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Comparative growth, biochemical and sensory analyses reveal the feasibility of large-scale development of selenium-enriched Tartary buckwheat sprouts

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Abstract
Selenium is an essential trace element in the human diet and has anti-aging, anti-tumor, and immunity boosting effects. In this study, the effects of selenium on Tartary buckwheat (*Fagopyrum tataricum* L.) sprouts were investigated. A suitable concentration of selenium had no adverse effects on the growth of Tartary buckwheat seedlings. The soluble sugar, protein, total flavonoid, and total phenol concentrations in the selenium-treated sprouts were higher than those in the untreated control sprouts. Electronic tongue measurements indicated that the taste characteristics, especially the sweetness and umami, of the selenium-treated and untreated groups were significantly different. Sensory analysis showed that selenium-treated Tartary buckwheat sprouts had good acceptability. Selenium-enriched Tartary buckwheat sprouts are a promising product, both in terms of flavor and as a useful source of selenium to increase human dietary intake.

Keywords: Tartary buckwheat; selenium; sprouts; electronic tongue.

Practical Application: In this study, the effects of selenium on the nutritional quality, growth and sensory characteristics of Tartary buckwheat sprouts, thus providing a promising product for their good flavor and as a useful source of selenium to increase human dietary intake.

1 Introduction

Buckwheat, which is known as qiaomai in China, is an annual or perennial dicotyledonous plant in the *Polygonaceae* family and *Fagopyrum* genus (Suzuki et al., 2020; Zhang et al., 2021). In the buckwheat genus, Tartary buckwheat (*Fagopyrum tataricum*) and common buckwheat (*Fagopyrum esculentum*) are the major cultivated species and are consumed as food and a raw material for food production (Brasil et al., 2021; Habtemariam, 2019). Buckwheat is becoming a very important alternative crop for use as a food source in areas of Europe and Asia. Compared with cereal crops like wheat and rice, buckwheat contains many bioactive components and is rich in flavonoids (Lv et al., 2017). Tartary buckwheat contains more flavonoids, such as quercetin and rutin, than common buckwheat (Mazahir et al., 2022). Furthermore, Tartary buckwheat has a more balanced composition of amino acids and trace elements, which are nutritionally complementary to staple food crops. In the human body, Tartary buckwheat has various beneficial effects, such as antioxidant, hypoglycemic, and hypolipidemic activities (Nishimura et al., 2016; Wu et al., 2018; Zhang et al., 2017a; Zhu, 2016). These results indicate that Tartary buckwheat has therapeutic potential for the prevention of metabolic syndrome. The biological activities of Tartary buckwheat can be attributed to the presence of flavonoids, which affect the regulation of glucose and lipid metabolism. Because of its therapeutic potential, Tartary buckwheat is recognized as both a medicinal and edible plant. In recent years, with increased demand for nutritional and functional foods, Tartary buckwheat products have become popular among consumers.

Sprouts are young shoots, seedlings, and vegetables that are grown from seed for human consumption. The nutritional qualities and bioactive components in sprouts are affected by the seed germination conditions (Wang et al., 2020). Sprouts are rich in vitamins, minerals, amino acids, and other nutrients needed in the human body and have high nutritional and medicinal value (Mir et al., 2021; Yiming et al., 2015). A previous study showed that buckwheat seed germination significantly changed the contents of proteins and essential amino acids (Zhang et al., 2017b). Tartary buckwheat sprouts germinated from healthy seeds are crisp and delicious with a unique flavor and contain abundant flavonoids and active functional ingredients (Gao et al., 2019; Qian et al., 2020). Consequently, these sprouts are attractive to consumers. Notably, because sprouts have a short production cycle that is unaffected by the climate or propagation site, they have good utilization value and broad market prospects.

Selenium is an essential trace element with various biological functions in the human body, including antioxidant activity and immunity-boosting effects (Vinceti et al., 2017). A deficiency in selenium leads to diseases such as Keshan disease (a cardiomyopathy) and Kashin-Beck disease (an osteoarthropathy). Dietary organic selenium exerts its biological functions mainly through selenoproteins in the human body (Rayman, 2012).

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Selenium cannot be synthesized by the human body and comes from the diet. Plants can take up inorganic selenium from the environment and convert it to organic selenium compounds (Gupta & Gupta, 2017). Inorganic selenium dramatically affects plant nutritional quality, growth and development (Malagoli et al., 2015). In buckwheat, a suitable concentration of selenium greatly improves the grain quality and yield, and soil with an appropriate concentration of Se improved the contents of crude protein, fat, and flavonoids in seeds (Cao et al., 2021; Liu et al., 2022). Previous studies have suggested that buckwheat is highly tolerant to selenium accumulation, which makes it suitable for development of selenium-enriched food products (Golob et al., 2015; Golob et al., 2016). However, further investigation is required to evaluate the effects of selenium enrichment on the taste and nutrient composition of Tartary buckwheat sprouts.

In this study, we investigated the effects of selenium on the nutritional quality, growth, and sensory characteristics of Tartary buckwheat sprouts.

2 Materials and methods

2.1 Plant material

Tartary buckwheat seeds were harvested from the Experimental Station of the Key Laboratory of Coarse Cereal Processing, Ministry of Agriculture and Rural Affairs, at Chengdu in Sichuan, China. After collection, mature healthy seeds were selected and stored in paper bags at 4 °C until required for use.

2.2 Cultivation of Tartary buckwheat sprouts

Seeds in the experimental group were soaked in sodium selenite solution (5 mg/L) for 24 h. The control group was treated with water. Germination and growth assays with the experimental and control groups were performed with plants in a light incubator under identical conditions. The planting substrate was a mixture of equal proportions of nutrient soil and vermiculite. Whole seedlings were harvested after 12 d of continuous cultivation.

2.3 Measurement of growth indexes and selenium concentrations

The seedling height and stem diameter were measured using Vernier calipers. The fresh and dry seedlings were weighed using an electronic balance. The selenium concentrations were determined according to Chinese National Standard GB 5009.268-2016 (National Health and Family Planning Commission of the People's Republic of China, 2016). Sample powder (approximately 0.5 g) was accurately weighed and placed in the digestion tube, and 10 mL of concentrated nitric acid was added. After digestion, the solution was cooled to room temperature and filtered through a 0.45-μm filter membrane. The selenium concentrations were determined by inductively coupled plasma mass spectrometry (iCAP RQ, Thermo Fisher Scientific, Bremen, Germany).

2.4 Analysis of sprout quality

The soluble protein, soluble sugar, and fat contents in the sprouts were estimated using the Coomassie brilliant blue method, anthrone-sulfuric acid method, and Soxhlet extraction method, respectively. Chlorophyll (a and b) and carotenoids were extracted from the sprouts using acetone and analyzed using the colorimetric method. The total flavonoid concentration was evaluated using the aluminum chloride colorimetric method using rutin as a standard. The absorbance was measured using a SynergyHTX multifunctional microplate reader (BioTek, Winooski, VT). The six concentrations of major flavonoids (rutin, quercetin, vitexin, isovitexin, orientin, and isoorientin) in buckwheat were determined by high-performance liquid chromatography (HPLC) according to an established method. HPLC was performed on a LC-20A instrument (Shimadzu, Japan) with an Agilent C18 column (4.6 × 250 mm, 5 μm).

2.5 Electronic tongue analysis

Electronic tongue experiments were carried out with an ASTREE electronic tongue system (Alpha MOS, Toulouse, France) equipped with seven sensors. AHS (sourness), CTS (saltiness), NMS (umami), SCS (bitterness), and ANS (sweetness) sensors were used for the five basic tastes, and PKS and CPS were used as general sensors. Powdered sprout samples (5 g) were added to 150 mL of ultrapure water and then mixed thoroughly. The mixtures were centrifuged at 4000 rpm for 10 min, and the supernatant was collected and analyzed immediately using the electronic tongue system. Data were recorded every second over a total acquisition period of 120 s. An average of 100-120 s of data were taken and analyzed, and principal component analysis (PAC) and discriminant function (DFA) analysis were performed using the accompanying software (AlphaSoft).

2.6 Sensory evaluation

The consumer acceptance was studied using a sensory evaluation conducted with 16 panelists that were selected from a group of graduate students majoring in food sciences (Chengdu University). The nine-point evaluation method, which includes evaluation of the color, flavor, taste, juiciness, and crunchiness, was used to determine the sensory scores.

2.7 Statistical analysis

All results are provided as the mean ± standard deviation. Statistical analysis was performed using Microsoft Excel 2010 and IBM SPSS Statistics 26.0 software. Significant differences (at $P < 0.05$) were analyzed using Duncan's multiple range test.

3 Results and discussion

3.1 Selenium concentration and sprout growth

Seeds in both the experimental and control groups successfully germinated and grew well during the experiments (Figure 1A). To evaluate the effects of selenium on the growth of Tartary buckwheat, the fresh mass, dry mass, seedling height, and stem diameter were measured (Figure 1B). Compared with the control samples, the experimental samples had higher fresh mass (by 1.69%), dry mass (by 8.85%), seedling height (by 5.35%), and stem diameter (by 5.20%). However, these differences were not significant. Therefore, the treatment used in the experiments did not negatively affect seedling growth. Our results are consistent with those from previous reports in that selenium within a certain concentration range is safe and does not have a negative impact on buckwheat growth (Liu et al., 2022; Ma et al., 2020; Song et al., 2019). In addition to the concentration of selenium, plant biomass and growth characteristics are also associated with selenium speciation and the plant species and variety (Jiang et al., 2018; Rao et al., 2021). The concentration of selenium in the treated Tartary buckwheat sprouts in the experimental groups (0.057 mg/kg) increased by 142.36% compared with the control group. The results showed that Tartary buckwheat had a good selenium enrichment capacity, and that the selenium concentration in the Tartary buckwheat sprouts was affected by the selenium concentration in the environment. The sprouts obtained by this method meet the daily recommended intake of selenium from various institutions and international organizations.

3.2 Pigment content and nutrient composition

The color of sprouts is important for determining consumer acceptance. Pigment is an essential component in leaves and largely determines the leaf color. The results for chlorophyll a, chlorophyll b, and carotenoids are shown in Figure 1C. Compared with the control group, the contents of chlorophyll a and chlorophyll b in the experimental group decreased by 0.93% and 0.53%, respectively, and the level of carotenoids increased by 2.7%. There were no significant differences between the two groups in terms of the chlorophyll and carotenoid contents ($p > 0.05$). Therefore, certain concentrations of exogenous selenium do not affect pigment formation or have apparent adverse effects on sprout color. Conversely, selenium could promote the biosynthesis of chlorophyll in broccoli (Ghasemi et al., 2016). The effects of selenium may vary by plant type, and this is probably caused by mutual antagonism between selenium and other elements, which indirectly influences leaf pigment. Interestingly, the soluble sugar and protein contents of sprouts treated with selenium were higher than those in the control sprouts, and these differences were statistically significant (Figure 2). The crude fat content exhibited no significant differences. Previous studies have found that exogenous selenium influences the protein content or distribution in plants (Kovács et al., 2021). Although selenium is not considered an essential trace element for plants, numerous authors have confirmed its beneficial effect on plant life (Gupta & Gupta, 2017; Malagoli et al., 2015). This conclusion inspired us to improve the protein quality in sprouts.

3.3 Concentrations of bioactive compounds

Active ingredients, especially flavonoids, are a distinguishing feature of buckwheat compared with other stable grains. Our results showed that the concentrations of total polyphenols in Tartary buckwheat sprouts were higher than those of total

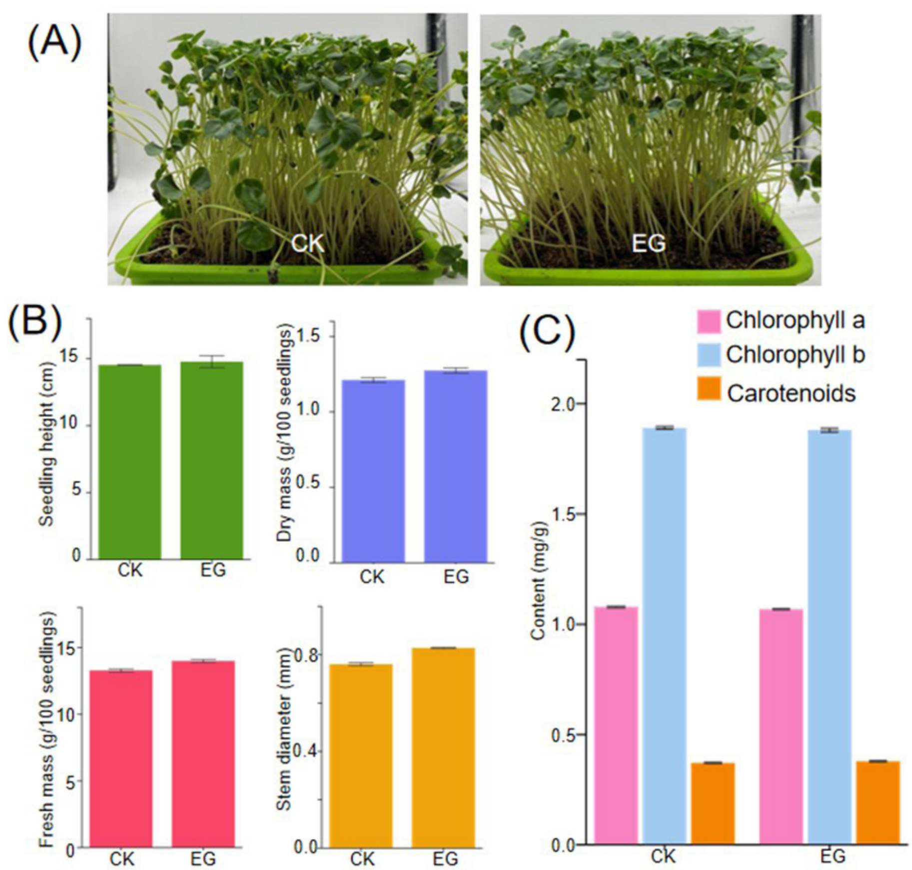

Figure 1. The morphological (A), growth characteristics (B) and pigment content (C) of the control groups and experimental groups. Ck represents the control group and EG represents the treatment group. Vertical bars indicate the means ± standard error (n = 3).

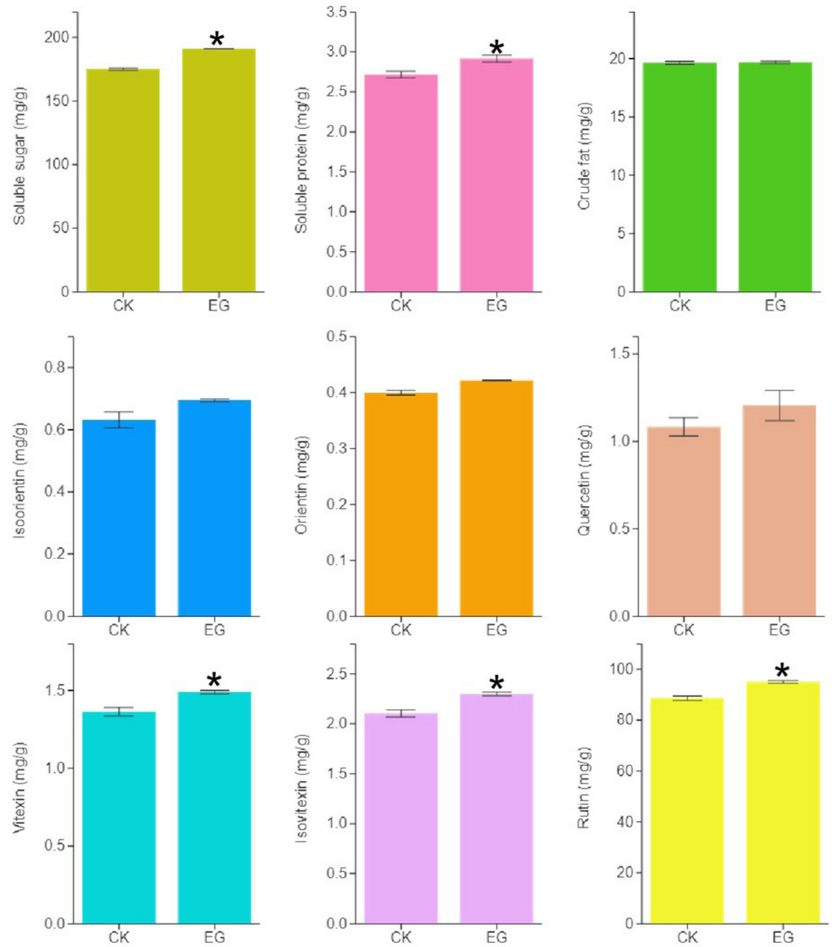

Figure 2. Content of nutrient composition and flavonoids of the control groups (CK) and experimental groups (EG). Error bars indicate standard error means of means. Asterisk indicates the significant difference ($P < 0.05$).

flavonoids in all groups. The total polyphenol and flavonoid concentrations increased by 7.18% and 17.45%, respectively, in the sprouts treated with selenium, and these differences were significant compared with the control. HPLC analysis was used to determine the concentrations of rutin, quercetin, vitexin, isovitexin, orientin, and isoorientin (Figure 2). Among the six flavonoids, rutin was the most abundant in Tartary buckwheat sprouts, followed by isovitexin, and then quercetin. Although all six flavonoids were detected in all samples, their concentrations were altered in the sprouts treated with selenium. The concentrations of the main flavonoid components were higher in the experimental samples than in the control samples. The rutin concentration reached 94.97 mg/g and was 7.16% higher than that in the control. Our results agree with those from other studies on tomatoes and groundnuts, and demonstrate that a suitable selenium concentration promotes flavonoid biosynthesis (Hernández-Hernández et al., 2019; Hussein et al., 2019). Exogenous selenium may be a multifunctional regulatory factor that promotes flavonoid formation to a certain extent. In addition to the selenium concentration, the high flavonoid concentration in Tartary buckwheat sprouts is a very attractive characteristic for consumers.

3.4 Sensory evaluation and electronic tongue

Sensory evaluations of food quality (e.g., appearance, flavor, taste, and texture) are commonly used to assess consumer acceptance of novel products (Cheng et al., 2020; Savini et al., 2020). This method is appropriate for sprouts. The selenium-treated and untreated sprouts were evaluated for color, flavor, taste, juiciness, and crunchiness. The selenium-enriched Tartary buckwheat sprouts were bright green and shiny with a slightly sweet taste, plenty of juice, crisp texture, and high quality. The scores of the five parameters showed no significant differences between the experimental and control groups (Figure 3A). Therefore, selenium did not affect the sensory qualities and all products were well accepted by the subjects. However, this sensory evaluation is subjective. As an alternative, an electronic tongue is an intelligent instrument that can simulate human gustatory evaluation to detect taste (Wu et al., 2022; Zaukuu et al., 2021). Therefore, we used an electronic tongue to evaluate the taste profiles of the samples. Data from the seven sensors were plotted in a radar chart (Figure 3A). The sweetness sensor showed the highest response in all Tartary buckwheat sprouts, followed by bitterness, and then sourness. Compared with the control group,

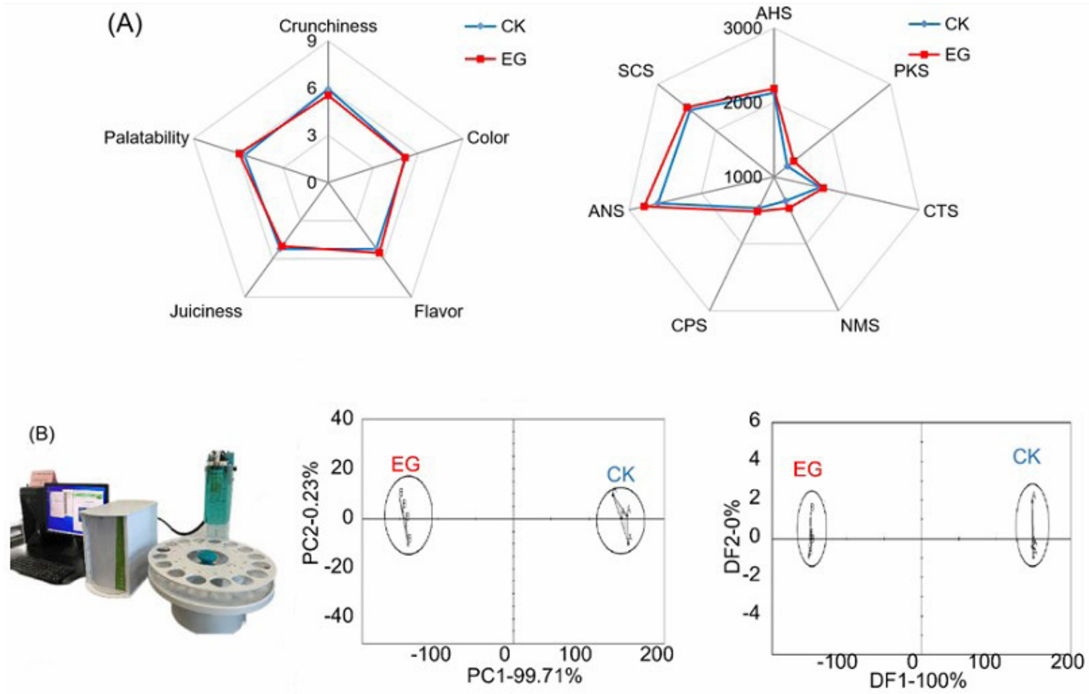

Figure 3. Averaged radar charts of sensory evaluation (left figure) and electronic tongue (right figure) for the control groups and experimental groups (A). Alpha MOS electronic tongue, principal component analysis and discriminant factor analysis (B).

the selenium-treated group showed higher values for all sensors and these differences were statistically significant. PCA and DFA were able to discriminate between the two groups, which further supported the statistical significance of the difference between the control and experimental groups (Figure 3B). However, the electronic tongue results were inconsistent with the results of the sensory evaluation. The high sensitivity and distinguishing ability of the electronic tongue could overcome the issues with subjective human sensory evaluation. In the future, we will investigate the mechanism by which selenium influences food taste. As a raw material for food, buckwheat has been processed into various products, such as breads, noodles, tea, beer and vinegar (Brasil et al., 2021). Quercetin can be generated in Tartary buckwheat products as a result of rutin degradation in food processing, which may contribute to the bitter taste (Sun et al., 2022). The bitter flavor of Tartary buckwheat largely influences consumer acceptability. Interestingly, Tartary buckwheat sprouts are an excellent dietary source of phenolic compounds, which can improve the taste and eventually increasing its market value. Our team has developed a series of Tartary buckwheat sprouts products, and some related products have become available.

4 Conclusion

The addition of exogenous selenium to Tartary buckwheat increased the selenium concentration in the sprouts and did not adversely affect growth. The sprouts obtained by this approach met the requirements for daily human selenium intake. Although the total polyphenol and flavonoid concentrations increased in the sprouts treated with selenium, sensory evaluation scores showed no significant differences. An electronic tongue with high sensitivity and distinguishing ability enabled an improved evaluation of the selenium-enriched sprouts. Selenium-enriched Tartary buckwheat sprouts are a promising product for their good flavor and as a useful source of selenium to increase human dietary intake.

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