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## RESEARCH NOTE

# Seeds Yield and Quality of Quinoa (*Chenopodium quinoa* Willd.) Plants Grown Under Different Nitrogen Fertilization Doses

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## Abstract

**S. Contreras, J. Molina, M. García, J. Sánchez, R. Chorbadjian, F. Fuentes, and F. Albornoz. 2024. Seeds Yield and Quality of Quinoa (***Chenopodium quinoa* Willd.) Plants **Grown Under Different Nitrogen Fertilization Doses. Int. J. Agric. Nat. Resour. 68-74.** Quinoa (*Chenopodium quinoa* Willd.) has been cultivated for more than 7,000 years in the mountain regions (Andes Mountains) of South America. Five ecotypes differing in their phenology, morphology and stress resistance can be found. One of these ecotypes corresponds to the coastal ecotype, which is cultivated mainly in Chile. Little information is available regarding the response of this ecotype to nitrogen (N) fertilization. In the present study, three N fertilization doses (low: 30, adequate: 140 and high: 280 kg ha<sup>-1</sup>) were evaluated in plants grown in containers. Seed yield and quality attributes, including seed weight, nitrate content, germination, and seed longevity, were evaluated. These results indicate that increasing N fertilization treatments did not affect germination, seed longevity was reduced by increasing the N fertilization dose.

**Keywords:** Quinoa coastal ecotype, seed dormancy, seed germination, seed longevity, thousand seed weight.

## Introduction

Quinoa (*Chenopodium quinoa* Willd.) is an important crop in the Andean Region of South America, where it has been cultivated for more than 7,000 years (Jarvis et al., 2017). This crop is gaining attention worldwide mainly because

of its tolerance to adverse climatic conditions and the high nutritional value of its grains (Vázquez-Luna et al., 2019). Different quinoa ecotypes are available depending on the region of cultivation. In Chile, the cultivation of the coastal quinoa ecotype has been reported since the arrival of Spanish colonizers (Von Baer et al., 2009). However, little information is available regarding the effect of fertilizer management on grain yield and seed quality in this ecotype. The nitrogen (N) supply to the crop plays an important role in determining quinoa yield, with the literature indicating an optimum dose

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between 120 and 170 kg N ha<sup>-1</sup> (Schulte et al., 2005; Almadini et al., 2019; Bascuñán-Godoy et al., 2018). This supply of N ensures a protein content in the grain between 11 and 21%, a value much higher than that found in common cereals (Thanapornpoonpong et al., 2008).

With regard to seed quality, important attributes such as germination, dormancy and longevity (i.e., the capacity of the seed to preserve its germination potential during storage) have been shown to vary according to the environment of the mother plant. For instance, relatively high temperatures and long photoperiods result in deeper dormancy in guinoa seeds (Ceccato et al., 2011): however, seed longevity is not affected by these environmental parameters across different quinoa ecotypes (Ayala et al., 2020). Little information is available regarding seed quality attributes in grains obtained from plants exposed to different N concentrations. Therefore, the aim of the present study was to assess seed yield and quality in a coastal ecotype of quinoa cultivated under different nitrogen fertilization doses.

#### **Materials and Methods**

#### Growth conditions and nitrogen treatments

Seeds of quinoa cv. 'Regalona-Baer' were sown in 3 L containers using a mixture of peat (Kekkilä DSM 0, Kekkilä-BVB, Vantaa, Finland) and perlite (Harbolite® A-6 Harbolite Chile Ltda., Santiago, Chile) at a ratio of 2:1 (v/v). Six seeds were sown per container and thinned 10 days after emergence to leave one plant in each container. The plants were established on November 18, 2020, and were grown under summer environmental conditions, which corresponds to their natural growth period. The average temperature and relative humidity (RH) during the day were 24 °C and 51%, respectively, while during the night, they were 16 °C and 73%, respectively. The day length varied from 14 to 12 hours during the growing period.

Plants were divided into three groups, and each group received one of the following N fertilization treatments: a low dose equivalent to 30 kg N ha<sup>-1</sup>. an optimum dose of 140 kg N ha-1, or a high dose equivalent to 280 kg N ha-1. Nitrogen was supplied using KNO<sub>2</sub>. Plants also received the equivalent of 240 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> (applied as KH<sub>2</sub>PO<sub>4</sub>) and 480 kg K<sub>2</sub>O ha<sup>-1</sup> (applied as KNO<sub>2</sub> and K<sub>2</sub>SO<sub>4</sub>). The complete dose of each treatment was divided into four applications: 10%, 35%, 35% and 20% of the total fertilizer applied in phenological growth stages 12, 20, 30 and 40 (Sosa-Zuñiga et al., 2017), respectively. Additionally, at the 12th stage, 15 mg of Fetrilon plant<sup>-1</sup> (Compo Expert, Münster, Germany) was applied to provide micronutrients. Plants underwent regular watering to achieve saturation, with intervals typically occurring every 1 to 2 days. The plants were arranged in a randomized complete block design with 10 replicates per treatment.

#### Measurements

## Seed yield and quality

Seeds were harvested 120 days after sowing (stage 95 according to Sosa-Zuñiga et al., 2017), and the seed yield was recorded as grams of seeds per plant. Seed harvesting and cleaning were performed manually, and the clean seeds from each plant were stored in paper bags at room temperature until evaluation.

The thousand-seed weight (TSW) was determined for four samples for each replicate. For this purpose, 100 seeds were weighed, and the TSW was calculated as 10 times the average seed weight of 100 seeds.

The seed nitrate (NO<sub>3</sub><sup>-</sup>) content was determined by colorimetry at 410 nm. Three-gram samples were suspended in 10 mL of deionized water. The suspensions were incubated at 45 °C for 1 hour. Then, 0.2 mL aliquots of the extracts were added to 50 mL Erlenmeyer flasks and mixed with 0.8 mL of 5% (w/v) salicylic acid in concentrated  $H_2SO_4$ . After 20 minutes at room temperature, 19 mL of 2 N NaOH was added, and the samples were cooled to room temperature for later determination of the absorbance at 410 nm.

Standard and physiological germination was evaluated at 20 °C by sowing seeds in Petri dishes (90 mm) over two layers of filter paper (87 g m<sup>-2</sup>; Munktell Filter) saturated with 6 mL of distilled water or a polyethylene glycol solution (PEG 8000, Sigma-Aldrich). For standard germination, three Petri dishes with 25 seeds each were evaluated per replicate, and normal seedlings were counted seven days after sowing (González & Aranciaga, 2021). For physiological germination (PG), a Petri dish with 50 seeds was evaluated per replicate, and germinated seeds (radicle emergence > 2 mm) were counted daily until 7 days after sowing. PG was evaluated in water (0 MPa) or in PEG solutions with osmotic potentials between -0.3 and -0.9 MPa (Sánchez et al., 2022). The germination index (GI) was calculated according to the following equation (Avala et al., 2020):

GI = 
$$\sum_{i=1}^{7} [(\text{fraction of seeds germinated on day } i)/i]$$

For seed longevity evaluation, an accelerated aging treatment consisting of subjecting seeds to 41 °C and 96% RH in hermetic containers with a saturated solution of  $K_2SO_4$  (Sánchez et al., 2022) was implemented. The containers were kept in a

water-jacketed incubator (Sheldon Manufacturing, Cornelius, OR) during the aging period. Seed PG was subsequently assessed after 10, 15 or 20 days of accelerated aging.

## Statistical Analysis

Germination variables were analyzed using a generalized linear model (GLM) with logit links and binomial distributions. All variables were evaluated by ANOVA (p<0.05), and mean comparisons were performed using Tukey's test. Analyses were carried out using R statistics software (v.4.1.1) through the RStudio console.

#### Results

As shown in Table 1, increasing the N supply to the mother plants resulted in significant increases in seed yield, weight, and nitrate content (p<0.001). However, no significant differences were detected for standard germination (p=0.380), PG (p=0.180), or GI (p=0.184) (Table 1).

As expected, the PG was affected when seeds were soaked in solutions with reduced osmotic potentials (Figure 1). However, seeds from the different N treatments presented no significant difference in the response of their germination percentage (p=0.525) or GI (p=0.487) to the different osmotic potential solutions.

Table 1. Yield and quality attributes of quinoa seeds produced under three different nitrogen supply treatments. The values represent the mean  $\pm$  S.E. of ten replicates. Different letters in the same row denote significant differences (p<0.05).

	Nitrogen fertilization dose, kg ha-1			n voluo
	30	140	280	p value
Seed yield (g plant <sup>-1</sup> )	13.2 ±1.1 c	$20.2 \pm 0.9 \text{ b}$	25.0 ±2.2 a	< 0.001
Thousand seed weight (mg)	2427 ±4 b	2487 ±3 b	2720 ±5 a	< 0.001
Nitrate (mg KNO <sub>3-</sub> kg <sup>-1</sup> )	185.0 ±5.4 c	233.7 ±6.2 a	212.5 ±5.3 b	< 0.001
Standard germination, %	98 ±0.01	$99 \pm 0.01$	$98 \pm 0.01$	0.380
Physiological germination, %	$100\pm\!\!0.0$	99 ±0.2	98 ±0.4	0.180
Germination Index	$0.499 \pm 0.003$	$0.495 \pm 0.003$	$0.492 \pm 0.003$	0.184

Seeds subjected to accelerated aging presented a significant reduction in the germination percentage (Figure 2A) and germination rate (Figure 2B), especially after 20 days of treatment. The aging effects were greater in seeds from the higher N fertilization treatment than in those from the lower N fertilization treatment (p<0.05), which indicates that increasing the N supply to the mother plant decreases seed longevity.

## Discussion

Previous studies using cv. Titicaca and Chipava showed a significant and positive effect of up to 160 kg N ha-1 fertilization on quinoa grain yield (Alandia et al., 2016; Almadini et al., 2019), which is similar to the response observed in our study. However, our results indicate that further increases in the N supply, up to 280 kg N ha<sup>-1</sup>, significantly increased the seed yield per plant and seed weight. This response could be attributed to an increase in photosynthetic radiation use efficiency due to an increase in chlorophyll and protein content in the leaves (Bascuñán-Godoy et al., 2018). Due to the experimental conditions of this study (plants growing in pots with soilless media), the 280 kg N ha-1 dose should not be considered a recommendation for producing quinoa in open fields. However, these results emphasize the importance of considering the cultivated genotype when determining the optimal fertilization rate.

With regard to seed quality, the range of the N supply evaluated in the present study had no effect on seed germination, regardless of expression as standard (percentage) or physiological (percentage and IG) germination, indicating that these seed quality attributes are less sensitive to N fertilization than are yield components (g plant<sup>-1</sup> and TSW). Similar results were reported when three fertilization doses were evaluated for tomato seed production (Sánchez et al., 2022). In contrast, a reduction in seed yield and germination was observed in lettuce when fertilization was increased from 10 to 20 mM of N in the watering solution in soilless culture (Albornoz et al., 2019).

In tomato, N fertilization of the mother plants affected the sensitivity of the seeds to germination when soaked in a solution with negative osmotic potential (-0.2 MPa; Sánchez et al., 2022), and the authors interpreted those results as modifications in seed physiological dormancy. Additionally, it has been argued for Arabidopsis that increasing the nitrate content in seeds reduces dormancy (Yan & Chen, 2020). In quinoa, nondeep physi-



**Figure 1.** Effects of different nitrogen fertilization treatments on the physiological germination of seeds soaked in solutions with osmotic potentials ranging from 0.0 to -0.9 MPa. Germination is presented as a percentage. Each symbol represents the mean  $\pm$  S.E. of ten replicates.



Figure 2. Effect of nitrogen fertilization on seed germination after accelerated aging at 41  $^{\circ}$ C and 96% relative humidity. A. Total germination after 0, 10, 15 and 20 days of aging. B. Accumulated germination after 20 days of aging. Each symbol represents the mean  $\pm$  S.E. of ten replicates.

ological dormancy has been reported for some coastal genotypes, including cv. 'Regalona' (Ceccato et al., 2011; Ayala et al., 2020), which is important for preventing preharvest sprouting of seeds (McGinty et al., 2021). In the present study, significant differences in nitrate content were observed among seeds from the different N fertilization treatments (Table 1). However, when seeds were soaked in solutions with low osmotic potentials, our results showed no difference in germination among the seeds from the different N treatments (Figure 1), indicating no difference in seed dormancy and, therefore, preharvest sprouting resistance.

In relation to longevity, the results showed that the highest N dose supplied to quinoa mother plants resulted in the lowest seed longevity (Figure 2), which is consistent with previous results reported for lettuce (Albornoz et al., 2019). However, the opposite was observed in tomato, where the highest fertilization dose of N to the mother plant resulted in seeds with greater longevity (Sánchez et al., 2022). A possible explanation for these contrasting results could be related to the different types of fruits that hold the seeds in these species: dry fruits in lettuce and quinoa versus wet fruits in tomato. However, the mechanism underlying the effects of N fertilization on seed longevity remains unknown and deserves further study. In agreement with what has been observed by other authors, longevity was shown to be a more sensitive seed quality attribute, as differences could not be detected through germination tests (Albornoz et al., 2019; Sánchez et al., 2022).

### Conclusion

Under the experimental conditions used in the present study, the seed yield of the coastal ecotype of quinoa cv. 'Regalona' increased in response to increasing nitrogen fertilization doses up to 280 kg ha<sup>-1</sup>. No significant effects on seed germination or dormancy were observed under nitrogen fertilization doses between 30 and 280 kg ha<sup>-1</sup>. However, a significant reduction in seed longevity occurred when seeds were produced under the highest N fertilization treatment, which should be considered when the cultivation objective is the production of high-quality seeds for planting.

#### Resumen

S. Contreras, J. Molina, M. García, J. Sánchez, R. Chorbadjian, F. Fuentes, y F. Albornoz. 2024. Rendimiento y calidad de semillas de quinoa (*Chenopodium quinoa* Willd.) producida con diferentes dosis de fertilización. Int. J. Agric. Nat. Resour. 68-74. La quinua (*Chenopodium quinoa* Willd.) se cultiva desde hace más de 7.000 años en las zonas montañosas (Cordillera de Los Andes) de América del Sur. Existen cinco ecotipos que difieren en su fenología, morfología y resistencia al estrés. Uno de estos ecotipos corresponde al ecotipo de la costa, cultivado principalmente en Chile. Hay poca información disponible con respecto a la respuesta de este ecotipo a la fertilización con nitrógeno (N). En el presente estudio se evaluaron tres dosis de fertilización nitrogenada (baja: 30, adecuada: 140 y alta: 280 kg ha<sup>-1</sup>) en plantas de contenedor. Se evaluaron los atributos de rendimiento y calidad de la semilla, incluido el peso, el contenido de nitrato, la germinación y la longevidad. Los resultados indican que el aumento de la fertilización nitrogenada promueve el rendimiento y el peso de la semilla. En cuanto a los atributos de calidad de semilla, aunque los tratamientos de fertilización no afectaron la germinación, la longevidad de la semilla se redujo al aumentar las dosis de fertilización nitrogenada.

Palabras clave: Ecotipo de la costa, dormancia de semilla, germinación, longevidad, peso de mil semillas.

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