c	40.7d	41.4e
105 d with weeds	1.8a	5.3b	12.5b	36.5c	38.5e	40.2c	40.5d	41.0e

The lower-case letters indicate significant differences between treatments (LSD test  $P \leq 0.05$ ).

**Table 4. Plant height response to weed-free growth periods expressed as days after emergence (DAE) of the quinoa ‘Regalona’ crop.**

Treatments	0 DAE	15 DAE	30 DAE	45 DAE	60 DAE	75 DAE	90 DAE	105 DAE
0 d weed-free	1.8a	5.0b	12.8b	49.2c	54.0f	54.6f	54.9f	55.0f
15 d weed-free	1.8a	5.9a	13.0b	64.9b	69.3e	70.6e	70.6e	70.9e
30 d weed-free	1.9a	5.9a	14.6a	66.9ab	76.7d	78.2d	79.8d	80.0d
45 d weed-free	1.8a	5.8a	14.7a	68.4ab	78.9cd	80.2d	80.5d	80.7d
60 d weed-free	1.8a	5.9a	14.9a	69.3ab	83.4bc	84.0c	84.2c	84.4c
75 d weed-free	1.8a	5.8a	15.0a	70.3ab	86.4b	87.7b	88.1b	88.2b
90 d weed-free	1.8a	5.9a	15.1a	71.0a	91.3a	91.9a	92.1a	92.2a
105 d weed-free	1.9a	5.8a	15.2a	72.3a	92.1a	92.8a	92.8a	93.0a

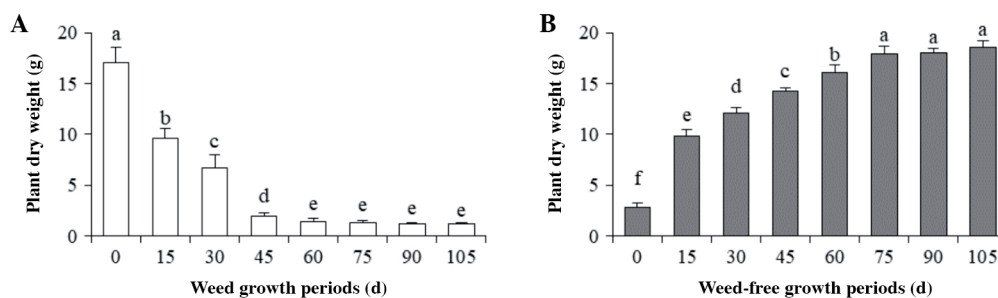
The lower-case letters indicate significant differences between treatments (LSD test  $P \leq 0.05$ ).

the same treatments with weeds at 0 and 15 d maintained the trend and showed higher PH (80.5 and 75.1 cm, respectively) than the other treatments. In the same period, the weed biomass growth curve, adjusted to the Gompertz equation (Figure 2), continued its upward trend, while weed density adjusted to a quadratic model (Figure 1) reached its maximum value. Finally, the same trend was observed at 105 DAE when the two treatments (0 and 15 d with weeds) were significantly different with values of 83.9 and 79.4 cm, respectively, and these PH values were higher than for the other treatments. The treatments with lower PH at 105 DAE were those for 105 and 90 d with weeds; there were no differences between the two and PH was 41.4 and 41.0 cm, respectively.

In the weed-free growth periods (Table 4), trends were similar to the weed growth periods, and PH up to 30 DAE did not exhibit much difference between treatments. At 45 DAE, the 105 and 90 d weed-free treatments had higher values than the mean of the other treatments, 72.3 cm and 71.0 cm, respectively, and there were no differences between the two. At 60 DAE, the same 105 and 90 d weed-free treatments maintained the trend and exhibited the highest PH with 92.1 and 91.3 cm, respectively; there were no differences between the two but were higher than the other treatments. A similar trend was maintained at 105 DAE in which the two treatments (105 and 90 d weed-free) did not differ one from the other with values of 93.0 and 92.2 cm, respectively, and they were significantly higher than the other treatments. The treatment that had significantly lower PH was the 0 d weed-free with a value of 55.0 cm at 105 DAE.

Plant biomass and the grain number per plant variable were significantly ( $P < 0.05$ ) influenced by the treatments with weed and weed-free growth periods. For plant biomass, the treatment with 0 d with weeds (Figure 4A) was higher than the rest of the treatments and had a value of 16.99 g in the weed growth periods, while in the weed-free growth periods (Figure 4B), treatments with 105, 90, and 75 d weed-free were significantly equal and had values of 18.5, 18.0, and 18.0 g, respectively. The highest value for the grain number per plant variable occurred in the treatment at 0 d with weeds (Figure 5A); the value of 4312 grains plant<sup>-1</sup> was significantly different from the other treatments in the weed growth periods. Meanwhile, in the weed-free growth periods (Figure 5B), the highest values were found in treatments with 105 and 90 d weed-free; values were significantly equal at 5110 and 4671 grains plant<sup>-1</sup>, respectively. On the other hand, the 1000 grains weight variable was constant ( $P > 0.05$ ) in the weed growth periods (Figure 6A) and in the weed-free growth periods (Figure 6B); values varied between 3.1 and 2.9 g in weed growth periods and between 3.0 and 2.9 g in weed-free growth periods.

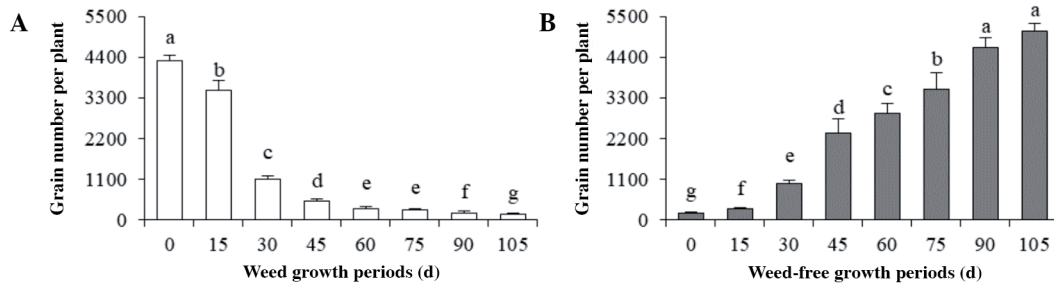
**Figure 4. Plant biomass response to weed interference in the quinoa ‘Regalona’ crop in the growth periods (A) with weeds and (B) weed-free.**



The error bars represent the standard deviation of the mean in each treatment and lower-case letters indicate significant differences between treatments (LSD test  $P \leq 0.05$ ).

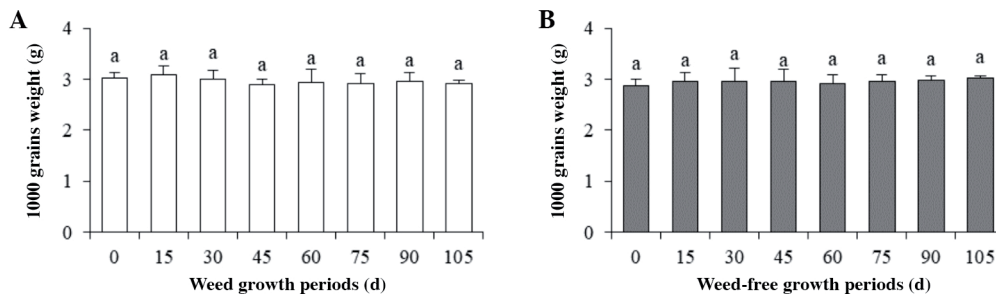


**Figure 5. Grain number per plant response to weed interference in the quinoa ‘Regalona’ crop in the growth periods (A) with weeds and (B) weed-free.**



The error bars represent the standard deviation of the mean in each treatment and lower-case letters indicate significant differences between treatments (LSD test  $P \leq 0.05$ ).

**Figure 6. Response of 1000 grains weight to weed interference in the quinoa ‘Regalona’ crop in the growth periods (A) with weeds and (B) weed-free.**



The error bars represent the standard deviation of the mean in each treatment and lower-case letters indicate significant differences between treatments (LSD test  $P \leq 0.05$ ).

The PH, plant biomass, and grain number per plant showed their highest values in those treatments that remained weed-free during the experiment, while the lowest values were recorded in the treatments that were weed-infested. This is due to the effect of weeds on the development of crops, competing for water, light, nutrients, CO<sub>2</sub> and space, behaving as hosts of pests and diseases (Page et al., 2009), also by the shade effect caused by the highest weeds that reduce available light for photosynthesis and thus biomass production, resulting in decreased yield components (Vasilakoglou and Dhima, 2012).

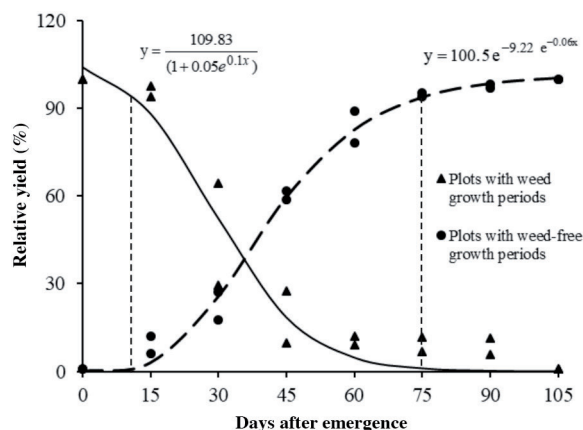
Akhter et al. (2009) reported a decrease in the yield components of a pea crop under weed conditions. Similar studies in other crops have demonstrated decreased yield components due to weed-crop interference. Safdar et al. (2016) reported decreased yield components in a corn crop and Singh et al. (2016) in a pea crop. Weed interference did not affect grain filling, so that the 1000 grains weight variable was the most stable. It is possible that the quinoa plants that were exposed to a longer period of weed interference responded by only forming the number of grains they were able to fill.

Finally, by the logistic and Gompertz regression equations adjusted for grain relative yield (%), CPWI was determined for quinoa ‘Regalona’ at the experimental site between 10 and 75 d after the emergency corresponding to the phenological stages of 2 true leaves and flowering respectively (Table 5) to not have production losses greater than 5% (Figure 7). The estimated parameters used in the equations were adjusted to the model ( $P < 0.05$ ); parameters for the Gompertz equation were the asymptote  $a = 100.5$ , exponential rate  $b = 9.2$ , and the inflection point  $X_0 = 0.06$ . Meanwhile, parameters for the logistic equation were asymptote  $a = 109.8$ , yield rate reduction measurement  $b = 0.05$ , and duration of weed interference  $X_0 = -0.01$ . In similar studies with other crops, Ahymadvand et al. (2009) reported that weight of tubers per plant and total tuber production in a potato crop decreased when the duration of weed interference increased, calculated on the basis of 10% yield losses by the logistic and Gompertz equations. Tursun et al. (2016) reported that the production of three types of corn was influenced by the duration of weed-free and weed-infested periods and CPWI was determined from the development V1 (1 unfolded leaf) stage and maintained until the V12 (12 unfolded leaves) stage.

**Table 5. Phenological stages of quinoa ‘Regalona’ crop at each treatment application in two seasons.**

Days after emergence	Phenological stages
0	Emergence
15	2 true leaves
30	6 true leaves
45	Start of panicle formation
60	Start of flowering
75	Flowering
90	Milky grain
105	Physiological maturity

**Figure 7. Effect of weed control periods on quinoa ‘Regalona’ yield.**

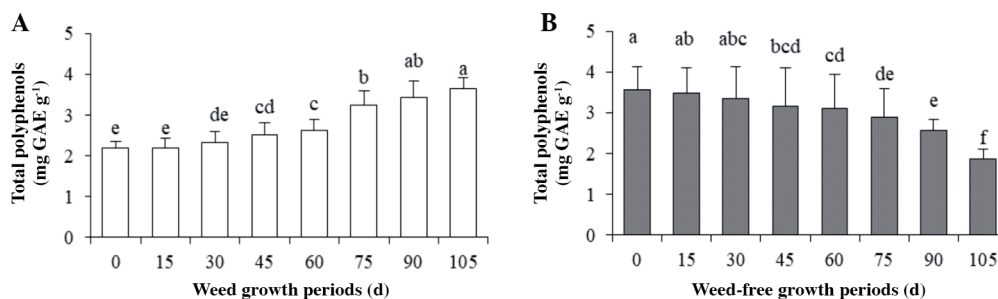


The Gompertz equation was used in weed-free plots and the logistic equation adjusted to grain yield (% yield) was used in weed-infested plots. The weed-free critical periods to achieve 95% of the maximum yield are shown between the vertical dashed lines.

### Total polyphenols

Total polyphenols varied and increased as the duration of weed interference increased and decreased when the duration of weed interference decreased ( $P < 0.05$ ). In the weed growth periods (Figure 8A), polyphenols were increased between 45 and 105 d with weeds, the treatments at 105 and 90 d with weeds were significantly equal and exhibited the same high total polyphenol values, 3.6 and 3.4 mg GAE  $g^{-1}$ , respectively, while the lowest values were observed in the totally or partially weeded treatments. The at 0, 15, and 30 d with weeds were significantly equal and exhibited the lowest total polyphenol values, 2.2, 2.2, and 2.3 mg GAE  $g^{-1}$ , respectively. In the weed-free growth periods (Figure 8B), polyphenols

**Figure 8. Total polyphenol response to weed interference in the quinoa ‘Regalona’ crop in the growth periods (A) with weeds and (B) weed-free.**



The error bars represent the standard deviation of the mean in each treatment and lower-case letters indicate significant differences between treatments (LSD test  $P \leq 0.05$ ). GAE: Gallic acid equivalent.



were decreased between 0 and 90 d weed-free, the weed-free treatments at 0, 15, and 30 d were statistically equal and exhibited the highest total polyphenol values, 3.6, 3.5, and 3.4 mg GAE g<sup>-1</sup>, respectively, while the lowest value was 1.9 mg GAE g<sup>-1</sup> in the totally weeded treatment (105 d weed-free). Most secondary metabolites found in plants accomplish important functions such as protecting against parasites, conferring attractive characteristics for pollinators and seed dispersers, as well as an important role in plant-plant competition in plant-microorganism symbiosis (Olivoto et al., 2016). In the present study, stress caused by weed interference increased total polyphenol concentration as weedy periods increased. Polyphenols are secondary metabolites that are important in plants and accomplish functions in response to stress conditions (Miranda et al., 2013). Reported mean values in some studies indicate that total phenol content in quinoa grains is 1.1 mg GAE g<sup>-1</sup>, and these values are higher than those obtained in traditional cereal seeds such as barley, wheat, rice, and millet that varied between 0.16 and 0.36 GAE g<sup>-1</sup> (Asao and Watanabe, 2010; Djordjevic et al., 2010). Meanwhile, Fischer et al. (2013) determined the variation of the antioxidant capacity in quinoa subjected to different water stress levels and found an increase in total polyphenol content between 3.3 and 4.5 mg GAE g<sup>-1</sup> as water restriction increased in the quinoa 'Regalona' crop under field conditions. Miranda et al. (2010) also determined a notable reduction in total polyphenol content when quinoa seed was subjected to high temperatures with hot air.

## CONCLUSIONS

The critical period of weed interference was determined between 10 and 75 d after the emergency, which corresponded to the phenological stages of two true leaves and flowering, respectively. The determination of this period will allow crop management decision-making to minimize losses produced by weed interference. Stress caused by weed interference altered polyphenol contents and affected quinoa production. Among the yield components, the grain number per plant showed the greatest differences and directly affected yield. Total polyphenols varied and increased with a longer weed interference period, while they decreased with a shorter weed interference period. Thus, in the weed growth periods, polyphenols increased between 45 and 105 d with weeds, whereas polyphenols decreased between 0 and 90 d weed-free in the weed-free growth periods. Higher amount total polyphenols in the quinoa crop and lower crop yield were due to stress caused by weed interference.

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