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Research Article

Iodine biofortification impacts the nutraceutical compounds content and agronomic characteristics of lettuce (*Lactuca sativa* L.)

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Abstract

Currently, agronomic biofortification through foliar fertilization is an effective process for correcting micronutrient deficiencies in the population in a sustained manner. Some studies have shown that iodine has a positive effect on crop metabolism and development, as well as on the absorption of this element and its buildup in the edible organs of the vegetable, mitigating the problem of iodine deficiency in the population. In this work, the response of potassium iodide (KI) on lettuce plants (*Lactuca sativa* L.) produced in a hydroponic system (NFT) was evaluated using a completely randomized design. Five treatments, one control, and five replicates per treatment (control, 5, 10, 15, 20, 25 μmol L^{-1}). The morphological variables, chlorophyll content, phytochemicals, and mineral concentration were quantified. The results show that the dose of 20 μmol L^{-1} of (KI) increases compared to the control treatment in the morphological variables, including plant height (41.83%), leaf length (41.30%), leaf width (45.45%), crown diameter (29.79%), number of leaves, root fresh weight (99.58%), and dry matter (77.7%). At concentrations of 20 μmolL^{-1} of (KI), chlorophyll showed better results (43.5%) than the control. At doses of 15 and 20 μmolL^{-1} of (KI), phytochemical variables showed increases in phenols (56.5%), flavonoids (31.8%), antioxidant capacity (5.5%), and vitamin C (3.33%) compared to the control. Iodine concentration increased with higher doses, showing the maximum concentration at 25 μmol L^{-1} of (KI) (48.04%) compared to the control. Doses of 20 μmolL^{-1} of (KI) increased the accumulation of certain minerals (N, P, Ca^{+2}, Mg^{+2}, Cu, Mn, Na) (7.7 to 35.7%) compared to the control. In contrast, higher doses of 15 μmolL^{-1} of (KI) caused antagonism, decreasing the concentration of Fe and Zn. The application of adequate doses of iodine is a good alternative for biofortification.

Keywords: deficit; foliar fertilization; functional feed; metabolism; micronutrients; phytochemicals

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Introduction

Micronutrient deficiency in Latin America constitutes a critical public health issue, affecting millions and contributing to various disorders and diseases (FAO, 2020). Among essential micronutrients, iodine is important in regulating metabolism and cognitive development, and its deficiency can lead to serious consequences such as goiter and thyroid dysfunction (Sularz *et al*., 2020; Lopez, 2022). For this reason, the World Health Organization (Allen *et al*., 2017) suggests a daily intake of 150 µg of iodine for adults and 90 to 120 µg for children. Although universal salt iodization has been the predominant strategy to combat iodine deficiency, its effectiveness has been limited by the instability of the compound, highlighting the need to explore more efficient and sustainable alternatives (Leija-Martínez *et al*., 2016).

In this context, agronomic biofortification emerges as a promising solution capable of naturally and safely increasing the iodine content in foods (Sida *et al*., 2015). This approach not only allows offers for improving micronutrient intake through the diet but can also offer additional benefits on plant growth, production, and stress resistance and in terms of nutraceutical quality by promoting the accumulation of bioactive compounds in vegetables (Kiferle *et al*., 2020). Iodine is related at the level of the plant transcriptome, as demonstrated by Kiferle *et al.* (2021) through bioinformatics analysis in which they found a regulation of the expression of genes that encoded in enzymes of the antioxidant and defense system, in addition to protein-iodine association present in chloroplasts and photosynthesis, consequently positively affecting biomass accumulation and quality parameters.

In recent years, the benefits of biofortification of crops have been widely documented (Ofori *et al.,* 2022). In the case of iodine biofortification by different methods, it has successfully increased the iodine content in the target organs of several crops (Duborská *et al.,* 2020); however, the method can influence the fact that root application is highly justified to correct deficiencies or increase nutrients. This can present essential challenges since some elements are of low mobility, such as iodine, difficult transport via apoplast or symplast to the aerial part, and drastically decreasing the assimilated concentration (Riyazuddin *et al.,* 2023).

Even though the iodine content increased in target organs in various crops, completing the requirements that a person needs daily, such as carrot, potato, tomato, chili, and strawberry. However, these experiments have in common that the biofortification was carried out through the root system, requiring higher concentrations with the risk of radical toxicity, volatilization (Zhang *et al.,* 2023), and increased fertilizers wasted and costs. Therefore, there is a need to improve the application of iodine to plants in small doses.

The above was validated through the iodine foliar biofortification in tomato, a fruit vegetable, in the form of KI at concentrations of 1 to 9 µMol, resulting in increased fruit I content and stimulated growth and development (Ikram *et al.,* 2024); while a significant increase in the accumulation of I in leaves and roots of carrots was observed when iodine was applied foliar at concentrations of 10 to 20 mMol L^{-1}. However, there was no effect on either the yield or the quality of the commercial product (Rakoczy-Lelek *et al.,* 2021); similar results were found in potatoes in which the presence of iodine in the tubers increased with low doses applied because the translocation of iodine via phloem from the aerial to the underground part is easier than the xylem, although yield and quality parameters remained unaffected (Ledwożyw-Smoleń *et al.,* 2017).

But without a doubt, the most significant benefit of iodine foliar biofortification is in leafy crops due to the direct topical effect on the leaves. The antioxidant mechanism is activated (Dobosy *et al.,* 2024), as well as improving the agronomic characteristics of the crops as the concentration of iodine in the leaves obtaining higher commercial and phytochemical quality harvest, as demonstrated Krzepiłko *et al.* (2019) in spinach, and Dobosoy *et al.* (2024) in kohl. However, there is also evidence of presenting phytotoxicity at doses up to 20 mMol under a hydroponic system in tomatoes, negatively affecting vegetative and reproductive growth, while at low doses, the leaves remained greener (Landini *et al*., 2011)

Lettuce (*Lactuca sativa* L.), a vegetable widely consumed globally and produced in large quantities in Mexico, stands out as a model for iodine biofortification because of its high capacity for absorption and accumulation of the mineral (Smoleń *et al*., 2014)

Recent research has shown promising results in the biofortification of lettuce with potassium iodide (KI), demonstrating significant increases in iodine concentration in leaves, which could lead to an improvement in the nutraceutical quality of the vegetable (Figueiredo *et al*., 2022).

In another experiment, Lawson *et al*. (2015) found a positive effect of iodine-applied foliar in butterhead lettuce on commercial parameters and concentration of phytochemicals of the antioxidant system. Also, Blasco *et al*. (2008), at low doses in lettuces cultivated in vermiculite substrate, observed an increase in carbohydrates and enzymatic activity of the antioxidant system as well as the accumulation of biomass.

This study evaluated the effect of the foliar application of different KI low concentrations on the yield, commercial quality, and nutraceutical quality of lettuce grown in a hydroponic system and the concentration of iodine accumulated in the leaves.

Materials and Methods

Study area

The experiment was conducted at the Instituto Tecnológico de Torreón (ITT) facilities located in the city of Torreon Coahuila, México at coordinates 25° 36′ 37″ N -103° 22′ 33″ W, at 1120 m. a. s. l., during the autumn-winter cycle of 2020-2021, from November to February, with a maximum temperature of 30 °C, minimum temperature of 5 °C, average temperature of 17 °C, and average relative humidity of 33.7%. The shade netting structure is provided with anti-aphid mesh (white color) with 25 x 25 UV-treated polyethylene threads, 720 gauge, 30% shade, and diffuse light, with external radiation assimilation of 9189 μmol s^{-1} m^{-2}.

Plant material and treatments evaluated

Seeds of butterhead lettuce (*Lactuca sativa* L.) Vita® from Rancho los Molinos® (Morelos, Méxcio) were used. They were sown and germinated in phenolic foam plates. After 15 days, the seedlings were transplanted when they presented 3 to 4 true leaves to a hydroponic system NFT Nutrient Film Technique (Steiner 1961) elaborated with hydraulic PVC tubes of four inches in diameter and six meters long, with a slope of 10% and with a space between cavities of 25 cm. The system maintained an electrical conductivity (EC) of 2.0 dS m^{-1} and a pH between 5.5-6. Highly soluble commercial fertilizers available in the region (MKP, $MgNO_3$, K_2 SO_4, NKS, $CaNO_3$ -) and micronutrients of the Nutrisol® brand (New Delhi, India) were used for its preparation.

The potassium iodide (KI) (Jalamek ®) at 71% purity was used as a source of iodine. Five increasing doses and one control were evaluated for each treatment with five replicates (Table 1).

Table 1. Description of KI treatments applied to lettuce cultivation in a hydroponic system

Treatment	**KI concentration (μmol L^{-1})**
T1	Control (tridistilled water)
T2	5
T3	10
T4	15
T5	20
T6	25

A completely randomized experimental design with six treatments and five replications was used, resulting in 30 experimental units.

Before the application of the treatments, the potassium iodide was diluted in tridistilled water with 1-2 mL of surfactant adherent (INEX-A) per liter of water. Industrial atomizers were used to spray the KI solution in the mornings (8:00-9:00 am) homogeneously to the entire plant with an interval of 15 days, accumulating five applications during the experiment. Harvesting was carried out 70 days after transplanting when the leaves of the lettuce crop had reached commercial maturity.

Morphological parameters

Once the crop cycle had elapsed, when the lettuce plants reached commercial maturity, they were removed in the morning from the hydroponic system to begin data collection, such as measuring the fresh weight of leaves, stems, and roots using precision scale with an accuracy of ± 0.01 g (Truper base-5EP modelo 3V cc). Other parameters measured were leaf height, which was measured from the base of the stem to the apex of the leaf using a flexometer, as well as head diameter and number of leaves.

For the determination of the dry matter, at the time of harvest and after weighing the fresh plant material (leaves, stems), the leaves were washed with deionized water and placed in the induction oven (memmert Single) at 70 °C for 48 h until a constant weight was obtained to obtain dry matter, which was expressed in g.

Lyophilization of samples

The selected samples were frozen for 24 h and, subsequently, placed in freeze-drying equipment (Labconco Freezone Triad* Freeze Dry Systems$^{®}$) (ultra-cooling for dehydration of the vegetable sample using the sublimation principle) at a temperature of -20 °C for 5 days, followed by grinding.

Chlorophyll estimation

Chlorophyll content was determined using Lichtenthaler and Wellburn's method (1983). 10 mg of lyophilized tissue was suspended in 95% ethanol and homogenized. The mixture was centrifuged at 1500 g for 20 minutes at 4 °C (Sorwall ST 8R Thermo Scientific) centrifuge. With the supernatant obtained, the absorbance was read at different wavelengths (665, 649, and 470 nm) using a UV-visible spectrophotometer (Jenway 7305). The results were reported using specific equations. (1-3):

$$\text{Chlorophyll (a)} = 13.95\ A665 - 6.88\ A649 \tag{1}$$

$$\text{Chlorophyll (b)} = 24.96\ A649 - 7.32\ A665 \tag{2}$$

$$\text{Total chlorophyll} = \text{Chl(a)} + \text{Chl(b)} = (1000\ A470 - 2.05\ \text{Chl(a)} - 114.8\ \text{Chl(b)}) / 245 \tag{3}$$

Extraction of extracts

100 mg of sample was used in 10 mL of 80% ethanol in screw-capped plastic tubes, placed in a rotary shaker (ATR Inc., USA) for 24 h at 2,000 rpm at 5 °C. The homogenate was centrifuged at 1500 g for 5 min, and the supernatant was extracted for further analysis.

Phytochemical compounds

The method described by Singleton *et al.* (1999) was used for total phenols determination. 300 µL of the extract was mixed with 1080 µL of water in a test tube and 120 µL of Folin-Ciocalteu's reagent. The mix was vortexed for 10 seconds. After 10 minutes, 0.9 mL of 7.5% (w/v) Na_2CO_3 was poured and gently shaken for 10 seconds. At room temperature, the samples were reacted for 30 minutes, and then absorbance was measured at 765 nm using a UV-Vis spectrophotometer (Jenway 7305). The standard was prepared with gallic acid (GA), and the results were expressed as mg equivalent mg GA 100 g^{-1} dry weight.

The total flavonoids were determined following the method of Hidalgo et al. (2019). 250 µl of ethanol extract, 1.25 ml of water, and 75 µl of 5% $NaNO_2$ were poured into a test tube; the mixture was vortexed and rested for 5 min at room temperature. Subsequently, the mixture was reacted with 150 µl of $AlCl_3$. After the reaction, 500 µl of 1 M NaOH and 275 µl of water were added. The mixture was shaken vigorously after reading the absorbance at 510 nm UV-Vis spectrophotometer (Jenway 7305). The standard was prepared with quercetin dissolved in absolute ethanol, and the results were expressed as mg QE 100 g^{-1} dry weight.

The antioxidant capacity was determined using the method 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical method (Hsu *et al.,* 2003) with brief modifications. A solution of DPPH in ethanol was prepared at a concentration of 0.025 mg mL^{-1}. 50 µL of ethanolic extract was mixed with 1950 µL of DPPH solution, and after 30 minutes, the absorbance of the samples was read on a UV-Vis spectrophotometer (Jenway 7305) at 517 nm. The results were expressed as µM equivalent in Trolox 100 g^{-1} dry weight.

Vitamin C

Vitamin C content was determined by the method reported by Padayatt *et al.* (2001), suspending 100 mg of lyophilized tissue with 10 mL of 2% (v/v) hydrochloric acid. It was filtered and made up to 100 mL with distilled water in an Erlenmeyer flask. With 10 mL of the aliquot, it was titrated with 2,6 dichlorophenolindophenol (1×10^{-3} N), and the vitamin C content was determined by equation (4):

$$\text{Vitamina C} = \frac{(2.6\text{ mL of diclorofenolindofenol})(0.088)(\text{ total volume})(100)}{(\text{volume alicuota})(\text{sample weight})} \tag{4}$$

Determination of iodine in leaves

The alkaline digestion technique was used to determine the iodine content (Medrano-Macias *et al.,* 2021). 1 g of dry lettuce leaf tissue, 2 mL of 2 M KOH, and 1 mL of 2 M KNO_3 were added. The mixture was placed in an oven at 100 C for 2 h. Subsequently, the sample was digested in a muffle at 580 °C for 3 h. Finally, the iodine was extracted with 2 mL of 2 mM KOH. Iodine determination was used by the catalytic method of plasma mass spectrometry (ICP-MS) (Verma *et al.,* 1992; Krishna *et al.,* 1992). The concentration of iodine in the samples was evaluated using a calibration curve obtained with the standard, and the results were reported in mg kg^{-1} dry weight (dw).

Determination of leaf minerals

The nitrogen (N) content was determined through the Kjeldahl method (Plank, 1992), in which, in principle, organic N is converted into ammonium (NH_4^+) by digestion of the sample with concentrated sulfuric acid (H_2SO_4), and the result of digestion is measured by the amount of NH_4^+produced expressed as a percentage.

Phosphorus (P) concentration was determined in 0.5 grams of dry lettuce leaf sample through the colorimetric method based on the intensity of the yellow color measured at 430 nm in a spectrophotometer (Jenway 7305 UV-Vis) as a product of the reaction of molybdenum and ammonium vanadate (Wang *et al.,* 2023) in an acidic medium and reported as a percentage of Phosphorous from a curve with KH_2PO_4 as standard at different concentrations.

The dried leaf sample was weighed (1 g) into digestion tubes, and 10 ml of nitric acid 65% was added to determine the total contents of K^{+1}, Ca^{+2}, Mg^{+2}, Cu^{+2}, Fe^{+2}, Zn^{+2}, and Mn^{+2} according to Kawashima and Valente-Soares (2003). The tubes were heated in an infrared digestion apparatus. The solution was allowed to dry when the contents of the tubes were clear. The residue was dissolved with sufficient 1% nitric acid and 0.5% lanthanum solution until reaching a capacity of 10 ml in a volumetric flask. The potassium, calcium, magnesium, copper, iron, zinc, and manganese solutions were read in a Flame Atomic Absorption Spectrometry (F-AAS Thermo Scientific iCE). Calibration curves with standards were used to determine concentration. Results were expressed as mg element kg dry $weight^{-1}$.

Statistical analysis

The results presented here for all variables evaluated were analyzed by analysis of variance and mean comparison test using the Tukey method ($P \leq 0.05$) using the statistical package SAS (Statistical Analysis System Institute) version 9.4. The general description of the polynomic regression model to investigate the relationships between the KI concentration applied and the response variables: concentration of phytochemicals (total phenols and flavonoids), vitamin C in lettuce leaves was as follows: $Y = \beta 0 + \beta 1x^2 + \beta 1x + \epsilon$, where Y is a response variable, β0 is constant or intercept, β1 is the slope, x is the independent variable and Є is the error term.

Results

Lettuce plant morphology

Foliar iodine spraying caused changes in morphological traits in the lettuce crop, according to the results obtained in the analysis of variance, showing significant differences ($P \leq 0.05$), observing that the concentration of 20 μmol L^{-1} of KI increased the characteristics such as plant height (32.50 cm), leaf length (30.50 cm), leaf width (24.00 cm), crown diameter (46.00 cm), leaf number (75.50) and root fresh weight (359.25 g) as represented in Table 2.

Table 2. Effect of foliar biofortification with KI on lettuce crop morphological characteristics

(KI) (μmol L^{-1})	Plant height (cm)	Leaf length (cm)	Blade width (cm)	Crown diameter (cm)	Sheet number	Root fresh weight (g)
0	23.00 ± 0.00 d*	21.50 ± 1.00 d*	16.50 ± 0.50d *	35.44 ± 1.94 d*	59.00 ± 1.00 b	180.00 ± 7.00 d*
5	26.10 ± 0.10 c	25.25 ± 0.25b,c	17.37± 0.62 c,d	40.50 ± 1.00 b,c*	64.19 ± 1.44 b	281.50 ± 7.50 c*
10	27.00 ± 0.50 c	26.50 ± 1.00b,c	18.75 ± 0.75 b,c	41.25 ± 1.25 b,c	64.33 ± 1.52 b	299.62 ± 5.62 c
15	28.75 ± 0.50 b	27.00 ± 0.50 b	20.15 ± 0.15 b	43.25 ± 1.25 a,b	71.25 ± 0.25 a	321.00 ± 11.00 b
20	32.50 ± 1.00 a	30.50 ± 0.50 a	24.00 ± 1.00 a	46.00 ± 0.50 a	75.50 ± 3.50 a	359.25 ± 8.75 a
25	26.00 ± 0.00 c	25.12 ± 0.12 c	17.12 ± 0.62 c,d	38.75 ± 1.25 c,d	60.25 ± 2.25 b	279.12 ± 4.12 c
CV	1.842	2.523174	3.478405	3.111597	2.963454	2.669092
R^2	0.980576	0.961628	0.956957	0.911051	0.931199	0.987204
DSM	1.3758	1.7977	1.811	3.4873	5.3441	20.99

Comparison of means, plant height, leaf length and leaf width, crown diameter, leaf number and root fresh weight in lettuce plants with foliar applications of potassium iodide. * Values with different letters within each column are statistically different according to Tukey's test ($P \leq 0.05$).

The results of the analysis of variance in fresh weight and leaf dry weight showed a statistically significant difference ($P \leq 0.05$) due to the effect of foliar application of KI on the lettuce crop. It was observed that the best treatment concerning the control was the application of 20 μmol L^{-1} KI, with a value of 1178.00 g, exceeding the control by 77.7%. There was a decrease in weight at the concentration of 25 μmol L^{-1} KI (Figure 1).

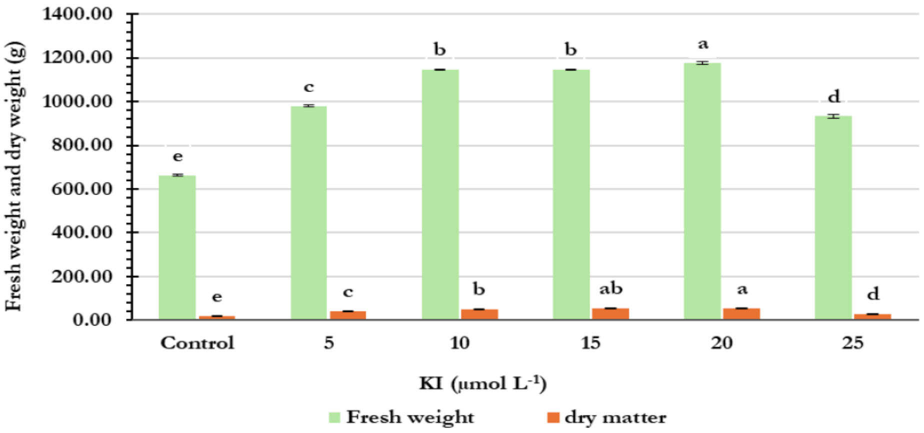

Figure 1. Effect of foliar application of different concentrations of (KI) on fresh weight and dry weight in lettuce plants produced in an NFT system

Data are shown as means (n= 30) ± standard deviation. According to Tukey's test ($P \leq 0.05$), bars with different letters are statistically different

Phytochemical compounds

Chlorophyll content

Figure 2 shows the chlorophyll content in lettuce leaves, indicating a significant difference ($P \leq 0.05$). It shows an increase as the KI dose increases, obtaining the best treatment of 20 µmol L^{-1} with an increase of 43.5% more than the treatment control; on the other hand, the treatment of 25 µmol L^{-1} decreased in 4.9% of chlorophyll.

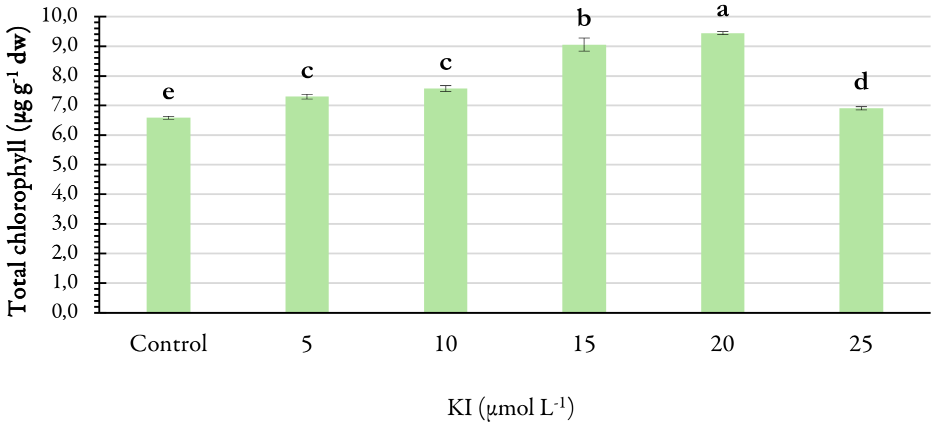

Figure 2. Effect of foliar application of KI on total chlorophyll content in lettuce leaves produced in an NFT system

According to Tukey's test ($P \leq 0.05$), bars with different letters are statistically different

Total phenols content

Figure 3 shows the values for phenols, obtaining a significant statistical difference ($P \leq 0.05$). The highest value of this bioactive compound (162.05 and 153.35 mg GA 100 g^{-1} dw) was recorded with the concentration of 15 and 20 µmol L^{-1}. This value is 56.5 and 48.1% higher than recorded in the control treatment, which was 103.55 mg GA 100 g^{-1} dw.

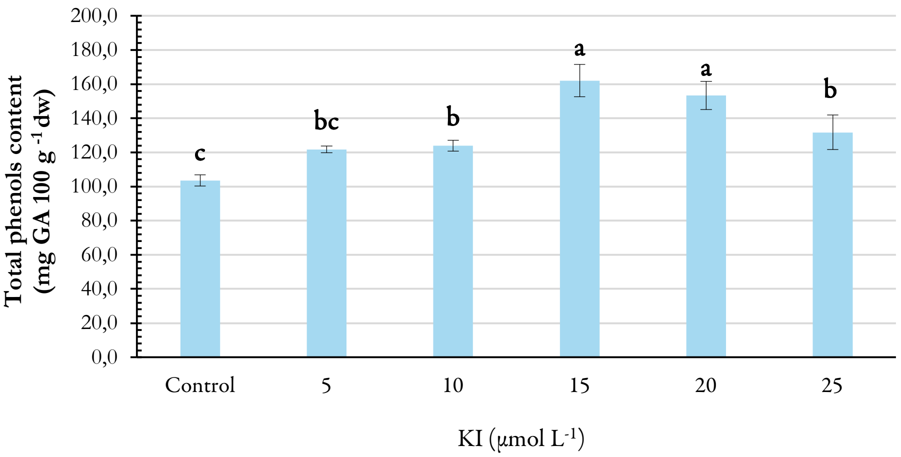

Figure 3 Effect of foliar application of KI on phenol content in lettuce plants grown in an NFT system
Data are shown as means (n= 30) ± standard deviation. Bars with different letters are statistically different according to the Tukey test ($P \leq 0.05$)

Total flavonoids content

The results of the variance analysis for flavonoids showed a significant difference. The highest value of this bioactive compound was recorded with a concentration of 15 and 20 µmol L^{-1}, with a value of 196.293 and 197.143 mg QE 100 g^{-1} (dw). In the flavonoids variable, this value exceeds 31.3 and 31.8%, the value recorded in the control treatment 149.537 mg QE 100 g^{-1} (dw) (Figure 4).

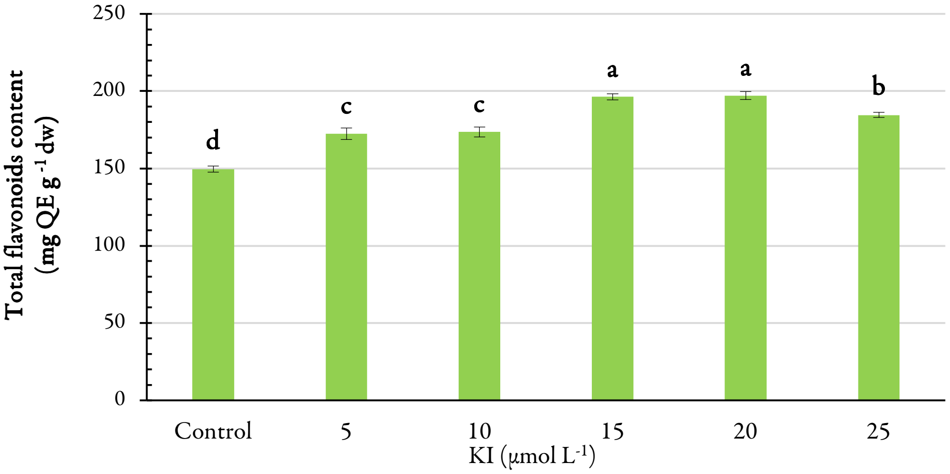

Figure 4. Effect of foliar application of KI on the content of Flavonoids in lettuce plants produced in a NFT system
Data are shown as means (n= 30) ± standard deviation. Bars with different letters are statistically different according to the Tukey test ($P \leq 0.05$)

Antioxidant capacity

For the antioxidant capacity, the analysis of variance showed a significant statistical difference ($P \leq 0.05$) due to the effect of the KI foliar spray on the lettuce leaves. The highest values were 176.36 and 176.13 mg equiv. Trolox 100 g^{-1} (dw) was recorded with the concentration of 15 and 20 µmol L^{-1}, this value was 5.6 and 5.5% higher than the value recorded in the control treatment 166.97 mg equiv. Trolox 100 g^{-1} (dw) (Figure 5).

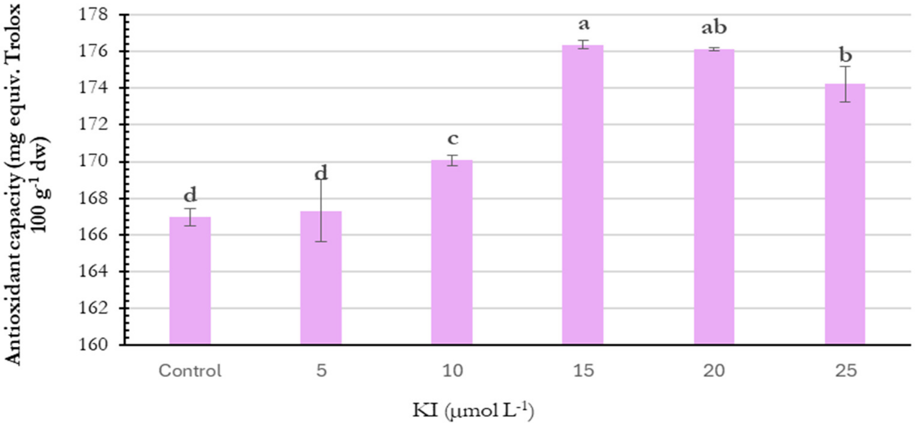

Figure 5. Effect of foliar application of KI on the antioxidant capacity in lettuce plants grown in an NFT system)

Data are shown as means (n= 30) ± standard deviation. Bars with different letters are statistically different according to the Tukey test (P ≤ 0.05)

Vitamin C

Vitamin C obtained a statistically significant difference according to the analysis of variance where the highest value was recorded with 0.21 g 100 g^{-1} dw, corresponding to the concentration of 20 µmol L^{-1}, this value is 3.33% higher than the value recorded in the control treatment of 0.09 g 100 g^{-1} dw (Figure 6).

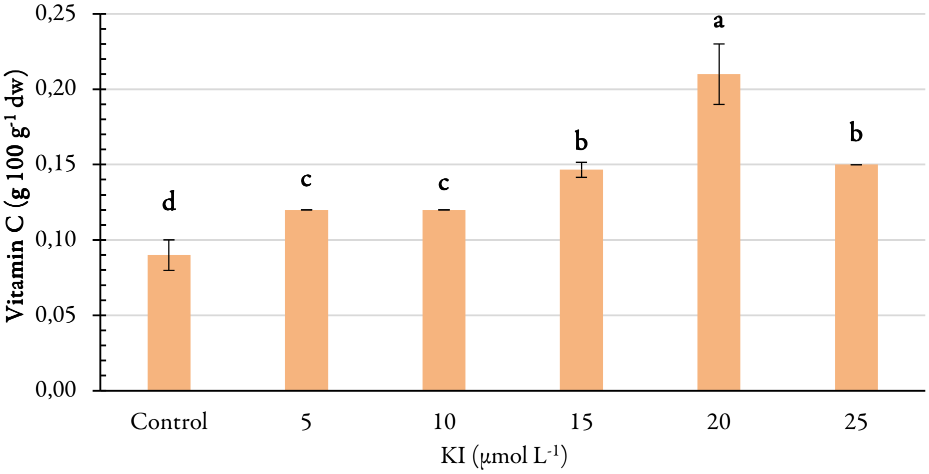

Figure 6. Effect of foliar application of KI on vitamin C content in lettuce leaves, produced in a NFT system

Bars with different letters are statistically different according to the Tukey test (P ≤ 0.05)

Leaf iodine concentration

The results obtained for the concentration of iodine in the leaves of the lettuce plant increased as the doses sprayed on the plant increased, observing the highest content 4.0372 mg kg^{-1} dw, with 25 µmol L^{-1} KI, surpassing the rest of the treatments by 48.04% to the control treatment 0.6226 mg kg^{-1} dw. (Figure 7).

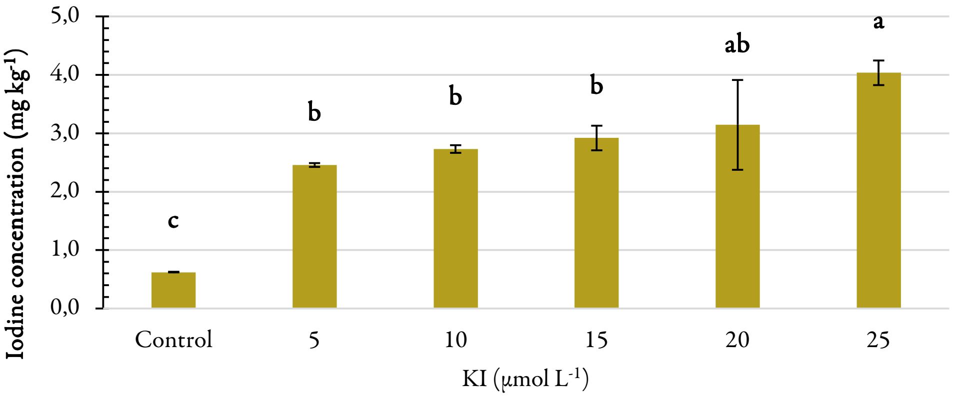

Figure 7. Effect of foliar application of different Kl concentrations on total iodine content in lettuce plants produced in a NFT system

Data are shown as means (n= 30) ± standard deviation. Bars with different letters are statistically different according to the Tukey test ($P \leq 0.05$)

Table 3 shows the results of the regression analysis in 5 variables that were susceptible to fitting the polynomial type model, that each of the variables analyzed presented similar patterns, in which higher concentration of KI increased the fresh weight of the lettuce heads ($R^2 = 0.9672$), as well as the concentration of total phenols (R2 = 0.7934)and flavonoids (R2 = 0.9453), and vitamin C (R2 = 0.6880), up to the point that for more KI applied there is a reduction in the performance of the variables analyzed; except the iodine concentration in the lettuce leaves, which in each of the concentrations resulted in a more significant amount of accumulated iodine, however, even in this variable, the polynomial regression fit better (R2 = 0.8839) than the linear one (Figure 7).

Table 3. Polynomial model regression analysis in 5 response variables to different concentrations of KI as a biofortified

Variable	Equation of the polynomial model regression	R2
Fresh weight (g)	Y = -53.214x2 + 439.5x + 273	0.9672
Total phenols (mg GA 100 g-1 dw)	Y = -5x2 + 41.714x + 64	0.7934
Total flavonoids (mg QE 100 g-1 dry weight)	Y = -3.375x2 + 29.71x +124.7	0.9453
Vitamin C (g 100 g-1 dw)	Y = 0.0041x2 + 0.0455x + 0.044	0.6880
Iodo concentration (mg kg-1 dry weight)	Y = -0.0893x2 + 1.1864x - 0.14	0.8839

Mineral content

The concentration of minerals was quantified (Table 4), observing a statistically significant difference ($P \leq 0.05$) in elements such as N, which showed an increase of this element as the KI dose increased, obtaining as the best treatment the concentration of 20 µmol L^{-1} by 35.01% concerning the control treatment. The same effect was seen for the element P, where the increase in concentration was 34.5% with the dose of 20 µmol L^{-1} compared to the control, but a notable decrease of 10.9% in P was observed when applying doses of 25 µmol L^{-1}.

The results obtained for the element Ca showed a notable increase of 9.7 and 12.1% of this element in the doses of 15 and 20 µmol L^{-1} of KI compared with the control treatment. For the mineral Mg, statistically significant differences were observed in the doses of 15 and 20 µmol L^{-1} of KI with 5.1 and 7.7%; however, KI showed no significant difference for the treatments 0, 5, 10, and 25 µmol L^{-1}.

For microelements such as Cu, a significant difference was observed in the doses of 25 μmol L^{-1} with an increase of 44.7% compared to the control; on the other hand, with doses of 0.5 and 10 μmol L^{-1} KI, the concentration of this Cu decreased.

For Mn content, there was a statistically significant difference and an increase of 20.9% in the 25 μmol L^{-1} KI treatment higher than the control. In contrast, there was no significant difference with the 5, 10, and 20 μmol L^{-1} treatments. For Na, the results obtained showed statistically significant differences ($P \le 0.05$). The best treatment was the dose of 25 μmol L^{-1} KI, obtaining an increase of 20.0% compared to the control treatment; on the contrary, with 5 and 10 μmol L^{-1} doses, the Na concentration decreased by 7.5%.

For the microelements Fe and Zn, the results showed a statistically significant difference ($P \le 0.05$); the best treatment was the concentration of 15 μmol L^{-1} KI, obtaining an increase of 53.20 and 28.3%, respectively. For both elements Fe and Zn, a notable decrease of 4.8% and 4.1% in the concentration of this mineral in the lettuce leaves was observed when the application was increased to 25 μmol L^{-1} KI.

Table-4. Impact of KI foliar spraying on mineral content in lettuce crop

(KI) (μmol L^{-1})	N	P	Ca	Mg
	%			
0	2.77±0.09d	0.55±0.00d	1.65±0.01b	0.39±0.00b
5	2.78±0.03d	0.67±0.01b	1.75±0.04a,b	0.39±0.00b
10	3.29±0.02c	0.68±0.00b	1.78±0.02a,b	0.39±0.00b
15	3.34±0.00c	0.62±0.01b	1.81±0.02a	0.41±0.00a
20	3.74±0.04a	0.74±0.01a	1.85±0.01a	0.42±0.00a
25	3.54±0.0b	0.61±0.01c	1.76±0.02a,b	0.38±0.00b
CV	1.400066	1.542416	2.774470	1.310336
R^2	0.990167	0.982366	0.689976	0.885496
DSM	0.1959	0.0274	0.1345	0.0145

(KI) (μmol L^{-1})	Cu	Fe	Zn	Mn	Na
	mg kg^{-1}				
0	5.71±0.12e	283.60±0.51e	42.99±0.13e	120.65±2.93c	0.40±0.00d
5	6.49±0.08d	300.62±0.82d	43.87±0.22d	141.18±0.04b	0.43±0.01c
10	6.89±0.10c	347.79±0.17c	52.30±0.58a	141.74±0.56b	0.43±0.01c
15	7.04±0.12c	667.04±3.44a	55.15±0.0.19a	144.37±1.01ab	0.45±0.00b
20	7.39±0.17b	337.77±9.50b	44.82±0.00c	141.46±0.70b	0.46±0.00b
25	8.26±0.00a	297.15±0.33d	44.75±0.03c	145.91±1.49a	0.48±0.00a
CV	1.646201	1.094291	0.572959	1.042782	1.589004
R^2	0.985920	0.999351	0.997736	0.980833	0.952569
DSM	0.3146	11.374	0.7435	3.9815	0.0194

Different letters are statistically different according to Tukey's test ($P \le 0.05$)

Discussion

Iodine is not yet classified as an essential plant nutrient (Sarrou *et al.,* 2019). However, it is recognized as a beneficial micronutrient for them because iodine is associated with 82 proteins that perform a very important job in the functions of biological processes (Kiferle *et al.,* 2020). Studying this trace element for the benefits attributed to plants and human health is essential because it is related to human thyroid hormones. In this research, the spraying of KI in the plant contributed to increased plant characteristics.

In the case of morphological characteristics, iodine increased the development of the plant, observing a difference in the overall growth of the plant compared between control plants and plants treated with I^-, this effect on the development was presented in other investigations such as in eggplant cultivation (Lara *et al.,* 2023), bell pepper (Cortés *et al.,* 2016), also Chen *et al.* (2024) in lettuce grown in pots confirmed the effect of applying controlled release iodized fertilizer to the soil on biomass gain in both the aerial part and the roots; but, the best results have been obtained when applying iodine compounds by foliar spray in leafy vegetables (Leija-Martínez *et al*., 2016), for example, in a study carried out on spinach under hydroponic cultivation, different chemical forms of iodine, it was found that even with low concentrations an increase in biomass was observed (Weng *et al*., 2008). The KI applications acted topically since there was a direct influence on the increase in the fresh and dry weight values of the aerial parts, as well as the increase in the concentration of photosynthetic pigments, without detracting from the fact that the roots also benefited from increasing the accumulation of biomass although to a lesser extent, as were reported in lettuce foliar biofortified with zinc oxide nanoparticles (Fortis-Hernández *et al.,* 2024). This is because iodine in adequate concentrations is beneficial for plant growth, where iodinated proteins play a vital role in nitrogen metabolism, phytohormone regulation, and energy production in both root and leaf cells (Kiferle *et al.,* 2020).

On the other hand, some reports affirm that there is no significant effect of biofortification with iodine on the morphometric parameters and increasing the pigment content, and the effectiveness of the iodine applied will depend on the species, variety, form of iodine or the type of application (Puccelini *et al.,* 2024). Likewise, iodine interfered negatively with biomass accumulation with high I- doses. This coincides with the report by Blasco *et al.* (2013) and Dávila *et al.* (2019) on lettuce iodine biofortified.

High iodine concentrations can interfere with ROS metabolism and cell signaling, inhibiting growth and other plant processes (Caffagni *et al.,* 2011). It has been found that adequate doses function as an elicitor contributing to plant growth, development, and yield (Lawson *et al.,* 2015).

Iodine is also involved in chlorophyll content, as was observed in a study on fig plants, where the application of KI increased chlorophyll (Salinas *et al.,* 2022). This may be due to the fact that, in adequate concentrations, iodine acts as an auxiliary for the correct metabolism and synthesis of chlorophyll. However, at high concentrations, iodine can be toxic, decreasing stomatal conductance and, in turn, the photosynthetic rate (Medrano *et al.,* 2016).

For the phytochemical part of this work, an increase of bioactive compounds such as phenols was also observed, this effect obtained from KI is due to the fact that this compound is susceptible to oxidizing agents, giving a protective function against abiotic stress causing the increase of ROS due to excess light, salinity and high temperatures, among others (Halka *et al.,* 2020). Therefore, phenolic structures recover their reduced state through a redox balance, interacting with chemical structures of other functional groups through different metabolites. Once oxidized, they recover their hydroxyl group and thus their antioxidant capacity, avoiding the oxidation of other elements such as proteins, nutrients, and sugars (Annunziata *et al.,* 2020). Therefore, as the KI dosage increased, the phenol content increased, and a similar effect was reported by Kiferle *et al.* (2020) in basil cultivation. For flavonoids, it was the same response as in the phenolic compound, probably because it affects the metabolism of nitrogen biosynthesis, proteins, and amino acids, in particular phenylalanine, which is a precursor of phenolic compounds including flavonoids (Germ *et al.,* 2017). This effect is like the results reported by Golubkina *et al.* (2018) in mustard. For antioxidant activity, the best results were with concentrations of 15 and 20 μmol L^{-1} KI in other studies performed on tomatoes. Kiferle *et al.* (2013) observed a similar effect mainly due to an increase in the levels of ascorbic acid in its reduced form. This molecule has a high capacity to donate electrons and directly neutralize free radicals. Suggesting that iodine above a certain threshold could trigger a moderate antioxidant response in fruit, probably against mild stress caused by iodine itself (Blasco *et al.,* 2011).

For vitamin C, there was also a response to the foliar application of KI in lettuce plants, increasing vitamin C as the dose increased. A similar effect was reported by García *et al.* (2014) in nopal, this response was probably caused by the fact that ascorbic acid participates in the detoxification of hydrogen peroxide H_2O_2, as well as the regulation of growth and in the process of xylem lignification, which is why the plant species when affected by abiotic factors, tend to accumulate more ascorbic acid than malate, for lower energy expenditure, this effect causes a tolerance in the plant against oxidative stress (García *et al.,* 2014).

As previously mentioned, iodine plays a vital role in the physiological and biochemical responses of the plant (Smoleń *et al.,* 2017). Still, it is also a very important element in human health due to its involvement in the functioning of thyroid hormones, which control the body's metabolism (Dobosy *et al.,* 2020). Iodine is an element that is absorbed by the root or through cuticular waxes with a high degree of unsaturation and high capacity to take up iodine (Shaw *et al.,* 2007), both in dissolved form and in gas form as I_2 and CH_3 I, cuticular waxes that interact with iodine can be alternatives for pre- and post-harvest biofortification of fruits and seeds (Medrano *et al.,* 2016). Once iodine is taken up, it is transported through the xylem, finding its redistribution through the phloem to be low, so it accumulates in more significant amounts in leaves than in fruits and seeds. However, in lettuce plants treated with iodine via the foliar route, evidence of iodine transport from leaves to roots has been found (Smoleń *et al.,* 2015).

In the present investigation, the iodine content in the lettuce leaf was increased compared to the control treatment. As the dose was increased, the iodine present in the plant was increased; this effect is similar to other studies, such as in chili cultivation (Cortes, 2016), in bell pepper (Li *et al.,* 2017) in lettuce (Smoleń *et al.,* 2022).

The best regression fit was obtained with the polynomial type in each of the variables analyzed by this method, this could be due to following the pattern of diminishing returns which consists of the increase in an element leading to an increase in performance, however, it reaches a point that begins to decrease as it was in the case of the variables of fresh weight, phenols and flavonoids total and vitamin C and a second explanation are iodine may contribute to phytotoxicity at higher concentrations affecting performance (Medrano-Macías *et al*., 2016).

But in turn, I- interacted with other minerals, both macroelements and microelements, with some causing synergism or antagonism. This causes a synergistic response with the following macroelements: N, P, Ca^{+2}, and Mg^{+2}. This indicates that the application of KI interacts synergistically with N at adequate concentrations, giving a beneficial effect on crop growth and production; however, at high concentrations, it probably causes a phytotoxic impact by altering the NO transporters, causing antagonism between this and iodine (Blasco, 2011). For P and N, significant differences were presented, obtaining an increase with respect to the control. Similar results were obtained by Blasco (2013), when applying concentrations higher than 20 μmol of iodine, he observed a decrease in P in the lettuce crop. This is because high concentrations of iodine cause antagonism and interfere with specific transporters for P absorption (Medrano *et al.,* 2016).

The calcium (Ca) and magnesium (Mg) elements increased with respect to the control. This result was similar to that reported by Sularz *et al.* (2020), who, upon applying salicylic iodine, found that both Mg and Ca positively affected plant growth due to their essential functions, such as protein synthesis and photosynthesis, which allows us to infer that the increase in micronutrients uptake performed as a cofactor to increase in fresh and dry weight, as well as the size of the lettuce.

On the other hand, for microelements such as Cu^{+2}, Fe^{+2}, Zn^{+2}, and Mn^{+2}, the results showed an increase concerning the control. This shows a synergistic effect resulting in increased uptake of both elements in the leaves. Possibly, the relationship between Cu and Iodine can oxidize I- to I_2 or HIO through copper oxidases enzyme, or probably, that higher amount of I- in plant tissue could increase the enzyme activity such as copper oxidases and possibly, Mn oxidases that dissipate the element (Schlorke *et al.,* 2016).

The treatment of 15 μmol L^{-1} KI accumulated Fe and Zn microelements in greater quantity than the control treatment. On the one hand, Smoleń *et al.* (2014) reported that the application of KIO_3 and SeO in lettuce plants did not affect the mineral composition. But on the other hand, Smoleń *et al.* (2015) found a negative correlation between Fe, Zn, and Cd with iodine content, this may occur due to a redox phenomenon by oxidation of I- to I_2 provides a reducing potential, thus having an impact on the bioavailability of these elements (Kato *et al.,* 2013).

Conclusions

In this study, foliar application of potassium iodide (KI) at concentrations of 15 and 20 μmol L^{-1} provided significantly better results in most of the variables evaluated in lettuce. These concentrations not only improved the plant's morphological characteristics, providing more robust development and greater resistance to abiotic factors but also positively influenced the mineral interaction and the phytochemical profile of lettuce. This phytochemical enrichment is crucial, as it turns lettuce into a functional food that can provide health benefits to the consumer by increasing the availability of essential nutrients such as iodine and contributing to the prevention of deficiency-related diseases. These findings support biofortification as a viable strategy to increase dietary iodine intake, which is particularly relevant in contexts where deficiency of this micronutrient remains a public health problem. However, it is necessary to continue researching and applying this technique to other plant species to broaden its scope and efficacy in crop nutritional improvement.

Authors' Contributions

Investigation: MFH, JSA and PPR; Formal analysis: MFH and APGG; Visualization: MFH, PPR, APGG ASE. and JSA; Writing—original draft: MFH, ASE, and JSA; Writing—review and editing,: MFH, PPR, APGG and JSA. All authors read and approved the final manuscript.

Ethical approval (for researches involving animals or humans)

Not applicable.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

References

Allen LH, De Benoist B, Dary O, Hurrell R (2017). Guías para la fortificación de alimentos con micronutrientes. In Guías para la fortificación de alimentos con micronutrientes. [Guidelines for food fortification with micronutrients]. Organización Mundial de la Salud. *https://iris.who.int/handle/10665/255541*

Annunziata G, Jiménez M, Capó X, Moranta D, Arnone A, Tenore GC, Sureda A, Tejada S (2020). Microencapsulation as a tool to counteract the typical low bioavailability of polyphenols in the management of diabetes. Food and Chemical Toxicology 139:111248. *https://doi.org/10.1016/j.fct.2020.111248*

Blasco B, Leyva R, Romero L, Ruiz JM (2013). Iodine effects on phenolic metabolism in lettuce plants under salt stress. Journal of Agricultural and Food Chemistry 61(11):2591-2596. *https://doi.org/10.1021/jf303917n*

Blasco B, Rios JJ, Cervilla L. M, Sánchez-Rodrigez E, Ruiz JM, Romero L (2008). Iodine biofortification and antioxidant capacity of lettuce: potential benefits for cultivation and human health. Annals of Applied Biology 152(3):289-299. *https://doi.org/10.1111/j.1744-7348.2008.00217.x*

Blasco B, Ríos JJ, Leyva R, Cervilla LM, Sánchez E, Rubio M, … Romero L (2011). Does iodine biofortification affect oxidative metabolism in lettuce plants. Biological Trace Element Research 142(3):831-842. *https://doi.org/10.1007/s12011-010-8816-9*

Caffagni A, Arru L, Meriggi P, Milc J, Perata P, Pecchioni N (2011). Iodine fortification plant screening process and accumulation in tomato fruits and potato tubers. Communications in Soil Science and Plant Analysis 42(6):706-718. *https://doi.org/10.1080/00103624.2011.550372*

Chen H, Jiang F, Zhou H, Zhang Z, Zhang Z, Liu B (2024). Controlled-release iodine fertilizer improved iodine accumulation, antioxidant capacity, and quality of lettuce. Agronomy 14(11):2578. *https://doi.org/10.3390/agronomy14112578*

Cortés C, Rodríguez MN, Benavides A, García JL, Tornero M, Sánchez P (2016). El yodo aumenta el crecimiento y la concentración de minerales en plántulas de pimiento morrón [Iodine increases the growth and mineral concentration in sweet pepper seedlings]. Agrociencia 50(6):747-758.

Dávila IE, Leija P, Medrano J, Fuentes LO, González S, Juárez A, Benavides A (2019). Iodine biofortification of crops. In: Jaiwal PK, Chhillar AK, Chaudhary D. Nutritional quality improvement in plants. Concepts and strategies in plant sciences. Springer. Cham, Switzerland, pp 79-113. *https://doi.org/10.1007/978-3-319-95354-0_4*

Dobosy P, Vetési V, Sandil S, Endrédi A, Kröpfl K, Óvári M, … Záray G (2020). Effect of irrigation water containing iodine on plant physiological processes and elemental concentrations of cabbage (*Brassica oleracea* L. var. capitata L.) and tomato (*Solanum lycopersicum* L.) cultivated in different soils. Agronomy 10(5):720. *https://doi.org/10.3390/agronomy10050720*

Dobosy, P, Nguyen HTP, Záray G, Streli Ch, Ingerle D, Ziegler Ph, … Fodor F (2024). Effect of iodine species on biofortification of iodine in cabbage plants cultivated in hydroponic cultures. Science Reports 14:15794. *https://doi.org/10.1038/s41598-024-66575-z*

Duborská E, Urík M, Šeda M (2020). Iodine biofortification of vegetables could improve iodine supplementation status. Agronomy 10(10):1574. *https://doi.org/10.3390/agronomy10101574*

FAO.Organización de las Naciones Unidas para la Alimentación y la Agricultura, el Fondo Internacional de Desarrollo Agrícola (ops), la Organización Panamericana de la Salud (OPS), el Programa Mundial de Alimentos (WFP) y el Fondo de las Naciones Unidas para la Infancia (UNICEF) 2020. Panorama de la seguridad alimentaria y nutrición en América Latina y el Caribe [Regional Overview of FoodSecurity and Nutrition in Latin America and the Caribbean]. Santiago de Chile. *https://doi.org/10.4060/cb2242es*

Figueiredo M, Prieto H, Filomeno EA, Carmo, C, Silva JM (2022). Agronomic biofortification with iodine in lettuce plants cultivated in floating hydroponic system. Revista Ceres 69(2):210-217. *https://doi.org/10.1590/0034-737X202269020012*

Fortis-Hernández M, Sánchez-Estrada A, Hernández-Cruz D, Lagunes-Fortiz E, Betancourd-Galindo R, Fortiz-Hernández J (2024). La biofortificación con nanopartículas de óxido de zinc vía foliar aumenta la acumulación de biomasa y calidad fitoquímica de la lechuga. ITEA-Información Técnica Económica Agraria 120(4):344-359. *https://doi.org/10.12706/itea.2024.009*

García HT, Benavides A, Rivas C, Morales EM, Verde MJ, Ruvalcaba RM (2014). Iodine application increased ascorbic acid content and modified the vascular tissue in *Opuntia ficus-indica* L. Pakistan Journal of Botany 46 (1):127-134.

Germ M, Luthar Z, Benković E, Zhou M, Golob A, Kacjan N (2017). Fagopyrin and rutin concentration in seeds of common buckwheat plants treated with Se and I / Vsebnost fagopirina in rutina v semenih navadne ajde tretirane s selenom in jodom. Folia Biologica et Geologica 58(2):45-51. *https://doi.org/10.3986/fbg0027*

Golubkina N, Kekina H, Caruso G (2018). Yield, quality and antioxidant properties of Indian mustard (*Brassica juncea* L.) in response to foliar biofortification with selenium and iodine. Plants 7(4):80. *https://doi.org/10.3390/plants7040080*

Halka M, Smoleń S, Ledwożyw I (2020). Antioxidant potential and iodine accumulation in tomato (*Solanum lycopersicum* L.) seedlings as the effect of the application of three different iodobenzoates. Folia Horticulturae 32(2):203-219. *https://doi.org/10.2478/fhort-2020-0019*

Hidalgo A, Šaponjac V, Ćetković G, Travičić V, Čanadanović BJ, Chiosa D, Brandolini A (2019). Antioxidant properties and heat damage of water biscuits enriched with sprouted wheat and barley. LWT (114):108423. *https://doi.org/10.1016/j.lwt.2019.108423*

Hsu CL, Chen W, Weng YM, Tseng CY (2003). Chemical composition, physical properties, and antioxidant activities of yam flours as affected by different drying methods. Food Chemistry 83(1):85-92. *https://doi.org/10.1016/S0308-8146(03)00053-0*

Ikram NA, Ghaffar A, Khan AA, Nawaz F, Hussain A (2024). Foliar iodine application: A strategy for tomato biofortification and yield optimization. Journal of Plant Nutrition *48*(3):540–556. *https://doi.org/10.1080/01904167.2024.2407483*

Kato S, Wachi T, Yoshihira K, Nakagawa T, Ishikawa A, Takagi D, … Takahashi M (2013). Arroz (*Oryza sativa* L.) las raíces tienen actividad de reducción de yodato en respuesta al yodo. Plant Science (4):227. *https://doi.org/10.3389/fpls.2013.00227*

Kawashima LM, Valente LM (2003). Mineral profile of raw and cooked leafy vegetables consumed in Southern Brazil. Journal of Food Composition and Analysis 16(5):605-611. *https://doi.org/10.1016/s0889-1575(03)00057-7*

Kiferle C, Ascrizzi R, Martinelli M, Gonzali S, Mariotti L, Pistelli L (2020). Effect of Iodine treatments on *Ocimum basilicum* L.: Biofortification, phenolics production and essential oil composition. PLoS One 14(12):226559. *https://doi.org/10.1371/journal.pone.0226559*

Kiferle C, Gonzali S, Holwerda HT, Real R, Perata P (2013). Tomato fruits: a good target for iodine biofortification. Frontiers (4):4-7. *https://10.3389/fpls.2013.00205*

Kiferle C, Martinelli M, Salzano AM, Gonzali S, Beltrami S, Salvadori PA, … Perata P (2021). Evidences for a nutritional role of iodine in plants. Frontiers Plant Science 12:616868. *https://doi.org/10.3389/fpls.2021.616868*

Krishna, Jain A, Verma A (1992). Determination of iodide by high-performance liquid chromatography after precolumn derivatization. analytical. Chemistry 64 (13):1484-1489. *https://doi.org/10.1021/ac00037a030*

Krzepilko A, Prazak R, Skwarylo-Bednarz B, Molas J (2019). Agronomic biofortification as a means of enriching plant foodstuffs with iodine. Acta Agrobotanica 72(2):1-9. *http://dx.doi.org/10.5586/aa.1766*

Landini M, Gonzali S, Perata P (2011). Iodine biofortification in tomato. Journal of Plant Nutrition and Soil Science 174(3):480-486. *https://doi.org/10.1002/jpln.201000395*

Lara AY, Rojas ÁN, Alcalá JA, Alia I, Méndez H, Hernández A (2023). Biofortificación con yodo, actividad antioxidante y minerales en frutos de berenjena (*Solanum melongena* L.) [Iodine biofortification and antioxidant activity in eggplant fruits (*Solanum melongena* L.)] Revista Fitotecnia Mexicana 46(4):399-399. *https://doi.org/10.35196/rfm.2023.4.399*

Lawson PG, Daum D, Czauderna R, Meuser H, Härtling JW (2015). Soil versus foliar iodine fertilization as a biofortification strategy for field-grown vegetables. Frontiers Plant Science 6:1-11. *https://doi.org/10.3389/fpls.2015.00450*

Ledwożyw-Smoleń I, Smoleń S, Rożek S, Sady W, Strzetelski P (2020). Iodine biofortification of potato (*Solanum tuberosum* L.) grown in field. Agronomy 10(12):1916. *https://doi.org/10.3390/agronomy10121916*

Leija P, Benavides A, Rocha A, Medrano JR (2016). Biofortificación con yodo en plantas para consumo humano. Revista Mexicana de Ciencias Agrícolas 7(8):2025-2036. *https://doi.org/10.29312/remexca.v7i8.134*

Li R, Wang D, Ping H, Lai C, Yi M, Xi Z, … Xin H (2017). Enhancing iodine content and fruit quality of pepper (*Capsicum annuum* L.) through biofortification. Scientia Horticulturae 214:165-173. *https://doi.org/10.1016/j.scienta.2016.11.030*

Lichtenthaler HK, Wellburn AR (1983). Determinations of total carotenoids and chlorophylls a and b of leaf extracts in different solvents. Biochemical Society Transactions 11(5):591-592. *https://doi.org/10.1042/bst0110591*

Lopez E (2022). Neuronutrition: repercussions of nutritional excesses and deficiencies Neuronutrição: o impacto dos excessos e deficiências nutricionais. Nutrición Clínica y Metabolismo 5(3):69-82. *https://doi.org/10.35454/rncm.v5n3.367*

Medrano-Macias J, Leija P, González S, Juárez A, Benavides A (2016). Use of Cr iodine to biofortify and promote growth and stress tolerance in crops. Frontiers in Plant Science 7:54-66. *https://doi.org/10.3389/fpls.2016.01146*

Medrano-Macias J, López-Caltzontzit MG, Rivas-Martínez EN, Narváez-Ortiz WA, Benavides-Mendoza A, Martínez-Lagunes PJ (2021). Enhancement to salt stress tolerance in strawberry plants by iodine products application. Agronomy 11:602. *https://doi.org/10.3390/agronomy11030602*

Ofori KF, Antoniello S, English MM, Aryee AN (2022). Improving nutrition through biofortification–A systematic review. Frontiers in Nutrition 9:1043655. *https://doi.org/10.3389/fnut.2022.1043655*

Padayatt S, Daruwala R, Wang Y, Eck PK, Song J, Koh WS, Levine M (2001). Vitamin C: from molecular actions to optimum intake. In: Cadenzas E, Packer L (Eds). Handbook of Antioxidants. CRC press. Washington DC, USA pp 117-145.

Plank C (1992). Plant analysis reference procedures for the southern region of the United States. Southern Cooperative Series Bulletin 368:1-13.

Puccinelli M, De Padova A, Vernieri P, Carmassi G, Incrocci L (2024). Response of aeroponically cultivated baby-leaf lettuce (*Lactuca sativa* L.) plants with different zinc, copper, iodine, and selenium concentrations. Horticulturae 10(7):726. *https://doi.org/10.3390/horticulturae10070726*

Rakoczy-Lelek R, Smoleń S, Grzanka M, Ambroziak K, Pitala J, Skoczylas Ł, … Kardasz H (2021). Effectiveness of foliar biofortification of carrot with iodine and selenium in a field condition. Frontiers in Plant Science 12:656283. *https://doi.org/10.3389/fpls.2021.656283*

Riyazuddin R, Singh K, Iqbal N, Nisha N, Rani A, Kumar M, … Gupta R (2023). Iodine: an emerging biostimulant of growth and stress responses in plants. Plant and Soil 486(1):119-133. *https://doi.org/10.1007/s11104-022-05750-5*

Salinas PA, Mendez FJ, Ortiz JC, Niño G, Sáenz E, Garza CA (2022). Iodine increases the concentration of phenolic compounds and photosynthetic pigments in three cultivars of *Ficus carica* L. subjected to salt stress. Revista Mexicana de Ciencias Agrícolas (28):309-318. *https://doi.org/10.29312/remexca.v13i28.3285*

Sarrou E, Siomos AS, Riccadona S, Aktsoglou DC, Tsouvaltzis P, Angeli A, … Martens S (2019). Improvement of sea fennel (*Crithmum maritimum* L.) nutritional value through iodine biofortification in a hydroponic floating system. Food Chemistry (296):150-159. *https://doi.org/10.1016/j.foodchem.2019.05.190*

Schlorke D, Flemmig J, Birkemeyer C, Arnhold J (2016). Formation of cyanogen iodide by lactoperoxidase. Journal of Inorganic Biochemistry 154:35-41. *https://doi.org/10.1016/j.jinorgbio.2015.11.005*

Shaw G, Scott LK, Kinnersley RP (2007). Sorption of caesium, iodine and sulphur in solution to the adaxial leaf surface of broad bean (*Vicia faba* L.). Environmental and Experimental Botany 59(3):61-370. *https://doi.org/10.1016/j.envexpbot.2006.04.008*

Sida JP, Sánchez E, Ávila GD, Zamudio PB (2015). Biofortificación con micronutrientes en cultivos agrícolas y su impacto en la nutrición y salud humana [Micronutrient biofortification in agricultural crops and their impact on human nutrition and health]. Tecnociencia Chihuahua 9(2):67-74. *https://doi.org/10.54167/tch.v9i2.591*

Singleton VL, Orthofer R, Lamuela RM (1999). Analysis of total phenols and other oxidation substrates and antioxidants by means of folin-ciocalteu reagent. Methods in Enzymology 299(7):152-178. *https://doi.org/10.1016/S0076-6879(99)99017-1*

Smoleń S, Kowalska I, Sady W (2014). Assessment of biofortification with iodine and selenium of lettuce cultivated in the NFT hydroponic system. Scientia Horticulturae 166:9-16. *https://doi:10.1016/j.scienta.2013.11.011*

Smoleń S, Kowalska I, Skoczylas Ł, Tabaszewska M, Pitala J, Mrożek J, Kováčik P (2022). Effectiveness of enriching lettuce with iodine using 5-iodosalicylic and 3,5-diiodosalicylic acids and the chemical composition of plants depending on the type of soil in a pot experiment. Food Chemistry 382:132347, *https://doi.org/10.1016/j.foodchem.2022.132347*

Smoleń S, Ledwożyw I, Halka M, Sady W, Kováčik P (2017) The absorption of iodine from 5-iodosalicylic acid by hydroponically grown lettuce. Scientia Horticulturae 225:716-725. *https://doi.org/10.1016/j.scienta.2017.08.009*

Smoleń S, Wierzbińska J, Włodzimierz S, Kołton A, Wiszniewska A, Liszka M (2015). Iodine biofortification with additional application of salicylic acid affects yield and selected parameters of chemical composition of tomato fruits (*Solanum lycopersicum* L.). Scientia Horticulturae 188:89-96. *https://doi.org/10.1016/j.scienta.2015.03.023*

Steiner A (1961). A universal method for preparing nutrient solutions of a certain desired compositions. Plant and Soil 15(2):134-154. *https://link.springer.com/article/10.1007/BF01347224*

Sularz O, Smoleń S, Koronowicz A, Kowalska I, Leszczyńska T (2020). Chemical composition of lettuce (*Lactuca sativa* L.) biofortified with iodine by KIO3, 5-Iodo-, and 3.5-Diiodosalicylic acid in a hydroponic cultivation. Journal Agronomy 10(7):2-17. *https://doi.org/10.3390/agronomy10071022*

Verma KK, Jain A, Verma A (1992). Determination of iodide by high-performance liquid chromato graphy after precolumn derivatization. Analytical Chemistry 64(13):1484-1489. *https://pubs.acs.org/doi/pdf/10.1021/ac00037a030*

Wang T, Lin Z, Zhang W, Qin Z (2023). Study of the rapid determination of phosphorus in electronic packaging material by phosphorus-molybdenum yellow spectrophotometry. Journal of Electronic Materials 52(11):7590-7595. *https://doi.org/10.1007/s11664-023-10689-z*

Weng HX, Yan AI, Hong CL, Xie LL, Yong WB, Qin YC (2008). Increment of iodine content in vegetable plants by applying iodized fertilizer and the residual characteristics of iodine in soil. Biological Trace Element Research 123(1-3): 218-228. *https://doi.org/10.1007/s12011-008-8094-y*

Zhang Y, Cao H, Wang M, Zou Z, Zhou P, Wang X, Jin J (2023). A review of iodine in plants with biofortification: uptake, accumulation, transportation, function, and toxicity. Science of the Total Environment 878:163203. *https://doi.org/10.1016/j.scitotenv.2023.163203*

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