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Original Research

Optimizing Quinoa Cultivation: Combined NPK Fertilization as a Key Agronomic Practice Steering Yield, Growth, and Nutritional Quality

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Abstract

Background: Quinoa (*Chenopodium quinoa* Willd.), valued as a nutrient-dense grain, has seen expanded cultivation in China. However, its yield and quality are often unstable due to suboptimal fertilization practices, and the specific regulatory roles of macronutrients (N, P, K) remain unclear. This study aimed to elucidate the effects of combined NPK fertilization on the growth, yield, and nutritional quality of quinoa. Methods: A pot experiment was conducted in a controlled climate chamber using the “Jingli 1” cultivar. Plants were subjected to four fertilization treatments: NPK, PK, NK, and NP. Results: The NPK treatment significantly enhanced growth throughout the development cycle, increasing total root length (45.0±8.5%), root surface area (32.8±6.2%), plant height (20.0±5.0 cm), and dry matter accumulation (71.6±15.3%). It also improved nitrogen use efficiency and key yield components, including panicle length (5.63±1.22 cm), thousand-grain weight (0.34±0.02 g), and grain weight per plant (1.41±0.23 g). Nutritionally, the NPK treatment elevated the content of essential amino acids (284.10±52.75 mg/g), total free amino acids (2414.65±428.30 mg/g), and increased levels of protein, starch, and fat. Deficiency analyses revealed distinct limitations: nitrogen deficiency primarily constrained aboveground growth and yield; phosphorus deficiency strongly inhibited root development and dry matter accumulation; while potassium deficiency had comparatively minor effects. The synergistic application of NPK fertilizers is crucial for optimizing quinoa production. It promotes robust root and shoot growth, enhances nitrogen uptake, and simultaneously achieves high yield and superior nutritional quality, providing a scientific basis for optimized quinoa cultivation practices.

Keywords: quinoa, nitrogen, phosphorus, potassium fertilizers, yield, nutritional quality

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Introduction

Quinoa (*Chenopodium quinoa* Willd.) is an annual dicotyledonous plant belonging to the genus *Chenopodium* and family Amaranthaceae. Originating in South America, it has gained widespread global attention in recent years. Its grains are rich in high-quality protein, essential amino acids, minerals, and dietary fiber, earning the reputation of a "superfood" [1-3]. It thrives in cool high-altitude regions and is now cultivated worldwide owing to its food safety and complete nutritional profile, making it a research focus for scholars internationally. Previous studies have primarily concentrated on the nutritional value, extraction, and processing of chemical components, stress resistance, and introduction of cultivation. Research confirms that quinoa protein content is as high as 18.0±4.0%, which is 2-3 times that of rice. The grains are rich in essential amino acids required for human activities, including lysine, which is often deficient in common cereals, making it easily absorbed by the human body [4]. Quinoa seeds are abundant in carbohydrates, with high proportions of D-xylose and maltose, and low contents of glucose and fructose (Ogungbenle [5]). Its high dietary fiber content makes it popular among individuals seeking weight loss [6]. Quinoa is rich in antioxidant active substances such as flavonoids and polyphenols, and thus possesses good antioxidant and antiaging properties [7]. It is also rich in minerals and unsaturated fatty acids and contains almost no gluten, making it an excellent food for people with celiac disease or allergies [8, 9]. Long-term consumption can enhance immunity and help prevent cardiovascular and cerebrovascular diseases, making it one of the most favored health foods in the market. Owing to its safety and complete nutrition, it can serve as a food source for astronauts [10]. Owing to its rich nutritional value, researchers, including [11, 12].

Preliminary studies have begun to explore the agronomic practices suitable for these new environments, investigating the effects of key macronutrients like nitrogen (N), phosphorus (P), and potassium (K) on quinoa growth and development [13-15]. However, the specific regulatory effects of these macronutrients, particularly their synergistic application on yield formation and nutritional quality, are not yet fully clarified. Introduced relatively late in China, quinoa has been trialed in some areas of Qinghai, Gansu, and Ningxia [16], achieving certain results. The earliest area of introduction in the Shanxi Province was Jingle County, Xinzhou City. As an introduced crop, quinoa is suitable for growing in cool and cold highland regions, and exhibits strong adaptability to different agricultural ecologies. Studies have found that quinoa can grow at temperatures from -4ºC to 38ºC, but its optimal growth temperature is 15-20ºC. It is a crop with high water-use efficiency and can yield acceptable harvests, even with rainfall between 100-200 mm [17]. Its growth period varies by cultivar, ranging from 90 to 225 d [18]. However, owing to the lack of standardized cultivation practices for quinoa, blind fertilization occurs in production, leading to imbalances in nutrient elements, which in turn triggers physiological disorders in quinoa, reducing yield and quality. Nitrogen (N), phosphorus (P), and potassium (K) are essential macronutrients for plant growth and play key roles in regulating crop physiological processes, dry matter accumulation, and grain formation [19]. Applying phosphorus at 200 mg/g phosphorus promoted root growth at the seedling stage, enhanced root activity, and improved antioxidant capacity and drought resistance [20]. Potassium application significantly increased plant height and stem diameter of quinoa. Increasing the application rate to 0.024 mg/g increased quinoa biomass and the cumulative uptake of N, P, and K, but these parameters decreased when the rate exceeded 0.024 mg/g [21]. The Tiaoshan Farm in Gansu showed that an N application rate of 60 kg/hm² with a base-to-topdressing ratio of 1:2 significantly increased quinoa yield and economic benefits, improving yield by 86.8% and increasing net income by 14,196.54 RMB/hm² compared to no fertilization [22]. A study at the Sanping Experimental Base of Xinjiang Agricultural University found that when N, P, and K application rates were 160, 140, and 75 kg/hm², respectively, quinoa responded best to nitrogen fertilizer, increasing yield by 30.01% compared to the control. Thus, previous research has mainly focused on the effects of nitrogen (N), phosphorus (P), and potassium (K) on the seedling growth, yield, and economic benefits of quinoa. However, the effects and mechanisms of two-factor (PK, NK, and NP, i.e., lacking N, P, or K, respectively) or three-factor (NPK) applications on quinoa yield and quality remain unclear. The Shanxi Province began breeding quinoa varieties around 2008, gradually starting commercial cultivation. The current planting area reached 35,000 mu, demonstrating adaptability. Therefore, this study employed pot experiments in Taigu, Shanxi to investigate the effects of PK, NK, NP, and NPK fertilization on quinoa growth, yield, and efficiency. It aims to reveal the response of quinoa to N, P, and K fertilizers and the agronomic and physiological mechanisms affecting yield and quality, providing theoretical support for stable quinoa production.

Materials and Methods

Experimental Site Overview

A pot experiment was conducted in 2024 in an artificial climate chamber at the Wheat Research Base of Shanxi Agricultural University. The plants were grown under controlled conditions with a temperature regime of 26ºC for 16 hours and 18ºC for 8 hours, 60% relative humidity, and a light intensity of 50,000 Lux. The test cultivar used was "Jingli 1" (JQ307), provided by Shanxi Jiaqi Agricultural Technology Co., Ltd.

The experiment included four fertilizer treatments: NPK (nitrogen, phosphorus, and potassium), PK (phosphorus and potassium), NK (nitrogen and potassium), and NP (nitrogen and phosphorus), with a total of 20 pots. The pots measured 40 cm in height and 20 cm in diameter. The potting medium was unweathered subsoil, with each pot containing 30 kg of soil. Fertilizer application rates were calculated based on field equivalents of 150 kg N/hm², 100 kg P_2O_5/hm², and 40 kg K_2O/hm², corresponding to a field soil weight of 225,000 kg/hm². The specific fertilizer applications per pot were as follows: NPK treatment received 1.5 g N, 1.0 g P_2O_5, and 0.4 g K_2O; PK treatment received 1.0 g P_2O_5 and 0.4 g K_2O; NK treatment received 1.5 g N and 0.4 g K_2O; and NP treatment received 1.5 g N and 1.0 g P_2O_5. Sowing was carried out on April 26, 2024. At the 6-8 leaf stage, seedlings were thinned to two plants per pot. After final seedling establishment, irrigation was maintained at 1000 mL per day. The key growth stages recorded were: branching stage on June 8, panicle emergence stage on June 28, and flowering stage on July 15. Manual weeding and pest control were performed as needed during the growth period. The plants were harvested on August 22, 2024.

Agronomic Traits

Root characteristics: At each growth stage, the entire root system was collected and scanned using a root scanner (Epson Perfection V850 Pro) to measure total root length, root surface area, total root volume, and root tip number.

Plant height: The plant height was measured at each growth stage, from the stem base to the highest point of the stem.

Branch number and effective branch number: Sampling was conducted at the panicle emergence, flowering, and maturity stages to determine the total number of branches (including those with and without panicles) and the number of effective branches (branches capable of bearing seeds).

Panicle traits: The panicle length and width were measured at the panicle emergence, flowering, and maturity stages. The length of the main panicle was measured with a tape measure, and the panicle width was measured using a vernier caliper.

Dry matter weight: At the seedling, branching, panicle emergence, flowering, and maturity stages, representative plants with uniform growth were collected from the field. After separating the organs, the samples were dried at 105ºC for 30 min for enzyme deactivation and then at 75ºC until a constant weight was achieved.

Plant Nitrogen Content

The dried samples of plant organs from each growth stage were crushed. Nitrogen content was determined using the method described by [23] (H_2SO_3-H_2O_2-indophenol blue colorimetry). Nitrogen accumulation and related indicators were calculated using the method described by [11].

Yield and its Components

Grain number per panicle: Harvesting was conducted when 80% of the quinoa leaves had turned yellow and began to fall. The effective panicles from each pot were threshed, sun-dried, and the number of grains per panicle was counted.

Thousand-grain weight: One thousand grains were counted, and three replicates were weighed.

Yield: After threshing and removing impurities, the grain weight per plant was measured as the yield.

Grain Nutritional Quality

Protein: The nitrogen content in the grains was determined using H_2SO_3-H_2O_2-indophenol blue colorimetry. Protein content was calculated as the nitrogen content multiplied by 5.7.

Amino acids: Quinoa grains were sent to Ji Yin (Shanghai) Biotechnology Co., Ltd. for analysis using LC-MS metabolomics instrumentation (ExionLC liquid chromatograph and AB 6500 Plus mass spectrometer).

Starch: The anthrone colorimetric method from Gao Junfeng"s Plant Physiology Experimental Guide was used to determine starch content [24].

Fat, crude fiber, and ash: Fat content was measured using a semi-automatic fat determination instrument (VELP/SER148/6) following GB/T 38475-2021. The crude fiber content was determined using a semiautomatic fiber analyzer (VELP/FIWE6) following GB/T 37492-2019. The ash content was measured using a muffle furnace for high-temperature ashing following GB 5009.4-2016.

Calculation Methods

Plant Nitrogen Accumulation (kg/hm²) = Plant Dry Matter Weight × Nitrogen Content

Pre-Anthesis:

Nitrogen Translocation (kg/hm²) = Nitrogen Accumulation in Vegetative Organs at Flowering Stage – Nitrogen Accumulation in Vegetative Organs at Maturity Stage

Contribution rate of Pre-Anthesis Nitrogen Translocation to grains (%) = (Pre-Anthesis Nitrogen Translocation / Grain Nitrogen Accumulation) × 100%

Post-Anthesis:

Nitrogen Accumulation (kg/hm²) = Plant Nitrogen Accumulation at Maturity Plant Nitrogen Accumulation at Flowering

Contribution Rate of Post-Anthesis Nitrogen Accumulation to Grains (%) = (PostAnthesis Nitrogen

Accumulation / Grain Nitrogen Accumulation) × 100%

Nitrogen Use Efficiency (kg/kg) = Grain Yield / Plant Nitrogen Accumulation

Statistical Methods

Data were organized using Excel 2019 (Microsoft Inc., Redmond, WA, USA). Variance analysis was performed using the SPSS software (IBM SPSS Statistics, version 26.0). Graphs were generated using Origin 2024 (version 10.10.178).

Results

Effects of Nitrogen, Phosphorus, and Potassium Fertilizers on Root System Characteristics of Quinoa at Various Growth Stages

As the growth period progressed, the total root length, root surface area, total root volume, and number of root tips of quinoa at each growth stage all exhibited a trend of first increasing and then decreasing, peaking at the mid-filling stage (Fig. 1). Compared to other fertilization methods, the combined application of NPK fertilizers significantly enhanced root system characteristics from the branching stage to the maturity stage. Specifically, it increased the total root length by 50.0±11.3%, root surface area by 32.8±8.2%, total root volume by 74.8±0.8%, and the number of root tips by 76.2±2.1%. The increases were more pronounced after the panicle emergence stage, especially when compared with the NK treatment. However, the NPK treatment had no significant effect on the root system at the seedling stage. The combined application of NPK fertilizers promotes root growth in quinoa. Nonetheless, the absence of any of the nitrogen, phosphorus, or K fertilizers adversely affects root development, with the lack of phosphorus fertilizer having a more noticeable impact.

Effects of Nitrogen, Phosphorus, and Potassium Fertilizers on Agronomic Traits of Quinoa

During the growth process, quinoa plant height showed a slow-fast-slow trend. Dry matter accumulation showed a “slow-fast-fast” trend (Fig. 2). Compared with other fertilization methods, the combined application of nitrogen, P, and K fertilizers can significantly increase the plant height of quinoa at each growth stage. At maturity, plant height increased by 10-30 cm, and the effect was more obvious than that of nitrogen and K fertilizers. It can significantly increase the dry matter accumulation of quinoa at each growth stage, reaching 71.6±29.8% at the maturity stage, and the effect is more obvious than that of nitrogen and potassium fertilizers. It can be seen that the combined application of nitrogen, P, and K fertilizers is beneficial to the growth of the aboveground part of quinoa, and the growth of the aboveground part is affected by the lack of nitrogen, phosphorus, or potassium fertilizers, especially phosphorus fertilizers.

Effects of Nitrogen, Phosphorus, and Potassium Fertilizers on Branch Number and Effective Branch Number of Quinoa at Different Growth Stages

With the development of the growth process, the number of branches and effective branches of quinoa at each growth stage showed a trend of increasing

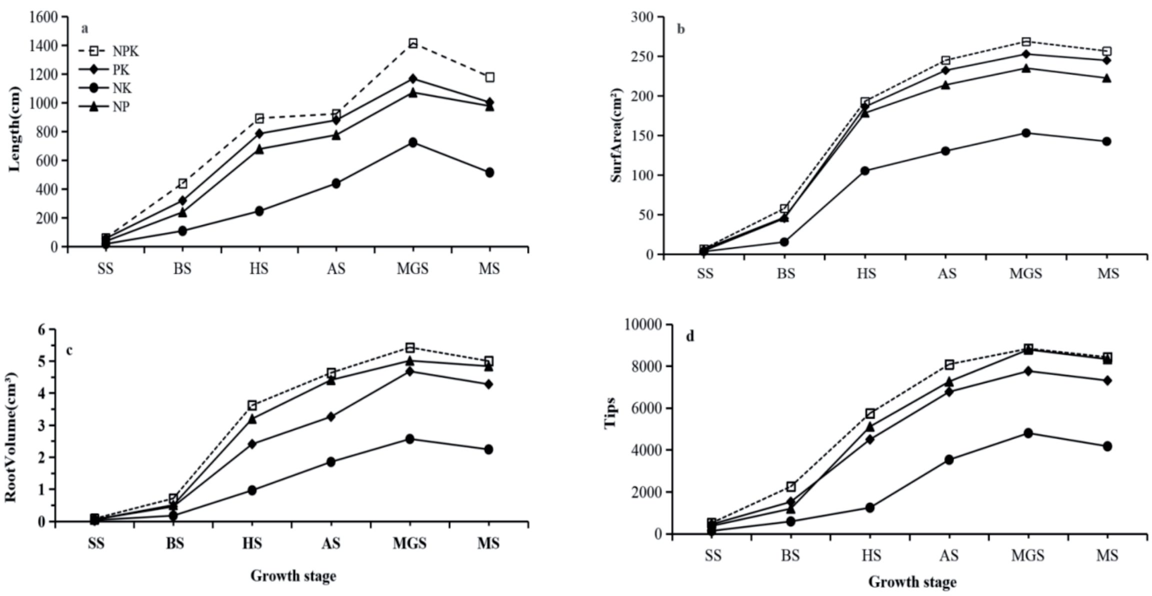

Fig. 1. Effects of nitrogen, phosphorus, and potassium on root characteristics of quinoa at different growth stages.
Note: SS, Seedling Stage; BS, Branching Stage; HS, Heading Stage; AS, Anthesis Stage; MGS, Mid Grain-Filling Stage; MS, Maturity Stage.

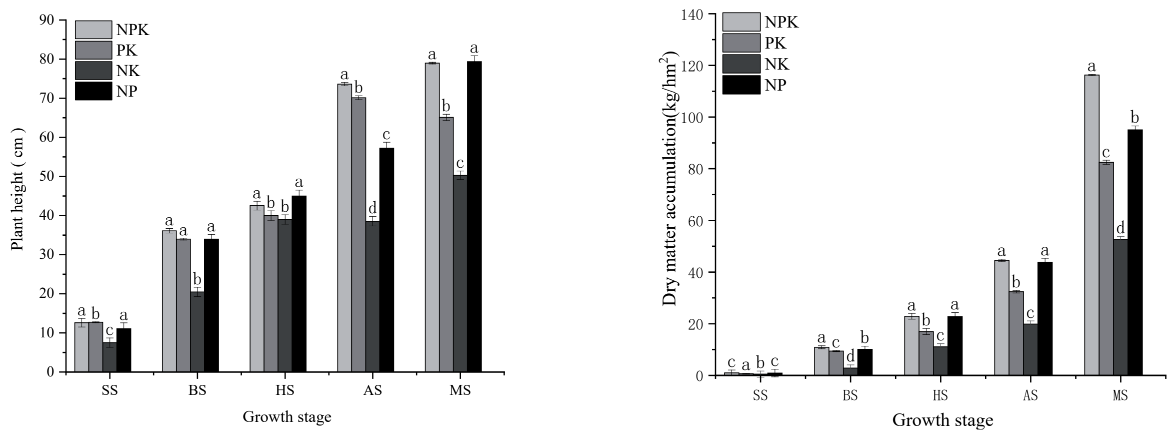

Fig. 2. The effects of nitrogen, phosphorus, and potassium on plant height and dry matter of quinoa at different growth stages.
Note: SS, Seedling Stage; BS, Branching Stage; HS, Heading Stage; AS, Anthesis Stage; MGS, Mid Grain-Filling Stage; MS, Maturity Stage.

first and then decreasing and peaked at the flowering stage, except for nitrogen and phosphorus fertilizer treatment (Table 1). Compared with other fertilization methods, the combined application of nitrogen, P, and K fertilizers can significantly increase the number of effective branches of quinoa at each growth stage, and the number of effective branches at the maturity stage reached 6.5-15. Compared with phosphorus potassium fertilizer, and nitrogen potassium fertilizer, the combined application of nitrogen, phosphorus, and potassium fertilizer significantly increased the effective branching rate of quinoa at the flowering stage, and there was no significant difference in the effective branching rate at the flowering stage compared with nitrogen and phosphorus fertilizers. Compared with other fertilization methods, the combined application of nitrogen, P, and K can significantly increase the effective branching rate of quinoa at the maturity stage, which is more obvious than that of phosphorus and K fertilizers and nitrogen and K fertilizers. It can be seen that the combined application of nitrogen, phosphorus, and potassium fertilizers is beneficial to the formation of effective branches of quinoa and increases the effective branch rate. The lack of nitrogen or phosphorus fertilizers affected the formation of effective branches, while the lack of potassium fertilizer had no significant effect on the formation of effective branches.

Effects of Nitrogen, Phosphorus, and Potassium Fertilizers on Nitrogen Accumulation of Quinoa Plants

With the passage of the growth process, the nitrogen accumulation of quinoa plants showed a gradual increase trend of “slow-fast-slow”, reaching the maximum at maturity, 69.91±13.31 kg/hm^2 (Table 2). Compared with other fertilization methods, nitrogen, phosphorus, and potassium fertilizers can increase the nitrogen accumulation of quinoa plants at each growth stage, and there are significant differences between nitrogen, phosphorus, and potassium fertilizers and nitrogen, potassium, and phosphorus and potassium fertilizers. There was a significant difference between nitrogen, phosphorus, and potassium fertilizers and nitrogen and phosphorus fertilizers at the flowering stage, but there was no significant difference in other periods. It can be seen that the combined application of nitrogen, phosphorus, and potassium fertilizers is most

Table 1. Effects of nitrogen, phosphorus, and potassium on branch number and the number of effective branch n

Treatment	HS			AS			MS		
	BN	NEB	EBR	BN	NEB	EBR	BN	NEB	EBR
NPK	16.33 a	13.00 a	79.60 b	22.67 a	15.50 a	68.40 a	21.67 a	15.00 a	69.20 a
PK	16.00 a	11.50 a	71.80 c	21.33 a	12.00 b	56.30 b	19.67 b	8.00 c	40.70 c
NK	15.00 a	5.50 b	36.70 d	15.00 c	8.50 c	56.70 b	15.67 c	6.50 d	41.50 c
NP	12.00 b	12.00 a	100 a	18.00 b	12.50 b	69.40 a	20.67 ab	13.00 b	62.90 b

Note: HS: Heading stage; AS: Anthesis stage; MS: Mature stage; BN: Branch number; NEB; The number of effective branch; EBR:effective branching rate. Different lowercase letters in the figure indicate significant differences at the 0.05 level ($P<0.05$), the same below.umber at different growth stages of quinoa (pcs).

Table 2. Effects of nitrogen, phosphorus, and potassium on nitrogen accumulation at different growth stages of quinoa (kg/hm^2).

Treatment	SS	BS	HS	AS	MS
NPK	2.79 a	20.11 a	53.59 a	91.59 a	96.52 a
PK	1.28 b	12.17 b	22.03 b	28.31 c	43.30 b
NK	1.27 b	5.91 c	22.40 b	34.37 c	46.52 b
NP	2.59 a	19.59 a	53.98 a	76.72 b	94.93 a

Note: SS:Seedling Stage ; BS:Branching Stage; HS:Heading Stage; AS:Anthesis Stage; MGS:Mid Grain-Filling Stage; MS:Maturity Stage The different lowercase letters showed significant differences at the 0.05 level (P<0.05).

conducive to the nitrogen accumulation of quinoa plants. The lack of nitrogen or phosphorus fertilizers affects the absorption of nitrogen by plants, and the lack of potassium fertilizers has little effect.

Effects of Nitrogen, Phosphorus, and Potassium Fertilizers on Yield Formation and Nitrogen Use Efficiency of Quinoa

With the development of the growth process, the spike length and width of the main spike of quinoa increased gradually after heading, and reached the maximum at maturity, with a spike length of 17.77±0.06 cm and a spike width of 3.49±0.08 cm (Table 3). Compared with other fertilization methods, nitrogen, phosphorus, and potassium fertilizers can increase the ear length of quinoa at the heading stage by 1.45±0.30 cm and the ear width by 0.16-0.31 cm, increase the ear length by 0.24±0.05 cm and the ear width by 0.38±0.17 cm at the flowering stage except nitrogen and phosphorus fertilizers, and increase the ear length by 5.63±1.22 cm and 1.37±0.11 cm at the maturity stage. There were significant differences between nitrogen, phosphorus, and potassium fertilizers and between nitrogen and potassium fertilizers. The difference was greater with nitrogen and potassium fertilizers, but there was no significant difference with nitrogen and phosphorus fertilizers.

At the maturity stage, the number of grains per plant was 3220.56±173.74, the 1000-grain weight was 1.66±0.06 g, and the grain weight per plant was 5.29±0.46 g (Table 4). The combined application of nitrogen, phosphorus, and potassium fertilizers can increase the number of grains per plant compared with phosphorus and potassium fertilizers, nitrogen and potassium fertilizers, and has significant differences with nitrogen and potassium fertilizers. Compared with nitrogen and phosphorus fertilizers, the number of grains per plant is reduced, but the difference between treatments is not significant. Compared with other fertilization methods, the combined application of nitrogen, phosphorus, and potassium fertilizers could increase the 1000-grain weight by 0.34±0.02 g, grain weight per plant by 1.41±0.23 g, and the overall nitrogen utilization efficiency. There were significant differences between nitrogen, phosphorus, and potassium fertilizers and between nitrogen and potassium fertilizers. There was a greater difference between nitrogen and potassium fertilizers, but there was no significant difference between nitrogen and phosphorus fertilizers. It can be seen that the combined application of nitrogen, phosphorus, and potassium fertilizers is beneficial to increase the number of grains per spike and 1000-grain weight, thereby increasing yield and nitrogen use efficiency.

Effects of Nitrogen, Phosphorus, and Potassium Fertilizers on Nutritional Quality of Quinoa Grains at the Maturity Stage

Free Amino Acid Content in Grain

The total free amino acid content of quinoa was 4088.35±454.68 mg/g, of which the essential amino acid

Table 3. Effects of nitrogen, phosphorus, and potassium on panicle length and panicle width at different growth stages.

Treatment	HS		AS		MS	
	SL (cm)	SW (cm)	SL (cm)	SW (cm)	SL (cm)	SW (cm)
NPK	6.57 a	1.61 a	12.63 a	2.26 a	17.88 a	3.51 a
PK	5.57 b	1.45 c	8.64 b	2.03 b	14.13 b	3.37 b
NK	4.67 c	1.30 d	6.84 c	1.54 c	9.78 c	1.92 c
NP	6.47 a	1.52 b	12.92 a	2.31 a	17.65 a	3.46 a

Note: HS: Heading stage; AS: Anthesis stage; MS: Mature stage; SL: Spike length; SW: Spike width. The different lowercase letters showed significant differences at the 0.05 level (P<0.05).

Table 4. Effects of nitrogen, phosphorus, and potassium on yield composition and nitrogen efficiency of quinoa.

Treatment	NGPS	TGW (g)	GWPP	NUE (%)
NPK	3384.88 a	1.84 a	6.22 a	10.35 a
PK	3315.61 a	1.53 b	5.07 b	5.74 c
NK	2972.82 b	1.47 b	4.36 c	7.13 b
NP	3469.29 a	1.78 a	6.17 a	10.26 a

Note: NPK: Nitrogen, phosphorus, and potassium complete application, PK: Nitrogen deficiency, NK: Phosphorus deficiency, NP: Potassium deficiency; NGPS: Number of grains per spike;TGW: Thousand-grain weight; GWPP: Grain weight per plant; NUE : Nitrogen use efficiency. The different lowercase letters showed significant differences at the 0.05 level ($P<0.05$).

content was 1629.93±260.68 mg/g, and the non-essential amino acid content was 2101.14-3099.29 mg/g (Table 5). Compared with other fertilization methods, the combined application of nitrogen, phosphorus, and potassium fertilizers can increase the content of essential amino acids by 284.10±52.75 mg/g, non-essential amino acids by 2230.71±362.70 mg/g, and total free amino acids by 2414.65±428.30 mg/g. The contents of essential amino acids, non-essential amino acids, and total free amino acids were significantly different between nitrogen, phosphorus, and potassium fertilizers and nitrogen and potassium fertilizers, especially phosphorus and potassium fertilizers. The difference was greater, but not significant for nitrogen and phosphorus fertilizers. In summary, nitrogen, phosphorus, and potassium (NPK) had the most significant effect on the improvement of most amino acid components. Nitrogen and potassium (NK) only significantly increased histidine levels. Nitrogen and phosphorus (NP) significantly increased some essential amino acids and non-essential amino acids, whereas phosphorus and potassium (PK) did not significantly increase amino acids. In addition, the combined application of nitrogen, phosphorus, and potassium fertilizers can also increase the proportion of essential amino acids, total free amino acids, and essential amino acids/non-essential amino acids, among which essential amino acids/non-essential amino acids can increase by 0.16±0.04 mg/g compared with other fertilization methods. Nitrogen, phosphorus, and potassium fertilizers are beneficial for the accumulation of free amino acids in quinoa grains, and the effect of a lack of nitrogen fertilizer is more obvious.

Grain Nutritional Quality

The protein content of quinoa grains was 16.62±1.46%, the starch content was 63.31±1.28%, the fat content was 4.66±0.52%, the crude fiber content was 4.06±0.52%, and the ash content was 4.06±0.52% (Table 6). Compared with other fertilization methods, the combined application of nitrogen, phosphorus, and potassium fertilizers can increase protein content by 2.27±0.55 percentage points, starch by 5.05±1.00 percentage points, fat by 1.12±0.23 percentage points, crude fiber by 0.59±0.24 percentage points, and ash by 0.88±0.25 percentage points. There were significant differences in protein, fat, crude fiber, and ash between nitrogen, phosphorus, and potassium fertilizers and phosphorus and potassium fertilizers, nitrogen and potassium fertilizers, especially phosphorus and potassium fertilizers. The difference was greater, but the difference was not significant with nitrogen and phosphorus fertilizers. There was a significant difference between starch nitrogen, phosphorus, and potassium fertilizers and phosphorus and potassium fertilizers, and there was no significant difference between nitrogen and potassium fertilizers and nitrogen and phosphorus fertilizers. It can be seen that nitrogen, phosphorus, and potassium fertilizers are beneficial to the formation of quinoa grain nutritional quality, and the effect of a lack of nitrogen fertilizer is more obvious.

Discussion

The root system is a vital organ for absorbing soil moisture and nutrients and exhibits high plasticity in various environments [25]. Nitrogen fertilizers promote root growth, increase root hair density, and enhance the physiological functions of roots. Appropriate nitrogen application significantly increases the total root dry mass, root length, root surface area, and root volume in maize [26]. Suitable phosphorus application depth and rate can effectively increase root length, root diameter, root active surface area, and root volume in tartary buckwheat, rice, wheat, and alfalfa, whereas low phosphorus stress significantly reduces root length, surface area, and diameter [27-29]. Potassium fertilizer significantly increases the total root length of peanuts and promotes the total root volume and total surface area to varying degrees under different moisture conditions [30]. This study demonstrated that compared to the NK, NP, and PK treatments, the combined application of NPK fertilizers significantly enhanced the root system characteristics of quinoa from the branching stage to the maturity stage. Specifically, it increased total root length by 50.00±11.3%, root surface area by 32.8±6.4%, total root volume by 74.8±0.8%, and the number of root tips by 76.2±1.1%. This effect was more pronounced than that of the NK treatment. This indicates that the absence

Table 5. Effects of nitrogen, phosphorus, and potassium on free amino acid and protein components in quinoa grains.

Amino acid components		NPK	PK	NK	NP
Essential amino acid (mg/g)	Val	431.91 a	235.25 b	288.99 b	468.11 a
	Thr	113.63 a	47.85 c	80.61 b	144.43 a
	Ile	261.74 b	143.21 d	184.86 c	318.26 a
	Leu	353.62 b	193.52 d	252.93 c	423.43 a
	Lys	213.23 a	145.21 b	191.43 a	197.03 a
	Met	34.64 a	15.75 c	25.72 b	36.93 a
	His	78.39 ab	59.94 c	84.29 a	62.96 bc
	Phe	301.32 a	171.25 b	217.96 b	341.76 a
	Trp	109.94 a	65.87 c	90.06 b	115.11 a
Nonessential amino acid (mg/g)	Arg	458.52 a	301.48 b	519.66 a	329.15 b
	Gln	626.32 a	530.57 ab	568.78 ab	363.29 b
	Glu	612.8 a	382.29 b	468.34 b	652.42 a
	Asn	100.01 a	88.56 a	102.25 a	94.61 a
	Ala	284.52 a	156.46 c	200.96 b	327.45 a
	Pro	154.04 a	98.16 b	172.52 a	174.88 a
	Tyr	602.58 a	363.17 c	466.93 b	665.39 a
	GABA	63.41 a	45.57 b	72.81 a	34.79 b
	Gly	162.78 a	103.38 c	132.66 b	153.37 ab
	Asp	406.79 a	242.36 b	299.28 b	466.7 a
	Ser	195.16 ab	127.92 c	164.73 bc	222.42 a
	Orn	34.95 a	24.39 b	26.61 b	28.31 b
Essential amino acid (mg/g)		1898.43 a	1077.85 c	1416.87 b	2108.01 a
Nonessential amino acid (mg/g)		3099.29 a	2101.14 b	2728.59 a	2847.38 a
Total Free Amino Acids (mg/g)		4997.71 a	3178.99 c	4145.46 b	4955.39 a
EAA/TFA		0.38 b	0.34 c	0.34 c	0.43 a
EAA/NA		0.61 b	0.51 c	0.52 c	0.74 a

Note: EAA:Essential amino acid;NA:Nonessential amino;TFA:Total Free Amino;The different lowercase letters showed a significant difference at the 0.05 level, *$P<0.05$; **$P<0.01$.

of nitrogen, phosphorus, or K fertilizers adversely affects quinoa root growth and development to some extent, with the lack of phosphorus fertilizer (i.e., the NK treatment) having the greatest impact [19]. Different phosphorus levels can significantly regulate quinoa root development at the seedling stage. Phosphorus deficiency strongly inhibits root growth by disrupting the homeostasis of hormonal signaling pathways that regulate root development [31]. The marked inhibitory effect of phosphorus (P) deficiency on quinoa root growth arises from the loss of P's core functions in plant physiological metabolism, signal regulation, and structural construction, which explains why root morphological traits under P-deficient conditions were significantly poorer than those under other treatments. As a key component for ATP synthesis, P deficiency reduces root cell energy supply, suppresses the expression of enzymes related to nucleic acid synthesis and cell division, and thereby inhibits root cell proliferation and elongation [31]. It also disturbs the polar transport and signaling balance of auxin, restraining lateral root emergence and root hair elongation [32], decreases the activities of key enzymes (e.g., plasma membrane H^+-ATPase, acid phosphatase), reduces nitrogen (N) and potassium (K) uptake (forming a vicious cycle of growth inhibition), and weakens the mobilization of soil organic phosphorus [33]. Additionally, P deficiency alters cell wall structure by inhibiting pectin methylesterase activity, which increases the mechanical resistance to root cell expansion and further restricts root growth

Table 6. Effects of nitrogen, phosphorus, and potassium on nutrient composition of quinoa grains.

Treatment	Protein (%)	Starch (%)	Fat (%)	Fiber (%)	Ash (%)
NPK	17.88 a	65.58 a	5.44 a	4.84 a	4.55 a
PK	13.56 c	61.03 b	3.87 b	3.28 c	3.17 c
NK	14.69 b	64.86 a	4.31 b	3.95 b	3.67 b
NP	17.12 a	64.62 a	5.12 a	4.67 a	4.38 a

Note:The different lowercase letters indicate significant differences at the 0.05 level (P<0.05). 6. Effects of nitrogen, phosphorus, and potassium on nutrient composition of quinoa grains.

[34]. Furthermore, the study showed that, as growth progressed, the NPK treatment resulted in a greater increase in root system parameters compared to other treatments, especially with the NK treatment (lacking phosphorus). This suggests that phosphorus fertilizer initiates root growth, and good early growth ensures favorable development in the middle and late stages.

Dry matter accumulation has a direct impact on yield, and an increase in dry matter accumulation serves as a foundation for yield improvement. The results of this study indicate that compared to other fertilization methods, the combined application of NPK fertilizers significantly increased plant height and dry matter accumulation at various growth stages of quinoa. At maturity, plant height increased by 20.0±5.0 cm, and dry matter accumulation reached 71.6±24.7%, with the effect being particularly more significant compared to the NK treatment. Nitrogen, phosphorus, and potassium fertilizers all promote the aboveground growth of quinoa, whereas the absence of any one of these elements inhibits its growth to some extent. The lack of phosphorus fertilizer had the most significant impact, primarily because of its effect on underground root growth. Some studies have found that nitrogen application rate greatly influences nutrient allocation in quinoa and is beneficial for increasing yield [35, 36].

Crop nutrient uptake serves as the foundation for dry matter accumulation. The nitrogen uptake of quinoa plants increases with the nitrogen application rate, but the rate of increase gradually slows, suggesting that nitrogen uptake approaches a threshold and further increasing nitrogen fertilizer will not enhance uptake [20]. The uptake of nitrogen, phosphorus, and K by quinoa plants initially increased and then decreased with increasing rates of phosphorus and K application [19]. Phosphorus application rate can improve phosphorus use efficiency in quinoa, while excess application reduces it. The results of this study revealed that, compared to other fertilization methods, NPK treatment increased plant nitrogen accumulation in quinoa at various growth stages. Significant differences were observed between NPK treatment and both NK and PK treatments. A significant difference was also observed between the NPK and NP treatments at the flowering stage, although the differences were not significant at other stages. This indicates that the combined application of NPK fertilizers is most conducive to nitrogen accumulation in quinoa plants. The absence of either nitrogen or phosphorus fertilizer affects plant nitrogen uptake, whereas the lack of K fertilizer has a lesser impact.

The effective branching of quinoa is a crucial yield component, and its extent significantly influences the grain yield [37, 38]. This study demonstrated that, compared to the phosphorus-potassium (PK) and nitrogen-potassium (NK) fertilizer treatments, the combined application of nitrogen, phosphorus, and potassium fertilizers (NPK) increased the effective branching rate of quinoa at the flowering stage by approximately 12.1 and 11.7 percentage points, respectively, and at the maturity stage by approximately 28.5 and 27.7 percentage points, respectively, with statistically significant differences observed among all treatments. In contrast, no significant difference in the effective branching rate was noted at the flowering stage when compared with the nitrogen-phosphorus (NP) fertilizer treatment. Evidently, both nitrogen and phosphorus fertilizers promoted quinoa branching and enhanced the effective branching rate, thereby increasing the number of effective branches, whereas the absence of potassium fertilizer had no significant impact on effective branching.

Nitrogen, phosphorus, and potassium fertilizers significantly regulate the crop population structure and yield [39]. Panicle traits form the basis of yield formation, and there is a synergistic improvement between yield components such as the number of grains per panicle, 1000-grain weight, grain weight per plant, and nitrogen use efficiency [22]. With nitrogen application of 160 kg/hm^2, the number of grains per panicle in quinoa increased by 30.01% compared with the control, indicating that nitrogen fertilizer promotes panicle development in quinoa. This study found that at maturity, the panicle length of the PK treatment (lacking nitrogen) was shortened by 3.75 cm, and the grain weight per plant was reduced by 1.15 g compared to the NPK treatment [22]. These results indicate that nitrogen deficiency reduces the number of grains per panicle and yield, thereby confirming the critical role of nitrogen in panicle elongation and grain filling [19]. The research findings demonstrated that phosphorus improves nutrient uptake by the root system, indirectly enhancing dry matter accumulation in the panicle.

The present experimental study revealed that the NK treatment (lacking phosphorus) exhibited the poorest panicle traits, with a panicle length of only 9.78 cm at maturity, which is 8.1 cm shorter than that of the NPK treatment. The grain weight per plant was 1.86 g less, and the nitrogen use efficiency was significantly lower at 7.13% (compared to that of 10.35% in the NPK treatment [19]). Phosphorus deficiency affects the translocation of nutrients to the panicle, indicating that phosphorus supports panicle development and yield formation by maintaining nutrient supply through the root system [20]. Potassium fertilizer increases quinoa biomass by enhancing the translocation of photosynthetic products to grains. This study found that compared to the NP treatment (lacking potassium), the NPK treatment increased the 1000-grain weight by 0.06 g and the grain weight per plant by 0.05 g. Although these differences were not statistically significant, they still suggest that potassium fertilizer promotes grain filling. The yield indicators of the NPK treatment were significantly higher than those of the two-factor treatment. The grain weight per plant increased by 35.2±7.1% and 42.7±0.5% compared with the PK and NK treatments, respectively, and nitrogen use efficiency improved by 45.4±0.1%. This demonstrates that the synergistic action of these three factors enhances yield: nitrogen promotes panicle development, phosphorus ensures nutrient translocation, and potassium improves the allocation of photosynthetic products. Their coordination synergistically increased the number of grains per panicle, 1000-grain weight, and grain weight per plant while also improving nitrogen use efficiency and reducing nutrient waste. In this study, the effect of potassium (K) deficiency on the growth, yield, and nutritional quality of quinoa cultivar Jingli 1 was relatively mild. This result is closely related to the nutritional demand characteristics of quinoa itself: quinoa is a K-loving crop, but its total K requirement is lower than that of nitrogen (N). There are significant differences in barren tolerance among different quinoa cultivars, and the total K uptake of some main cultivars during the whole growth period is only 330-340 kg/hm², showing strong tolerance to K stress [40]. Meanwhile, the available K content of the soil used in this experiment was 85.6 mg/kg, and the inherent K supply capacity of the soil could compensate for the nutrient loss caused by K deficiency treatment to a certain extent, further weakening the inhibitory effect of K deficiency on quinoa growth and development, hence only a mild effect was observed. In summary, the "mild" effect of K deficiency in this study is the combined result of the low K requirement of the quinoa cultivar itself and the sufficient K supply in the subsoil. Future studies can further distinguish the relative contributions of the two factors by setting strict K-free controls and soil background detection.

Nitrogen, phosphorus, and potassium fertilizers are essential elements for plant growth and development and are closely related to the formation of quality traits such as proteins, starches, and fats [41]. Wheat grains contain soluble proteins (including albumin and globulin) rich in essential amino acids such as lysine and tryptophan, which determine the nutritional quality of wheat. Previous studies investigated the effects of nitrogen, phosphorus, and potassium fertilizer application rates on soybean yield and quality. Their study on three soybean varieties found that NPK fertilizer application effectively improved soybean quality, notably increasing protein content and the combined protein and fat content [41]. Compared to the no-fertilizer treatment, protein content increased by 3.11%, whereas the increase in fat content was not significant [42]. NPK fertilizer application significantly increased both the protein content and water-soluble protein content in soybeans, albeit by only 0.35±0.15% and 0.20±0.08%, respectively. Changes in soybean quality are constrained by multiple factors, including variety, moisture, temperature, climate, and region. Consequently, conclusions from various studies on the effect of fertilization on plant quality may vary. However, nitrogen, phosphorus, and K fertilizers invariably influence the intrinsic quality of plants to different degrees [43, 44]. The results of this experiment indicate that compared to other fertilization methods, the combined application of NPK fertilizers increased protein content by 2.77±0.78 percentage points, starch by 3.05±0.75 percentage points, fat by 1.12±0.23 percentage points, crude fiber by 1.09±0.24 percentage points, and ash content by 1.04±0.17 percentage points. Among these, the absence of nitrogen fertilizer had a particularly significant impact on the nutritional quality of quinoa. The combined application of NPK fertilizers also increased the ratios of essential amino acids to total free amino acids, and essential amino acids to non-essential amino acids. Specifically, the ratio of essential amino acids to non-essential amino acids increased by 0.06±0.04 mg/g compared to other fertilization methods. This demonstrates that NPK fertilizer application is beneficial for the accumulation of free amino acids in quinoa grains, and that the absence of nitrogen fertilizer has a more pronounced negative effect. Nitrogen deficiency, phosphorus deficiency, and potassium deficiency have a significant inhibitory effect on the growth and development of quinoa. Among them, nitrogen deficiency will lead to plant dwarfing, leaf yellowing, biomass and grain yield decrease, and reduce protein and amino acid accumulation; phosphorus deficiency mainly inhibited root growth, reduced root length, root surface area, and nutrient absorption capacity, and had the most obvious limitation on biomass and yield. Potassium deficiency mainly affects grain filling, so that the 1000-grain weight is slightly reduced, and the overall effect is relatively small [45]. Under the condition of raw soil potting, due to the lack of soil basic nutrients, the symptoms of nutrient deficiency are prominent, which can clearly reveal the limiting effect of nitrogen, phosphorus, and potassium on quinoa; in the conventional field, after long-term tillage and fertilization, the soil has relatively sufficient basic nutrients, which can buffer the symptoms of nutrient deficiency, so that the performance of nitrogen, phosphorus, and potassium deficiency is weaker than that of the pot experiment, and it is difficult to accurately identify the core limiting factors. At present, most of the related studies have confirmed the negative effects of nutrient deficiency on quinoa based on pot experiments. However, in the farmland soil with a high fertility level, the real effect, performance degree, and suitable fertilization amount of nutrient deficiency are still unclear. Previous studies have shown that the immature subsoil has few nutrients without tillage and fertilization, which can minimize the interference of original soil nutrients, and more clearly see the effects of nitrogen, phosphorus, and potassium deficiency on the growth, yield, and quality of quinoa. It is an ideal experimental condition for studying the mechanism of nutrient stress in quinoa. Because of long-term fertilization management, there are a lot of nitrogen, phosphorus, and potassium in ordinary farmland soil, which will cover the performance of nutrient deficiency, and it is not easy to accurately find out the key nutrients that limit the growth of quinoa [46]. The effect of nutrient deficiency on quinoa in this study is consistent with the existing research results of nutrient stress under low-fertility soil. When studying tartary buckwheat on low-phosphorus poor soil, it was found that poor soil would significantly amplify the inhibitory effect of phosphorus deficiency on crop growth [26]; it was also proved that the raw soil of loess parent material will enlarge fertilization at the same time because of its low nutrient content [47].

Conclusions

The combined application of phosphorus and potassium fertilizers without nitrogen fertilizer affected dry matter and nitrogen accumulation in the aboveground parts of quinoa. The combined application of nitrogen and potassium fertilizers without phosphorus fertilizer inhibits root growth, thereby reducing plant height and dry matter accumulation in the aboveground parts, which significantly affects yield and quality. In contrast, the combined application of nitrogen and phosphorus fertilizers without K had a negligible effect on quinoa growth. However, the combined application of nitrogen, phosphorus, and K fertilizers most effectively promoted root and aboveground growth, enhanced nitrogen absorption and accumulation, and subsequently improved quinoa grain yield, amino acids, protein, fat, and other nutritional qualities.

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Conflicts of Interest

The authors declare no conflict of interest.

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