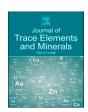
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Selenium in plant-based beverages: How can *in vitro* bioaccessibility contribute to an accurate daily intake?

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ABSTRACT

Background: Lactose intolerance, cow milk protein allergy, and environmental and ethical concerns drive the global market for plant-based beverages (PPBs). Despite this, data on the occurrence of selenium (Se) in these beverages are scarce.

Objective: The objectives of this study were to assess total selenium and determine the bioaccessible fractions in plant-based beverages (PBBs) from cereals, oilseeds, and pumpkin seeds, using an *in vitro* static method to simulate human digestion; and estimate the contribution these beverages have to the reference daily intake (RDI) for children and adults.

Methodology: The study involved beverages made under laboratory conditions. Samples and extracts were submitted to acid digestion (ultrasonic and block digester); total and bioaccessible Se levels were determined by ICP-MS. *In vitro* bioaccessibility was assessed using the INFOGEST protocol.

Results: The results revealed a wide variation in Se content in the studied plant-based beverages from $10.3~\mu g~kg^{-1}$ (sunflower seeds) to $3509~\mu g~kg^{-1}$ (Brazil nuts). The bioaccessibility of Se was found only in PBBs made from oilseeds with macadamia, cashew and Brazil nut, with a variation in values from 86 to 96 %. The Se content and its bioaccessible fraction of Brazil nut PBB may contribute to 1276~% and 1218~% of the RDI for children and adults, respectively.

Conclusions: The Se content varied among samples of cereals, oilseeds and pumpkin seeds, reflecting the diversity in the studied PBBs. Only PBBs containing oilseeds (macadamia, cashews, and Brazil nuts) exhibited measurable amounts of bioaccessible Se, indicating an association with the raw material. However, the consumption of the PBB containing Brazil nuts may pose a risk of adverse effects, due to its high contribution to the RDI in both children and adults.

1. Introduction

The global market for plant-based beverages (PBBs) was valued at \$27.3 billion in 2022, and with an annual growth rate of 10.4 %, is projected to reach \$44.8 billion by 2027 [1]. This growth is directly correlated with the health benefits associated with plant-based diets [2]. Furthermore, the consumption of PBBs can contribute to an adequate intake of Se, becoming crucial for health; especially given the increasing changes in dietary patterns. In this context, the global consumption of PBBs is also driven by health concerns (lactose intolerance, and search for healthy foods), the environment (reduction of greenhouse gas emissions from dietary choices) and ethics (animal welfare) [1,2].

The PBBs are produced by macerating grains in water at different ratios, ranging from 2 to 15 %. The main raw materials used include

cereals (rice, oats), pseudocereals (quinoa), legumes (soy, peanuts, peas), oilseeds (almonds, hazelnuts, walnuts, cashews, Brazil nuts), fruits (coconut), and other seeds (sesame) [1,3]. Vegetables such as hemp and macadamia have also been used [4]. PBBs can contribute appreciable amounts of unsaturated fatty acids, essential minerals, phenolic compounds, and other bioactive compounds [5]. Furthermore, plant-based beverages can be classified as functional foods and nutraceuticals due to the presence of dietary fibers, vitamins, antioxidant compounds, and some trace elements, such as selenium [6].

In adequate amounts, Se plays numerous roles in the body, which includes assisting in the protection against free radicals and regulating the immune and reproductive systems [7]. However, high dietary intakes of Se can contribute to the onset of chronic illnesses, such as diabetes and cardiovascular diseases [7]. In this context, a reference daily

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intake (RDI) of Se has been established, varying according to age groups. For example, for adults and children (\geq 4 years old), an RDI of 55 µg has been set, while for pregnant and lactating women, the RDI is 70 µg [8].

Food undergoes transformations in the gastrointestinal tract, with part of the nutrients being absorbed and used in biological functions [9]. Static *in vitro* gastrointestinal digestion models [10], such as the INFO-GEST protocol, simulate the physiological conditions of the organism during digestion; using synthetic fluids containing mineral salts and enzymes with known activity. Dynamic *in vitro* gastrointestinal digestion conditions, such as simulation of mechanical degradation, pH variations, and gastric emptying were not considered in these static protocols [11].

Although it is of utmost importance, there is still a noticeable lack of data regarding the presence of Se and its bioaccessibility in PBBs. Orlando et al. [12] evaluated the total Se content in beverages made from almonds, rice, oat, cashew (with or without Brazil nuts), coconut, and soy; and reported Se levels ranging from <1.2 to $160.5~\mu g~kg^{-1}.$ Astolfi et al. [4] studied PBBs produced with different raw materials and reported Se levels below the limit of quantification (LOQ) for plant-based beverages of almonds, hazelnut, walnut, hemp, and quinoa. The determination of bioaccessible trace element contents, such as Se, still poses an analytical challenge [13,14], with techniques of high sensitivity, such as the inductively coupled plasma mass spectrometer (ICP-MS), being used [15,16].

Given the above, this study aimed to investigate the total content and estimate the bioaccessibility percentages of Se by ICP-MS (using the INFOGEST protocol) in plant-based beverages prepared from cereals (oats and rye), oilseeds (hazelnuts, almonds, walnuts, sunflower seeds, macadamia nuts, cashews, and Brazil nuts), and pumpkin seeds. The results obtained will be used to calculate the intake estimate of ingestion, considering the RDI for children and adults. These findings may influence dietary guidelines and consumers' decisions about Se intake from PBBs, providing relevant data for more informed food choices.

2. Material and methods

2.1. Reagents

Concentrated nitric acid (Synth, Diadema, Brazil) purified by subboiling distillation (Berghof, Eningen, Germany); analytical grade hydrochloric acid (37 %) and hydrogen peroxide (30 %) (Merck, Darmstadt, Germany); salivary α -amylase (50 U/mg); porcine pepsin (3843 U/mg); porcine pancreatin (7,3 U/mg) and bile salts (200 mg/mL); sodium bicarbonate; ammonium carbonate; calcium chloride; magnesium chloride; potassium chloride; sodium chloride; potassium phosphate; analytical degree or higher sodium hydroxide (Sigma-Aldrich, St. Louis, EUA); certified reference materials (CRM) for skimmed milk powder (ERM-BD 151, European Commission, Geel, Belgium); peach leaves (NIST 1547, Gaithersburg, EUA) and standard solutions of Se, Ge and In (1000 mg/L, Quimlab, Jacareí, Brazil).

2.2. Equipment

Ultrasonic bath (Easy 180H, Elma, Singen, Germany); digester block and water bath with agitation (Tecnal, Piracicaba, Brazil); centrifuge (Centrifuge 5804 R, Hamburg, Germany) and inductively coupled plasma mass spectrometer (ICP-MS iCAP RQ, Thermo Fisher Scientific, Bremen, Germany) with optimized experimental conditions: Air flow rate, 14 L/min; Auxiliary air, 0.80 L/min; He flow rate, 5.0 mL/min; Nebulizer flow rate, *Micromist* 0.98 L/min; monitored isotopes, 78 Se, 82 Se, 72 Ge, 74 Ge and 115 In, and analytical curve in the range of 0.1 to 100 µg L^{-1} .

2.3. Samples

Samples of cereals (oats and rye), oilseeds (hazelnuts, almonds,

walnuts, sunflower seeds, macadamia nuts, cashews, and Brazil nuts) and pumpkin seeds were purchased at the Municipal Market in the city of Campinas-SP (Brazil), on three different days. Three samples of each raw material (cereals, nuts and pumpkin seeds) were obtained and used to evaluate the total Se content, as well as for use in the preparation of PBBs produced in the laboratory (non-commercial vegetable drinks) (n=3). All raw materials and PBBs were analyzed in triplicate, totaling 90 samples. Subsequently, samples were surface sterilized and maintained overnight (17 h) in water. After, samples were blended in a household blender for 3 min at a ratio of 1:4 (cereal, oilseed, and/or fruit: water), as described by Cunha Júnior et al. [15]. The beverages were filtered through a 0.25 mm polymeric membrane, prepared in triplicate, and refrigerated until the time of analysis.

2.4. Analytical procedure

All determinations were carried out in triplicate in the presence of analytical blanks.

2.4.1. Determination of total Se content

0.5 g of sample was weighed into graduated tubes (Corning, NY, USA), and added to 4 mL of nitric acid, and then left to rest overnight. Next, 2 mL of hydrogen peroxide was added, and acid digestion was carried out in an ultrasonic bath at 80 $^{\circ}\text{C}$ for 35 min. The digested solution was cooled, purified water was added to make up 20 mL, and then filtered for ICP-MS analysis to avoid undissolved constituents (PTFE, 0.45 μm , Agilent Technologies, Tokyo, Japan) [16].

2.4.2. Bioaccessible fraction of Se

For the estimation of Se bioaccessibility, the standardized *in vitro* digestion protocol proposed by INFOGEST (composed of oral, gastric, and intestinal phases) was employed [11,17]; using 2.5 g of plant-based beverage samples. The bioaccessible fraction was mineralized in a block digester at a maximum temperature of 130 °C for 4 h, using 4 mL of nitric acid and 2 mL of hydrogen peroxide [18].

2.4.3. Analytical control

The method was validated and deemed suitable according to AOAC [19] recommendations, considering the following figures of merit: linearity (r² \geq 0.99); limit of detection (LOD) (2.1 µg kg $^{-1}$) and limit of quantification (LOQ) (4.0 µg kg $^{-1}$); accuracy, with recovery values between 99 \pm 8 % (skimmed milk powder) and 111 \pm 6 % (peach leaves), as well as, 95 \pm 1 % and 101 \pm 5 % (spiked experiments); and repeatability, with coefficients of variation below 9 %. For bioaccessibility assays, the LOQ was 20 µg kg $^{-1}$ [20].

2.5. Contribution to reference daily intake (RDI) of Se

For calculating the contribution to Se RDI, a daily intake of 200 mL of plant-based beverage was considered for adults and children (\geq 4 years old), considering the average Se levels found in PBBs and their respective bioaccessible fractions (BF) [21]. The RDI value considered was 55 μ g, as established by the FDA [8].

2.6. Statistical analysis

The results were evaluated by analysis of variance (ANOVA, oneway), F-test, Tukey's test (95 % confidence), and coefficient of variation (CV) using Microsoft Office Excel (version 2016) and Statgraphics Centurion XVI (StatPoint Technologies, Inc. USA, 2010) software.

3. Results and discussion

3.1. Occurrence of selenium in the raw materials (cereals, oilseeds, and pumpkin seeds) and plant-based beverages

The levels of Se in the oats and rye (cereals), sunflower seeds, hazelnuts, almonds, walnuts, macadamia nuts, cashews, and Brazil nuts (oilseeds), and fruit (pumpkin seeds) raw materials, and in the beverages prepared from these ingredients, are presented in Table 1.

Selenium levels were detected in all evaluated raw materials (cereals, oilseeds, and pumpkin seeds), with the following behavior observed: Brazil nuts > cashews > pumpkin seeds > macadamia > sunflower seeds > walnuts > rye > almonds > hazelnuts > oats. Selenium levels varied from 6.9 $\mu g~kg^{-1}$ (oats) to 28,148 $\mu g~kg^{-1}$ (Brazil nuts), with the lowest levels found in oat and rye cereals (6.9 and $18 \mu g$ kg⁻¹, respectively). In the category of oilseeds, the lowest levels were found in hazelnuts and almonds (12.4 and 16.6 $\mu g \ kg^{-1}$, respectively). Although hazelnuts and almonds have low levels of Se, the other oilseeds could be considered rich in this trace element, with average values of 70.4, 137, 239, and 310 μ g kg⁻¹ for walnuts, sunflower seeds, macadamia, and cashews, respectively. The geographical location, soil type used in cultivation, use of fertilizers, and the bioavailability of different species contained in these plants influence Se levels [22]. Markiewicz-Żukowska et al. [23] evaluated various types of oilseeds and reported average Se levels higher than this study; 37, 46, 80, and 354 µg kg⁻¹ for almonds, hazelnuts, walnuts, and cashews, respectively.

For the plant-based beverages prepared using these raw materials, a similar pattern in Se levels was observed: Brazil nuts > cashews > macadamia > pumpkin > walnuts > sunflower seeds. The plant-based beverages derived from almonds, oat, hazelnut and rye showed Se values below the LOQ (< 4 $\mu g \ kg^{-1}$); behavior similar to that reported by De Paiva et al. [24] and Redan et al. [25] in commercial plant-based beverages samples of almonds, rice, oat, and soy.

From Table 1, the high levels of Se observed in Brazil nuts grown in Se-rich soil (Pará, Brazil) and in its plant-based beverage (28,148 and 3509 $\mu g\ kg^{-1}$, respectively) stands out. High levels of Se were reported by Sartori et al. [26] in their study of Brazil nuts (8714 $\mu g\ kg^{-1}$) and plant-based beverages (1060 $\mu g\ kg^{-1}$), and was related to the origin of the Brazil nut. Buffini et al. [27] reported that Se levels in foods, especially in wheat, cereals, and nuts, can vary according to the geochemistry of the soils where these products are cultivated. The Se content in plants is directly influenced by the Se content in the soil and water, varying geographically within and between countries where they are cultivated [28,29].

The beverages prepared with sunflower seeds, walnuts, and pumpkin seeds showed levels of 10.3, 12, and 19 μ g kg⁻¹, respectively; with the

Table 1
Selenium (Se) levels present in raw materials (cereals, oilseeds, and pumpkin seeds) and in plant-based beverages.

Categories*	Types	Se total ($\mu g \ kg^{-1}$)		
		Raw material	Plant-based beverages	
Cereals	Oat	$6.9 \pm 0.1^{\rm i}$	n.d.	
	Rye	18 ± 2^g	n.d.	
Oilseeds	Hazelnut	$12.4\pm0.5^{\rm h}$	n.d.	
	Almonds	$16.6\pm0.3^{\rm g}$	n.d.	
	Nuts	$70.4\pm0.5^{\rm f}$	$12\pm1^{\rm e}$	
	Sunflower seeds	$137\pm7^{\rm e}$	$10.3\pm0.1^{\rm e}$	
	Macadamia	$239\pm10^{\rm d}$	$35\pm2^{\mathrm{c}}$	
	Cashew nut	$310\pm10^{\rm b}$	$44 \pm 1^{\mathrm{b}}$	
	Brazil nuts	$28{,}148\pm49^a$	3509 ± 23^a	
Seeds	Pumpkin seeds	278 ± 5^{c}	19 ± 1^{d}	

According to Rego et al. [3]; n.d = not detected (< 4 μ g kg⁻¹). Results are expressed as mean \pm standard deviation (n = 3).

Different letters in the same column indicate significant differences (P < 0.05; ANOVA + Tukey's Test).

first two not being significantly different at the 95 % confidence level, and the results were similar to those reported by Vasquez-Rojas et al. [5] The beverages prepared with macadamia and cashew nuts showed Se levels of 35 and 44 $\mu g \ kg^{-1}$, respectively; higher than those reported by Orlando et al. [12] in commercial samples of cashew nuts (20 to 30 $\mu g \ kg^{-1}$).

In general, the Se content in plant-based beverages represented 6.8 % (pumpkin seeds), 7.5 % (sunflower seeds), 12.5 % (Brazil nuts), 14.2 % (cashews), 14.6 % (macadamia), and 17.0 % (walnuts) of the content found in the raw materials used. Factors such as the composition and solubility of proteins and their selenoamino acids can contribute to the transfer of Se. Sartori et al. [26] found that vegetable seeds have a high concentration of albumins (soluble in aqueous medium), while plant-based beverages have higher levels of globulins and/or convicilin (soluble in saline solution). Additionally, the binding of phenolic compounds (flavonoids, rutins, and phenolic acids) and tannins with Se molecules can form insoluble compounds, limiting the transfer of this element to the plant-based beverages [30,31].

It is worth noting that the impact of technological steps can influence the final Se content in PBBs. Examples include: cereal, oilseeds and pumpkin seed grinding, the integrity of the raw material components, the proportion between the raw material and beverage, and the solubility of Se in aqueous media. All these factors were taken into consideration during the handling of the PBBs characterized in this study.

3.2. Estimation of selenium bioaccessibility in PBBs of macadamia nuts, cashews, and Brazil nuts

The results obtained from the assessment of Se bioaccessibility in plant-based beverages after *in vitro* gastrointestinal digestion (INFO-GEST) are presented in Table 2. The results were expressed in solubility ($\mu g \ kg^{-1}$) and bioaccessibility (percentage of the soluble fraction relative to the initial amount). For the assessment of Se bioaccessibility estimation, only beverages with Se content above the LOQ of the method (20 $\mu g \ kg^{-1}$) were analyzed; these being the plant-based beverages prepared with macadamia nuts, cashews, and Brazil nuts.

According to Table 2, Brazil nut plant-based beverages, displayed the highest levels of total and bioaccessible Se (3509 and 3350 $\mu g \ kg^{-1}$, respectively), and the highest bioaccessible fraction (96 %), compared to macadamia (86 %) and cashew nut (93 %) beverages; with statistical differences found among the bioaccessible fractions of the studied plant-based beverages (P < 0.05).

In the literature, there is still a scarcity of studies regarding the bioaccessibility of Se in plant-based beverages. In general, plant-based beverages showed high percentages of Se bioaccessibility (86 to 96%), corroborating with Afonso et al. [30]; who reported high percentages of Se bioaccessibility (>90%) in food matrices. These authors still correlated the high bioaccessibility of Se with the presence of soluble proteins (around 80%). Dobrzyńska et al. [22] reported that organic forms of Se are absorbed more efficiently by the body, and Turck et al.

 Table 2

 Bioaccessibility of selenium (Se) in plant-based beverages.

Parameters	Plant-based beve	Plant-based beverages				
	Macadamia	Cashew nut	Brazil nuts			
Se total (μg kg ⁻¹)	35 ± 2^{c}	$44\pm1^{\rm b}$	3509 ± 23^{a}			
Se BF ($\mu g \ kg^{-1}$)	30 ± 1^{c}	41 ± 2^{b}	3350 ± 14^a			
% BF	86 ± 4^{c}	93 ± 2^{ab}	96 ± 1^a			
Se RF (µg kg ⁻¹)	n.d.	n.d.	94.2 ± 0.1^a			
% RF	_	_	2.7 ± 0.1^a			
Recovery%	86 ± 4^c	$93\pm2^{\rm b}$	98.7 ± 1^a			

BF: bioaccessible fraction; RF: residual fraction; n.d.: not detectable (< LOQ = $20~\mu g~kg^{-1}$).

Results are expressed as mean \pm standard deviation (n = 3).

Different letters in the same line indicate significant differences (P < 0.05; ANOVA + Tukey's Test).

[32] reported that 90 % of Se in plant sources occurs in organic form (Se-methionine), facilitating its absorption by the intestine.

Several factors can contribute to the high variability (10–97 %) of Se bioaccessibility percentages, such as: food composition, Se content, interaction between the food matrix and Se; as well as the use of different *in vitro* models to estimate the absorption gastrointestinal of this mineral [33]. Farooq et al. [33] also reported that the high variability of Se is due to the composition of the human intestinal microbiota, as it is capable of altering the chemical forms of Se, providing greater intestinal uptake. Lavu et al. [34] found that, when incubated in the human colon for 48 h, the absorption levels of Se from selenomethionine (SeMet) and selenate decreases to less than 40 % and 70 %, respectively. This fact can be explained by the high concentration of bacterial cells in the intestinal content, capable of absorbing Se as a micronutrient for its various metabolic processes.

Furthermore, variations in Se absorption percentage reductions are attributed to different forms of absorption by the intestinal microbiota. Mombo et al. [35] reported that the intestinal microbiota displays a preference for selenomethionine over selenate. With regard to the *in vitro* digestion method used, comparing the percentages of Se bioaccessibility in our study with other research becomes challenging, since the different *in vitro* digestion models differ in variables; such as pH, units of enzymatic activity, duration of digestion and the fraction considered as bioaccessible [36].

In this context, future studies on Se bioaccessibility percentages in various food matrices, including PBBs, should be monitored. A detailed analysis of the bioaccessible behavior of the different forms of Se (selenomethionine, selenate, selenite, and selenocystine) will expand our understanding of the absorption of this element by the human organism. Furthermore, the various chemical forms of Se could be fortification material for PBBs with low Se content. It is worth noting that the intake of Se in the diet needs to be strictly monitored to meet the RDI of $55~\mu g/day$ (children and adults), as well as to avoid the emergence of adverse effects after an intake greater than $255~and~400~\mu g/day$, as advised by the EFSA [32] and the Institute of Medicine [37], respectively.

3.3. Estimation of Se intake through consumption of macadamia, cashew, and Brazil nut PBBs

In Table 3, the results of the estimated Se intake through the daily consumption of plant-based beverages are presented. For this estimation, the consumption of one cup (200 mL) of plant-based beverages by adults and children was considered, the reference daily intake (RDI) of $55~\mu g$ [8], and the levels of Se (total and bioaccessible) in different beverages studied. Although we analyzed the bioaccessibility

Table 3Results of the daily selenium (Se) intake estimate, considering a consumption of 200 mL of plant-based beverage by children and adults.

Plant-based beverages	Se total (μg kg ⁻¹)	% of RDI	BF of Se (μg kg ⁻¹)	% of RDI
Pumpkin seeds	$19\pm1^{\text{d}}$	$\begin{array}{c} \textbf{7.0} \pm \\ \textbf{0.4}^{\text{d}} \end{array}$	n.d.	-
Cashew nut	44 ± 1^{b}	$16.1~\pm\\0.5^{\rm b}$	41 ± 2^b	15 ± 1^{b}
Brazil nuts	3509 ± 23^a	$\begin{array}{c} 1276 \ \pm \\ 2^a \end{array}$	$3350\pm14^{\text{a}}$	$1218 \pm \\ 5^a$
Macadamia	35 ± 2^c	13 ± 1^{c}	30 ± 1^c	$11.0 \pm 0.4^{\rm c}$
Nuts	$12\pm1^{\rm e}$	$4.3\pm0.5^{\rm e}$	n.d.	_
Sunflower seed	10.3 ± 0.1^{e}	3.7 ± 0.1^{e}	n.d.	-

BF: Bioaccessible fraction; n.d.: not detectable (< LOQ = 20 μ g kg⁻¹). Results are expressed as mean \pm standard deviation (n = 3).

Different letters in the columns indicate significant differences (P < 0.05; ANOVA + Tukey's Test).

RDI (Reference Daily Intake) = $55 \mu g$ [8].

percentage for all laboratory-prepared PBBs (non-commercial samples), only PBBs with Se content above 20 $\mu g\ kg^{-1}$ (LOQ of the method) could be quantified in this study. A recent publication from our research group also revealed that commercial PBBs containing Se levels between 4 and 20 $\mu g\ kg^{-1}$ exhibited bioaccessible extracts with non-quantifiable Se values. In this context, only PBBs formulated with macadamia, cashew, and Brazil nuts presented quantifiable Se in the bioaccessible fraction obtained by INFOGEST protocol, and were considered for RDI estimation for children and adults.

Despite static *in vitro* methods providing high analytical performance, reproducibility and absence of ethical restrictions; it is important to mention that these methods have some limitations, such as the food mechanical breakdown simulation and dynamic variable processes (pH, enzymes and gastric emptying) [11]. In the present study, these aspects were also considered, as well the PBBs macro and micronutrient composition.

The contributions of sunflower seed, walnut, and pumpkin beverages to Se intake was considered low, accounting for 3.7 %, 4.3 %, and 7.0 % of the Se RDI, respectively. For cashew nuts, values of 16.1 % and 15 % were observed when considering the total content and bioaccessible fraction, respectively. These values are slightly higher than those reported by Orlando et al. [12], who estimated between 9 % and 13 % of the RDI of Se from commercial beverage samples of cashew nut.

For adults and children, whose recommended daily intake of Se is 55 μ g/day, as established by the FDA [8], the intake of just 200 mL of the Brazil nut drink substantially contributes to a higher intake of Se, corresponding to 1276 % and 1218 % of the recommended daily value for the total content and bioaccessible fraction, respectively. The EFSA establishes a UL (Tolerable Upper Intake Level) value for Se at 255 μ g/day [32], while the Institute of Medicine recommends a UL of a maximum of 400 μ g/day [37]. Therefore, when considering the total consumption of Se through the ingestion of 200 mL of beverage of Brazil nut, we observed that both adults and children would ingest around 2.75 and 1.75 times the maximum value recommended by EFSA [32] and the Institute of Medicine [37], respectively. This result suggests the need for caution and attention when incorporating this drink into the daily diet of this target audience, especially in vulnerable populations.

Trace elements, such as Se, play a crucial role in normal physiological functions. Se acts as a potent antioxidant, protecting cell membranes against damage from free radicals, and is an essential micronutrient for humans [38]. Furthermore, Se is involved in hormonal metabolism, growth regulation, cellular function, the immune system and reproduction [39]. On the other hand, excess Se in the diet may pose a health risk. Excessive Se intake in the diet may lead to the loss of teeth, hair, and nails, and can also cause disturbances in the nervous system [40]. Evidence suggests a close association between selenium deficiency and various physiological disorders, from chronic inflammatory diseases to cardiac dysfunction, vascular blockage, and cancer [41]. Morgan et al. [42] further reported that excess Se accumulates as selenomethionine (SeMet) in hair and nail proteins, as well as in tissues of the heart, muscles, and skin, affecting their behavior and performance.

In general, the supplementation form of Se, genetic polymorphisms, and behavioral factors can impact the absorption rates of Se in the organism [42]. Considering that Se can be present in the diet from other sources (eggs, fish, and offal, among others [27]), monitoring the consumption of this nutrient is extremely important to prevent adverse effects, and contribute to the proper functioning of the organism.

In this context, this study highlights the need for future research into the impact of Se intake through various diets, including PBBs. In this study, we emphasize consumer awareness of the potential risks associated with excessive Se intake, especially in relation to the consumption of Brazil nut PBB. Furthermore, the implementation of precautionary measures and the promotion of informed choices are essential to ensure health and well-being.

4. Conclusions

For all raw materials studied (cereals, oilseeds and pumpkin seeds) it was possible to quantify the total Se content. The rye (cereals), Brazil nuts (oilseeds), and sunflower seeds (seeds) showed the highest levels of Se (18, 28,148, 278 $\mu g\ kg^{-1}$, respectively).

For PBBs, it was not possible to quantify the Se content in raw materials (cereals: oats and rye; oilseeds: hazelnut and almonds) and bioaccessible fractions (oilseeds: nuts and sunflower seeds; seeds: pumpkin seeds) that had Se content below the LOQ for the bioaccessibility method (< LOQ, < 20.0 $\mu g\ kg^{-1}$). The Brazil nut vegetable beverage (PBB) presented the highest content of total Se and soluble Se (3509 and 3350 $\mu g\ kg^{-1}$, respectively), as well as the highest percentage of bioaccessibility (96 %), when compared with the other samples of cashew nut (93 %) and macadamia nut (86 %).

The daily consumption of just one glass (200 mL) of Brazilian nut plant-based beverage (PBB) may pose a risk of adverse effects on the organism due to the high concentration of Se present in this drink. Therefore, it is imperative to regulate excessive Se intake in the diet, given that this mineral can be found in various food sources.

In this context, this study emphasizes future studies that aim to determine the total content and bioaccessible fraction of Se in other food matrices, including other types of PBBs, as well as research related to the fortification of PBBs with low Se content with different chemical forms of Se (selenate, and selenite, among others) and also studies aimed at the biofortification of vegetable crops with Se during planting.

Ethical statement

This article **does not** include experiments with tissues and/or animals.

This article does not include experiments on humans.

CRediT authorship contribution statement

José Luan da Paixão Teixeira: Writing – review & editing, Writing – original draft, Validation, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. Raquel Fernanda Milani: Writing – review & editing, Writing – original draft, Visualization, Supervision, Methodology. Marcelo Antonio Morgano: Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Investigation.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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