


Enhancing of nutritional properties of quinoa fermented by probiotics

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Abstract

Fermentation of quinoa by probiotics provides higher nutritional value and can be considered as a significant source of bioactive compounds and alive probiotics for the human body. Moringa leaves powder (MLP) at the levels of 0.25 and 0.50% were used as an additional prebiotic source to supply quinoa fermentation by *Lactobacillus plantarum* ATCC 14917 and *Lactobacillus delbrueckii ssp. Bulgaricus* EMCC 11102 and produce healthier quinoa products. The results indicated that supplementation of fermented quinoa products with MLP at both levels increased its contents of free phenolics and water extractable arabinoxylans as well as enhanced its antioxidant activity and phytate degradation. Fermented quinoa products with 0.50% MLP showed better chemical properties than fermented quinoa products with 0.25% MLP. Furthermore, supplementation of fermented quinoa products with MLP (0.50%) displayed positive effects on blood glucose levels of the experimental rats. Further studies are required on the test of more kinds of probiotics with different concentration of MLP in quinoa fermentation in the future.

Keywords Quinoa fermentation · Moringa · Antioxidant · Probiotics · Prebiotics · Phytate degradation

1 Introduction

Quinoa (*Chenopodium quinoa*) is a pseudo-cereal plant domesticated in the Indian region 5000 years ago [1]. It is known for its high nutritional value and high contents of essential amino acids, minerals and dietary fibres. Due to its significant content of phenolic compounds, quinoa showed antioxidant activity which associated with many health benefits [2, 3].

Probiotic bacteria are mainly used in dairy products and show the biggest share of the probiotic food market [4]. However, some disadvantages of dairy products containing probiotics have been recorded for many consumers worldwide such as lactose intolerance; allergy to β -caseine in cow's milk; high content of cholesterol and saturated fatty acids in dairy products as well as high costs of milk [5]. Probiotic cereal foods may be produced as a good alternative to avoid drawbacks of fermented dairy products [4]. Several studies have evaluated the fermentation of quinoa by probiotics for production of fermented quinoa products with higher healthy benefits [6–8].

Moringa oleifera leaves powder (MLP) produce numerous healthy benefits and have good amounts of crude protein, crude fiber, extract ether, carbohydrates, energy, minerals, vitamins, β -carotene and polysaccharides [9, 10]. Also, MLP displayed a prebiotic effect may be due to its significant content of prebiotic compounds such as oligosaccharides [11, 12]. Also, the high antioxidant activity and phenolic compounds of MLP were reported by many previous studies [13–15].

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This is the first study evaluates the effect of supplementation with MLP as a source of prebiotics to produce healthier fermented quinoa products. The changes in the free phenolic content and antioxidant activity as well as the levels of phytate and water extractable arabinoxylans of fermented quinoa products were determined during storage period. Also, the effects of MLP (0.50%) on the blood glucose levels of the experimental rats were estimated during the feeding period for 30 days.

2 Materials and methods

2.1 Materials

2.1.1 Raw materials and reagents

Quinoa seeds were purchased from Agricultural Research Center, Giza, Egypt. Moringa (*Moringa oleifera*) leaves were obtained from a local farm located in Albalyana city, Sohag, Egypt, and sugar was purchased from a local market in Assiut city, Egypt. Chemicals used in the analytical methods were produced by sigma chemical co. (St. Louis, MO, USA) and obtained from El-Gomhouria Trading Chemicals and Drugs Company, Assiut city, Egypt. The reagents used in the biological experiment were purchased from Spectrum Company in Assiut city, Egypt.

2.1.2 Probiotic strains

Lactobacillus plantarum ATCC 14917 and *L. delbrueckii ssp. Bulgaricus* EMCC 11102 were purchased from Microbiological Resources center (Cairo MIRCEN) Ain Shams University, Cairo, Egypt.

2.2 Methods

2.2.1 Preparation of quinoa fermentation

2.2.1.1 Preparation of raw materials and quinoa blends Grain quinoa seeds were washed and soaking for 24 h in water which was discarded and changed every 3 h. The soaked quinoa seeds was dried at 60 °C for 8 h and milled to get the whole quinoa flour (WQF). The Moringa leaves were dried and milled to produce MLP, which stored in a cool dry place until the experimental work. The blends of WQF, MLP and sugar were prepared and tap water was added to produce the final formulas as follow in Table 1:

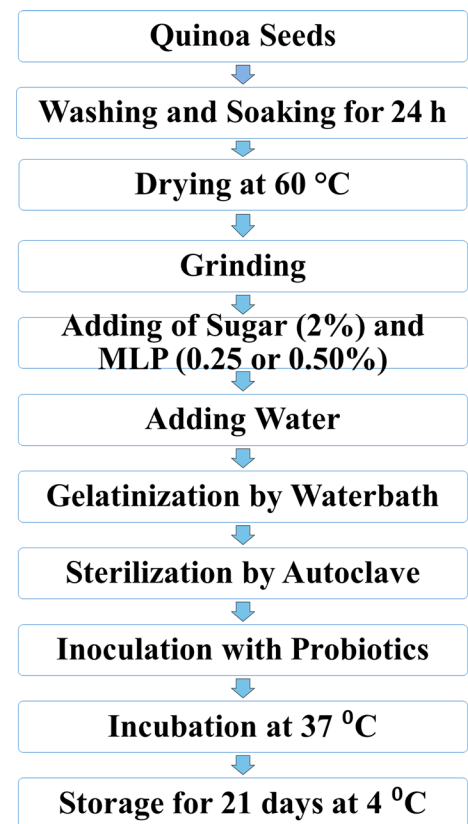
In the next step, the mixtures were gelatinized using water bath and autoclaved at 121 °C for 15 min Fig. 1.

Table 1 Formulas of fermented quinoa products

	Quinoa %	Sugar %	MLP %	Water %
Control	10.00	2.00	–	88.00
FQP	10.00	2.00	–	88.00
FQP1	10.00	2.00	0.25	87.75
FQP2	10.00	2.00	0.50	87.50
FQD	10.00	2.00	–	88.00
FQD1	10.00	2.00	0.25	87.75
FQD2	10.00	2.00	0.50	87.50

Control, Non-fermented quinoa; FQP, fermented quinoa by *L. plantarum* ATCC 14917; FQP1, fermented quinoa with 0.25% MLP by *L. plantarum* ATCC 14917; FQP2, fermented quinoa with 0.5% MLP by *L. plantarum* ATCC 14917; FQD, fermented quinoa by *L. delbrueckii ssp. Bulgaricus* EMCC 11102; FQD1, fermented quinoa with 0.25% MLP by *L. delbrueckii ssp. Bulgaricus* EMCC 11102; FQD2, fermented quinoa with 0.5% MLP by *L. delbrueckii ssp. Bulgaricus* EMCC 11102

Fig. 1 Production of fermented quinoa products



2.2.1.2 Inoculant strains and quinoa fermentation conditions The Lactobacillus bacteria strains were activated by inoculating in sterile MRS broth (9 mL) and incubation at 37 °C for 24 h. The cells were separated from the broth by centrifuging, and re-suspended in sterile saline solution (9 mL) with final concentration 10^8 CFU/mL [16].

The 24 h activated culture of probiotics (10^8 CFU/mL) were added to the previous quinoa mixtures by a concentration of 1%. A control sample without inoculation of bacteria was prepared. All treatments were incubated at 37 °C for 24 h for *L. plantarum* fermentation and 8 h for *L. delbrueckii ssp. Bulgaricus* fermentation. The fermented quinoa products were stored after fermentation at 4 ± 1 °C for 21 days. Chemical and microbiological properties of fermented quinoa products were estimated at 0, 7, 14 and 21 days.

2.2.2 Determination of titrable acidity

Total acidity was determined by titration method (10 g of fermented products with 90 mL distilled water, using 0.1 N NaOH solution and phenolphthalein as indicator) according to Coda et al. [17].

2.2.3 Determination of free phenolics and antioxidant activity

2.2.3.1 Extract preparation Free phenolic compounds were extracted from samples using the method of Acosta-Estrada et al. [18]. Briefly, 1 g of fermented quinoa samples were mixed with 10 mL of chilled ethanol/water (80:20 v/v) and shaken at 250 rpm for 10 min at 25 °C, then centrifuged at 3000 g for 10 min. The supernatant was recovered and the extraction repeated once. Supernatants were pooled and evaporated at 50 °C and 20 mbar. The resulting extracts were stored at – 20 °C until use.

2.2.3.2 Determination of free phenolic content Free phenolic contents were estimated in the previous ethanolic extracts of fermented quinoa products by Folin-Ciocalteu method according to Jaramillo-Flores et al. [19]. An 100 μ L of ethanolic extract was mixed with 900 μ L of Folin-Ciocalteu reagent (diluted 1:10 with distilled water) and was allowed to stand for 5 min at room temperature; 0.75 mL of sodium bicarbonate solution (7%) was added to the mixture and vortexed for 30 s, and allowed to stand at room temperature for 90 min. The absorbance was measured at 725 nm using a spectrophotometer

(6505 UV/Vis, Jenway LTD., Felsted, Dunmow, UK). A calibration curve of gallic acid (ranging from 0 to 1 mg/mL) was prepared and tested under similar conditions. All values were expressed as mean (mg of Gallic acid equivalents/100 g wet weight fermented quinoa).

2.2.3.3 Determination of DPPH radical scavenging activity Free radical scavenging activity was determined by the 1,1-diphenyl-2-picrylhydrazyl (DPPH) assay according to Elfahri et al. [20] with slight modification. Briefly, 900 μL of the DPPH reagent (0.1 mM DPPH dissolved in 95% methanol) was added to 100 μL of fermented product extracts in glass test tubes. The samples were shaken vigorously and incubated for 30 min in the dark at room temperature. The absorbance of the incubated samples was measured at 517 nm. The percentage of radical scavenging activity was expressed as scavenging (%) using Eq.:

$$\% \text{ Scavenging} = \frac{\text{Abs blank} - \text{Abs sample}}{\text{Abs blank}} \times 100$$

2.2.4 Determination of phytate content

The phytic acid (IP_6) was determined in terms of its phosphorous content, using the method described by [21]. Phytic acid was calculated from phytate phosphorus from the weight ratio of phosphorus atoms per molecule of IP_6 (1:3.52).

2.2.5 Quantification of water extractable arabinoxylans (WEAX)

The WEAX of fermented products was determined according to [22], with slight modification. Samples of fermented quinoa products (1 g) were extracted with distilled water (20 mL) under constant agitation and centrifuged for 10 min at 4000 rpm. Then, 100 μL of supernatant, 100 μL of distilled water and 2 mL of daily prepared working reaction solution (1 g of phloroglucinol dissolved in 5 mL of pure ethanol, 2 mL of hydrochloric acid, 110 mL of glacial acetic acid and 1 mL of a 17.5 g L^{-1} glucose solution) were added to stoppered glass tubes (12 mL, 16 \times 100 mm). The tubes were located for 25 min in a boiling water bath, next immediately cooled in the ice. The absorbance was measured using an UV-Vis spectrophotometer at 552 nm and 510 nm. D-(+)-Xylose was used as standard for the calibration curve (0.05–30 mg Kg^{-1}). Finally, the WEAX content was calculated subtracting the absorbance value at 510 nm, which corresponds to hexose interferences, from the absorbance value at 552 nm and the obtained value was compared with the regression equation.

2.2.6 Biological experiment

2.2.6.1 Adaptation and distributing of experimental animals The animals were housed as groups in wire cages under the normal laboratory conditions and fed on basal diets for 10 days as adaptation period. The rats were distributed to 4 groups containing control group and fed during experimental period (30 days).

2.2.6.2 Experimental design The rats were randomly divided into 4 groups (1, 2, 3 and 4), each group was contained 10 rats. Group 1 (control): fed on basal diet, Group 2: fed on basal diet containing of 30% of non-fermented quinoa, Group 3: fed on basal diet containing of 30% of FQP2, Group 4: fed on basal diet containing of 30% of FQD2.

2.2.6.3 Blood sampling At the day of blood sampling collection, rats were fasted overnight and anesthetized. Blood samples were collected from the retro-orbital plexus from all animals of each group into clean, dry and labeled tubes. The tubes were centrifuged at 3500 rpm for 15 min to separate the blood serum, which used to estimation of biochemical parameters [23].

2.2.6.4 Determination of fasting and postprandial blood glucose The concentrations of fasting and postprandial blood glucose were determined according to Lopes et al. [24] using enzymatic kits (SPECTRUM diagnosis, Germany).

2.2.7 Statistical analysis

Basic statistics and analysis of variance (ANOVA) were performed to data analysis and test the significance within replications and between treatments by using IBM SPSS software version 22. Duncan test was used to determine the differences among the means at the significance level of 0.05%.

Table 2 Changes in titrable acidity % of fermented quinoa products during storage at 4 °C ± 1 for 21 days

Treatments	Storage periods (days)			
	0	7	14	21
Control	0.27 ^{Da}	0.27 ^{Da}	0.28 ^{Da}	0.28 ^{Ca}
FQP	0.38 ^{Cc}	0.40 ^{Cc}	0.44 ^{Cb}	0.49 ^{Ba}
FQP1	0.40 ^{BCd}	0.43 ^{ABc}	0.46 ^{ABCb}	0.49 ^{ABa}
FQP2	0.41 ^{ABc}	0.43 ^{ABc}	0.47 ^{ABb}	0.51 ^{ABa}
FQD	0.40 ^{BCc}	0.42 ^{BCc}	0.45 ^{BCb}	0.50 ^{ABa}
FQD1	0.42 ^{ABc}	0.45 ^{Ac}	0.48 ^{ABb}	0.51 ^{ABa}
FQD2	0.43 ^{Ac}	0.45 ^{Ac}	0.49 ^{Ab}	0.52 ^{Aa}

Means within a column with different superscript capital letters are significantly different ($P > 0.05$); means within a row with different superscript small letters are significantly different ($P > 0.05$)

Table 3 Changes in free phenolic content (mg GAL /100 g ww) of fermented quinoa during storage at 4 °C ± 1 for 21 days

Treatments	Storage periods (days)			
	0	7	14	21
Control	19.96 ^{Ea}	20.15 ^{Ea}	21.06 ^{Ea}	21.54 ^{Da}
FQP	23.17 ^{DEb}	26.04 ^{Cab}	27.42 ^{Da}	27.60 ^{Ca}
FQP1	28.04 ^{BCb}	32.11 ^{Ba}	33.96 ^{Ca}	34.15 ^{Ba}
FQP2	32.85 ^{Ab}	36.81 ^{Aa}	38.14 ^{ABa}	37.24 ^{ABa}
FQD	24.88 ^{CDa}	24.50 ^{Ca}	25.69 ^{Da}	24.92 ^{CDa}
FQD1	29.15 ^{Bb}	32.93 ^{Ba}	34.79 ^{B^{Ca}}	35.10 ^{Ba}
FQD2	34.24 ^{Ab}	38.13 ^{Aa}	41.19 ^{Aa}	39.74 ^{Aa}

Means within a column with different superscript capital letters are significantly different ($P > 0.05$); means within a row with different superscript small letters are significantly different ($P > 0.05$)

3 Results and discussions

3.1 Changes in titrable acidity of fermented quinoa products

Variations in titrable acidity % of fermented quinoa products during storage at 4 °C ± 1 for 21 days were illustrated in Table 2. By fermentation process using *L. plantarum* and *L. delbrueckii ssp. Bulgaricus*, the titrable acidity increased from 0.27% to values ranged between 0.38 and 0.43%. The treatment of FQD2 showed the highest acidity value (0.43%) ($P > 0.05$) followed by FQD1 (0.42%) may be due to the high count of probiotic bacteria in these treatments, while FQP treatment presented the lowest acidity value (0.38%) after control sample. Lactate produced by lactic acid bacteria is responsible for the acidity increasing in the fermented quinoa products [25, 26]. Furthermore, the acidity values were continuously increased during storage period and the highest values were recorded at the end of storage for all treatments.

3.2 Changes in free phenolic content of fermented quinoa products

Generally, fermentation of quinoa by probiotic bacteria increased the free phenolic content of fermented quinoa products in all treatments compared to non-fermented quinoa products (Table 3). Also, the results showed that the adding of MLP at the two levels (0.25 and 0.50%) significantly enhanced the free phenolic content of fermented quinoa products more than fermented quinoa products without MLP, and the samples containing the higher level of MLP (0.50%) displayed the highest values of free phenolic content (34.24 and 32.85 for FQD2 and FQP2, respectively). Many previous studies indicated to the high antioxidant activity and phenolic compounds of MLP, this may explain the higher phenolic content of fermented quinoa samples supplemented with MLP [13–15].

During the first and second weeks, the free phenolic content of fermented quinoa continuously increased through the storage period may be because of the activity of probiotic bacteria. Călinoiu et al. and Hole et al. [27, 28], reported

that the increase of antioxidants such as phenolic compounds after fermentation may be due to its increased release or synthesis by some possible enzymes, such as β -glucosidases, esterase, cellulase, β -glucosidases and glycoside hydrolase that produced by some probiotic strains. The production of these enzymes during fermentation could release esterified and insoluble-bound phenolics in a time-dependent manner. These results are in line with those obtained from [29, 30], they stated that quinoa fermentation by probiotics improved the free phenolic contents and antioxidant activity.

3.3 Changes in free DPPH radical scavenging activity of fermented quinoa products

Changes in free DPPH of fermented quinoa during storage at $4\text{ }^{\circ}\text{C} \pm 1$ for 21 days were presented in Table 4. Significant differences were recorded in the free antioxidant activity of fermented quinoa supplemented and non-supplemented with MLP. Quinoa products contained MLP at the level of 0.50% and fermented by *L. delbrueckii ssp. Bulgaricus* and *L. plantarum* showed the highest antioxidant activity (7.75 and 6.62%, respectively) followed by fermented quinoa products contained MLP at the level of 0.25% (6.29 and 5.48%, respectively). The increase of free phenolic content results in improving the antioxidant activity of fermented products. The correlation between the phenolic content and the antioxidant activity was reported by many previous studies [27, 31]. Similar results were recorded by [29, 30], they indicated that the antioxidant activity of quinoa was increased after fermentation process by probiotics.

3.4 Changes in phytate content (mg/100 ww) of fermented quinoa products

Phytate is the primary storage form of phosphate in many cereals and it has anti-nutritional effect resulting in malabsorption of some important dietary minerals such as Fe^{2+} and Ca^{2+} as well as decreasing of protein solubility and amino acids bioavailability [32]. From the nutritional point of view, reducing of phytate levels is desirable and necessary to enhance the bioavailability of mineral and amino acids in foods [33]. Several methods have been applied to decrease the phytate concentration in foods such as germination, malting and soaking as well as fermentation, which considers the most common process, used to decrease the levels of phytate in food matrix [34].

Our results indicated that, the fermentation process by probiotics resulted in significant decrease ($P > 0.05$) in phytate concentrations of fermented quinoa products, and fermentation by *L. plantarum* (FQP) presented higher activity for phytate degradation than fermentation by *L. delbrueckii ssp. Bulgaricus* (FQD) (Table 5). Phytate levels were recorded as 67.06 and 71.88 mg/100 g ww for FQP and FQD, respectively compared with control sample, which had the highest phytate content 94.38 mg/100 g ww. Fermented quinoa products supplemented with MLP showed lower phytate concentrations than fermented quinoa products without MLP, and fermented quinoa containing the highest level of MLP showed the lowest phytate concentrations (51.82 and 57.21 mg/100 g ww for FQP2 and FQD2, respectively). All fermented quinoa products exhibited insignificant decreases ($P > 0.05$) in phytate levels through storage period for 21 days at $4\text{ }^{\circ}\text{C} \pm 1$, and FQP2 as well as FQD2 had the lowest phytate contents (44.47 and 47.59, mg/100 g ww respectively) at the end of storage.

Phytate degradation in the fermented quinoa products may be due to the exogenous phytase activity of *Lactobacillus* bacteria rather than endogenous phytase activity of quinoa because of the activity of quinoa endogenous phytase may be reduced by the thermal processes before fermentation [2] as we can observe in the control sample. These

Table 4 Changes in DPPH radical scavenging activity of fermented quinoa during storage at $4\text{ }^{\circ}\text{C} \pm 1$ for 21 days

Treatments	Storage periods (days)			
	0	7	14	21
Control	3.21 ^{Ca}	3.05 ^{Da}	3.26 ^{Ca}	2.84 ^{Ea}
FQP	4.78 ^{BCa}	5.65 ^{Ca}	4.91 ^{BCa}	4.25 ^{DEa}
FQP1	5.48 ^{ABa}	7.00 ^{BCa}	6.18 ^{Ba}	5.68 ^{CDa}
FQP2	6.62 ^{Bb}	8.19 ^{ABa}	8.47 ^{Aa}	7.95 ^{ABab}
FQD	5.31 ^{Ba}	6.14 ^{Ca}	5.74 ^{Ba}	4.63 ^{DEa}
FQD1	6.29 ^{ABb}	7.93 ^{ABab}	8.18 ^{Aa}	6.98 ^{BCab}
FQD2	7.75 ^{Aa}	9.11 ^{Aa}	9.44 ^{Aa}	9.08 ^{Aa}

Means within a column with different superscript capital letters are significantly different ($P > 0.05$); means within a row with different superscript small letters are significantly different ($P > 0.05$)

Table 5 Changes in phytic acid (mg/100) content of fermented quinoa during storage at 4 °C ± 1 for 21 days

Treatments	Storage periods (days)			
	0	7	14	21
Control	94.38 ^{Aa}	94.03 ^{Aa}	93.74 ^{Aa}	93.45 ^{Aa}
FQP	67.06 ^{BcCa}	62.30 ^{BCDa}	61.18 ^{Ba}	60.98 ^{BCDa}
FQP1	59.70 ^{CDEa}	56.00 ^{Ca}	55.85 ^{BcCa}	54.82 ^{Ca}
FQP2	51.82 ^{Ea}	46.21 ^{Ea}	44.58 ^{Da}	44.47 ^{Ea}
FQD	71.88 ^{Ba}	67.14 ^{Ba}	65.13 ^{Ba}	65.01 ^{Ba}
FQD1	63.63 ^{BCDa}	59.27 ^{BCDa}	57.32 ^{BcCa}	56.13 ^{BCDa}
FQD2	57.21 ^{DEa}	52.80 ^{DEa}	49.36 ^{CDa}	47.59 ^{DEa}

Means within a column with different superscript capital letters are significantly different ($P > 0.05$); means within a row with different superscript small letters are significantly different ($P > 0.05$)

results are agree with those obtained from [2], they stated that the fermentation by *Lactobacillus* strains degraded phytate and reduced it in the quinoa flour.

3.5 Changes in water extractable arabinoxylans (WEAX) (mg/100 g ww) of fermented quinoa products

WEAX have antioxidant activity against free radicals in gastrointestinal tract and food matrix and may help in inhibition of colorectal cancers by mitigating the toxicity of free radicals [35]. Also, Malunga and coworkers indicated to the potential antiglycemic effect of WEAX may be due to the direct inhibition of α -glucosidase enzyme activity [36].

The effect of fermentation process on the content of WEAX of fermented quinoa during storage at 4 °C ± 1 for 21 days was shown in Table 6. The content of WEAX increased from 12.18 for control sample to values ranged between 18.42 to 29.34 mg/100 g ww for fermented quinoa products. The results indicated that quinoa products fermented by *L. plantarum* showed higher content of WEAX than quinoa products fermented by *L. delbrueckii ssp. Bulgaricus*. Fermented quinoa by *L. plantarum* recorded 29.34, 24.06 and 21.30 mg/100 g ww for FQP2, FQP1 and FQP, respectively, while fermented quinoa by *L. delbrueckii ssp. Bulgaricus* recorded 24.54, 21.96 and 18.42 mg/100 g ww for FQD2, FQD1 and FQD, respectively. Interestingly, fermented quinoa containing higher content of MLP (0.50%) had the highest contents of WEAX and displayed more positive effect on improving WEAX content than fermented quinoa containing lower content of MLP (0.25%) for both probiotic bacteria may be the enhancing effect of MLP on the growth and survival of probiotics which responsible for the quinoa fermentation and increasing of quinoa WEAX. At the end of storage period, WEAX content reached to the highest values for all fermented quinoa products and FQP2 as well as FQP1 showed the highest contents as shown in (Table 6).

These results were in agreement with [33, 37], they stated that the content of WEAX was increased in fermented cereals after fermentation by lactic acid bacteria. Acidification and specific enzymes, such as xylan-degrading enzymes can hydrolyse the backbone of high molecular weight arabinoxylans and enhance its solubilization [33, 38].

Table 6 Changes in water extractable arabinoxylans (mg/100 g) of fermented quinoa during storage at 4 °C ± 1 for 21 days

Treatments	Storage periods (days)			
	0	7	14	21
Control	12.18 ^{Da}	12.38 ^{Da}	12.09 ^{Ea}	12.46 ^{Da}
FQP	21.30 ^{BcCa}	23.82 ^{BcCa}	23.50 ^{CDa}	24.02 ^{Ca}
FQP1	24.06 ^{Bb}	27.56 ^{ABab}	29.00 ^{ABa}	29.64 ^{Ba}
FQP2	29.34 ^{Ab}	30.72 ^{Aab}	31.74 ^{Aab}	33.36 ^{Aa}
FQD	18.42 ^{Ca}	20.40 ^{Ca}	20.88 ^{Da}	20.76 ^{Ca}
FQD1	21.96 ^{BCb}	24.80 ^{Bab}	26.92 ^{BcCa}	27.60 ^{Ba}
FQD2	24.54 ^{Ba}	27.14 ^{ABa}	28.02 ^{Ba}	28.44 ^{Ba}

Means within a column with different superscript capital letters are significantly different ($P > 0.05$); means within a row with different superscript small letters are significantly different ($P > 0.05$)

Table 7 Effect of fermented quinoa products on blood glucose (fasting and postprandial) levels of experimental rats

Parameters	Rat groups	Experimental periods (days)				↓ or ↑ %
		0	10	20	30	
FG (mg/dl)	Group 1	81.74 ^B	84.78 ^B	96.73 ^A	85.33 ^B	–
	Group 2	88.45 ^A	86.36 ^A	84.63 ^A	84.14 ^A	4.87↓
	Group 3	101.20 ^A	100.11 ^{AB}	94.93 ^{AB}	90.71 ^B	10.37↓
	Group 4	90.51 ^A	88.00 ^A	85.83 ^A	83.09 ^A	8.20↓
PP (mg/dl)	Group 1	138.25 ^A	124.33 ^B	142.20 ^A	136.36 ^A	–
	Group 2	122.70 ^A	123.02 ^A	118.61 ^A	113.16 ^A	7.78↓
	Group 3	119.12 ^A	115.53 ^{AB}	108.89 ^{BC}	102.65 ^C	13.82↓
	Group 4	130.04 ^A	126.16 ^A	122.43 ^{AB}	115.93 ^B	10.85↓

Group (1), control group; Group (2), fed with non-fermented quinoa; Group (3), fed with FQP2; Group (4), fed with FQD2; ↓, % Decrement; ↑, % Increment. Means within a row with different superscript small letters are significantly different ($P > 0.05$)

3.6 Determination of fasting (FG) and postprandial (PP) blood glucose

Type 2 diabetes mellitus (T2DM) is one of the most prevalent chronic diseases around the world. It is characterized by high level of blood glucose, resistance to insulin and relative insulin deficiency, accompanying with multiple systemic complications. According to International Diabetes Federation (IDF), T2DM has affected 415 million adults, and this number can increase to more than 600 million up to 2040 [39]. Data showed in Table 7 illustrated the effect of feeding with fermented quinoa products by probiotics on fasting and postprandial blood glucose levels of experimental rats. The results indicated that the feeding with fermented quinoa products by probiotics significantly decreased ($P > 0.05$) fasting blood glucose levels. Group 3 (Fed with FQP2) showed the highest decrement of fasting blood glucose level (10.37%). Moreover, group 2 (fed with non-fermented quinoa) and 3 (Fed with FQD2) exhibited insignificant decrease ($P > 0.05$) in the fasting blood glucose levels. Also, the results indicated that groups 3 and 4 presented higher decrement ($P > 0.05$) in postprandial glucose levels (13.82 and 10.85%, respectively) than group 2 (7.78%). The higher activity of reducing blood glucose which was recorded in the groups 3 and 4 may be due to the presence of probiotics and prebiotic effect of MLP. These results are in line with [40, 41], they reported that probiotics showed reducing blood glucose activity in bath in vitro and in vivo studies.

4 Conclusion

Symbiotic foods based on quinoa fermented by probiotics were produced as a significant source of bioactive compounds and alive probiotics for the human body. Moringa leaves powder (MLP) was added at the concentrations of 0.25 and 0.50% as additional prebiotic source to quinoa fermented by *Lactobacillus plantarum* ATCC 14917 and *Lactobacillus delbrueckii ssp. Bulgaricus* EMCC 11102. The results indicated that supplementation of fermented quinoa products with MLP at both levels increased its contents of free phenolics and water extractable arabinoxylans as well as enhanced its antioxidant activity and phytate degradation. Fermented quinoa products with 0.50% MLP showed better chemical properties than fermented quinoa products with 0.25% MLP with positive effect on the blood glucose level of experimental rats during feeding period. Supplementation of fermented quinoa products with MLP resulted in improving its nutritional value and healthy benefits. More studies are needed in the future to test more kinds of probiotics with different concentration of MLP in quinoa fermentation.

Author contributions AMA: conceptualization, data curation, formal analysis, investigation, methodology, resources, software, visualization and roles/writing—original draft. EAEL-N: supervision and validation. MNK: supervision, validation and writing—review and editing. All authors read and approved the final manuscript.

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Code availability Not applicable.

Declarations

Competing interests No competing interest exists in this paper.

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