

Heat treatment effect study on bioactive compounds of unconventional food plants

Estudo do efeito do tratamento térmico nos compostos bioativos em PANC

Estudio del efecto del tratamiento térmico sobre compuestos bioactivos en PANC

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Abstract

This study aimed to evaluate the effect of heat treatment (HT) on bioactive and antinutritional compounds of different PANC (*Cnidoscolus aconitifolius* (chaya), *Tallinum paniculatum* (jewels of opar), *Urtica dioica* L. (nettle leaf), *Costus spicatus* (spiked spirallflag ginger), *Portulaca oleracea* (purslane), *Mentha suaveolens* (pineapple mint) and *Sonchus oleraceus* (sow thistle). The concentrations of the bioactive compounds such as total phenolics (TF) and flavonoids, antinutritionals (tannins and oxalates) and antioxidant activity (AA) were carried out according to the Analytical Standards of the Adolfo Lutz Institute. The compounds contents (mg/100g) in PANC before the HT ranged as follows: bioactive compounds (TF from 2.04 ± 0.04 to 37.40 ± 0.46 , flavonoids from 145.96 ± 0.61 to 410.73 ± 1.93), anti-nutritional compounds (tannins from 4.47 ± 0.46 to 211.74 ± 2.96 and oxalates from 2.61 ± 0.13 to 4.41 ± 0.22), AA (IC50) from 2.13 ± 0.05 to 7.05 ± 1.36 . The HT applied to the samples provided an increase of compounds concentration percentage in most PANC, which ranged from: 38.13 to 99.84% (TF); 1.2 to 37% (flavonoids); 21.21 to 116.55% (tannins); 45.92 to 77.63 (oxalates), as well as in AA, which ranged from 14.5 to 67%. On the other hand, there was a reduction of compounds concentration percentage in other PANC from: 45 to 73% (TF); 8 to 29% (flavonoids); 15 to 74% (tannins); 5.36 to 27.44% (oxalates) and 44.49 to 79.84% (AA). It is concluded that, in general, the applied HT favored an increase in the concentration of bioactive compounds and AA in PANC, the same not happening for antinutritional activity. It is generally concluded that HT favored an increase of bioactive compounds and AA concentration in PANC, however the same did not occur for antinutritional activity.

Keywords: Unconventional food plants; Bioactive compounds; Heat treatment.

Resumo

Este trabalho objetivou avaliar o efeito do tratamento térmico (TT) nos compostos bioativos e antinutricionais em diferentes PANC (Beldroega, Bredo, Chaya, Cana de macaco, Hortelã com borda, Língua de vaca, Serralha e a Urtiga mansa). As concentrações dos compostos bioativos - fenólicos totais (FT) e flavonoides; dos antinutricionais (taninos e oxalatos) e a atividade antioxidante (AA) foram realizadas segundo as Normas Analíticas do Instituto Adolfo Lutz. Os teores (mg/100g) dos compostos nas PANC antes do TT variaram da seguinte forma: compostos bioativos - FT de $2,04\pm 0,04$ a $37,40\pm 0,46$, os flavonoides de $145,96\pm 0,61$ a $410,73\pm 1,93$; os compostos antinutricionais - taninos de $4,47\pm 0,46$ a $211,74\pm 2,96$, e oxalatos de $2,61\pm 0,13$ a $4,41\pm 0,22$; a AA (IC50) variou de $2,13\pm 0,05$ a $7,05\pm 1,36$. O TT aplicado nas amostras proporcionou aumento no percentual da concentração

dos compostos na maioria das PANC que variou de: 38,13 a 99,84% (FT); 1,2 a 37% (flavonoides); 21,21 a 116,55% (taninos); 45,92 a 77,63 (oxalatos), assim como na AA, que variou de 14,5 a 67%. Por outro lado, houve uma variação no percentual da redução nas outras PANC de: 45 a 73% (FT); 8 a 29% (flavonoides); 15 a 74 % (taninos); 5,36 a 27,44% (oxalatos) e de 44,49 a 79,84% (AA). Conclui-se que, de maneira geral, o TT aplicado favoreceu o aumento da concentração dos compostos bioativos e da AA nas PANC, o mesmo não ocorrendo para os fatores antinutricionais.

Palavras-chave: PANC; Compostos bioativos; Tratamento térmico.

Resumen

Este trabajo tuvo como objetivo evaluar el efecto del tratamiento térmico (TT) sobre compuestos bioactivos y antinutricionales en diferentes PANC (Verdolaga, Bredo, Chaya, Caña de mono, Menta con borde, Lengua de vaca, Serralha y Ortiga blanda). Las concentraciones de compuestos bioactivos: fenólico total (FT) y flavonoides; los compuestos antinutricionales (taninos y oxalatos) y actividad antioxidante (AA) realizaron de acuerdo con los Estándares Analíticos del Instituto Adolfo Lutz. El contenido (mg/100g) de los compuestos en los PANC antes de TT varió de la siguiente manera: compuestos bioactivos - FT de $2,04 \pm 0,04$ a $37,40 \pm 0,46$, flavonoides de $145,96 \pm 0,61$ a $410,73 \pm 1,93$; compuestos antinutricionales: taninos de $4,47 \pm 0,46$ a $211,74 \pm 2,96$ y oxalatos de $2,61 \pm 0,13$ a $4,41 \pm 0,22$; el AA (IC50) osciló entre $2,13 \pm 0,05$ y $7,05 \pm 1,36$. El TT aplicado a las muestras proporcionó un aumento en el porcentaje de concentración de compuestos en la mayoría de PANC, que osciló entre: 38,13 a 99,84% (FT); 1,2 a 37% (flavonoides); 21,21 a 116,55% (taninos); 45,92 a 77,63% (oxalatos), así como en AA, que varió de 14,5 a 67%. Por otro lado, hubo variación en el porcentaje de reducción en el resto de PANC de: 45 a 73% (FT); 8 a 29% (flavonoides); 15 a 74% (taninos); 5,36 a 27,44% (oxalatos) y 44,49 a 79,84% (AA). Se concluye que, en general, el TT aplicado favoreció un aumento en la concentración de compuestos bioactivos y AA en PANC, no sucediendo lo mismo con los factores antinutricionales.

Palabras clave: PANC; Compuestos bioactivos; Tratamiento térmico.

1. Introduction

Food is vital for human survival. In this way, the valorization of different food sources, especially vegetables, is extremely important. Vegetable diversity contributes to food

diversity, being source of vitamins, minerals, various sugars, fibers and bioactive compounds such as phenolics, among other essential nutrients for the maintenance of health and life. Thus, valuing plant species that are close to homes and properties is a good food alternative (Polesi, 2016).

Brazil is the country with the greatest biodiversity on the planet, which gives it a wide variety of biomes, and a fauna and flora of countless riches (Brasil, 2019). The low number of researches and studies regarding the diversity of edible plant species – with regard to cultivation and dissemination, biological and nutritional characteristics, forms of processing, among others – promotes neglect and the loss of all present food potential at PANC (Brack, 2016; Kinupp & Lorenzi, 2014). According to Kinupp (2018), the term PANC (Unconventional Food Plants) embraces the concept of plants that have one or more edible parts, whether they are spontaneous or cultivated, native or exotic, which are not part of the daily diet of the population, as well as not fully exploiting part of conventionally edible plants.

These vegetables also have characteristics that give them antioxidant, anti-inflammatory and therapeutic properties, in addition to having nutritional value as sources of vitamins and minerals. Thus, PANC consumption must be carried out, respecting their characteristics and ways of preparation, so that such properties are obtained safely. Due to the little knowledge of the physical-chemical characteristics of most PANC, it is suggested that further studies be conducted with regard to PANC nutritional and antinutritional substances (Paschoal & Souza, 2015).

Some plant species of the *Amaranthus* genus (*A. viridis*, *A. retroflexus*, *A. spinosus*) have been used in human and animal nutrition and, in Brazil, are known as slender amaranth, caruru or amarantos (Kinupp & Lorenzi, 2014; Silva *et al.*, 2016). These species are good sources of calcium, magnesium, nitrogen, iron, phosphorus, among other nutrients (Brasil, 2013; Souza *et al.*, 1999). Another PANC, known as purslane (*Portulaca oleracea*) is classified as a food of relevant nutritional value, since it has bioactive compounds with antioxidant activity and is rich in omega 3, thus contributing to the protection of some pathologies of the system central nervous system, such as Parkinson's disease (Liu *et al.*, 2000; Moraes & Colla, 2018).

Nettle leaf (*Urtica dioica* L.) is one of the most popular plants in several countries. Its leaves and buds have polysaccharides, proteins, essential fatty acids, minerals and vitamin C and low amounts of saturated fat (Jan *et al.*, 2016). *Mentha Suaveolens* known as pineapple mint has several antioxidant compounds, and 20 different compounds present in the essential

oil of this PANC have already been identified (Bouyahya *et al.*, 2019). According to Bouyahya *et al.* (2017), this vegetable has antidiarrheal, antihemorrhoidal and analgesic effects, and is used by the Moroccan population to treat pathologies of the digestive system, allergies and diabetes.

Antinutrients are present in a variety of plant-based foods that, when ingested, reduce the bioavailability of their nutrients and interfere with their digestibility, absorption or use. Oxalate, for example, can precipitate calcium and form insoluble crystals, increasing the risk of kidney stones. Tannins (polyphenols) can precipitate proteins, carbohydrates and minerals, triggering the decrease in nutritional value (Benevides *et al.*, 2011).

Nitrates and nitrites, phytates, cyanogenic glycosides and protease inhibitors are also considered antinutritional substances.

Bioactive compounds participate in the metabolism of the human body and, consequently, contribute to biological health-promoting activities, such as antioxidant, anti-inflammatory and hypocholesterolemic activity (Kris-Etherton *et al.*, 2002; Pinto *et al.*, 2008). Phenolic compounds (phenolic acids, flavonoids, anthocyanins, tannins, etc.), phytosterols, spilantol and terpenes (carotenoids, saponins, etc.) are among the most studied bioactive compounds worldwide for human health (Dubey *et al.*, 2013; Gerardi *et al.*, 2016).

Due to the great genetic variety, phenolic compounds can vary among leafy vegetables and, therefore, these vegetables should be included in the diet as potential suppliers of natural antioxidant substances (Khanam *et al.*, 2012; Singh *et al.*, 2018).

The heat treatment applied to some vegetables before consumption can interfere with the concentration of nutritional and antinutritional substances in them, thus interfering, positively or negatively, in the bioavailability and digestibility of some nutrients (Cardoso *et al.*, 2009).

Thus, aiming to know the nutritional and anti-nutritional characteristics of PANC, the present work analyzed the bioactive and anti-nutritional compounds, as well as the effect of thermal processing on the concentration of these substances in different PANC. In this way, it can contribute to the enhancement of plant biodiversity and the promotion of cultural rescue in relation to its consumption.

2. Methodology

This is an explanatory experimental study, carried out in the UNEB bromatology laboratory. The nature of the research is basic and its approach is qualitative-quantitative, whose method of procedure is experimental (Pereira *et al.*, 2018).

The PANC samples of purslane (*Portulaca oleracea L.*), slender amaranth (*Amaranthus viridis L.*), chaya (*Cnidoscolus aconitifolius*), spiked spiral-flag ginger (*Costus spicatus*), pineapple mint (*Mentha suaveolens*), jewels of opar (*Tallinum paniculatum*), sow thistle (*Sonchus Oleraceos*) and Nettle leaf (*Urtica dioica L*) were obtained from the green areas of the Biology Institute of the Federal University of Bahia (UFBA) and from vegetable gardens in the Metropolitan Region of the Municipality of Salvador-BA. Then the samples were sent to the Food Technology and Bromatology Laboratories of the Department of Life Sciences (DCV) of the State University of Bahia (UNEB), Campus I in Salvador-BA, where they were submitted to analysis. All PANC were cleaned with sodium hypochlorite (1% active chlorine) for 15 minutes, followed by rinsing in drinking water. After cleaning, the samples were removed from the stems (spiked spiralfalg ginger, pineapple mint, chaya, nettle leaf and slender amaranth) with the exception of jewels of opar and purslane. Subsequently, all samples were manually chopped, and part of this material was subjected to thermal treatment – HT (bleaching for 3 minutes) in the proportion of 160 g/40mL of water. Then, the two samples (with and without heat treatment) were dehydrated in an oven with forced air circulation (model 400/D) at 40°C until the humidity reached around 9%. The dehydrated samples were ground in a hammer mill (TE650 Wiley type mill) and filled in polypropylene jars, hermetically closed until the extract was obtained.

PANC extracts were obtained from the dry sample (0.8 g), previously sprayed and added with 50 mL of methanol. Then, the mixture was subjected to stirring for 15 minutes at room temperature and, subsequently, centrifuged at 3000 rpm/5 minutes. The supernatants were placed in polyethylene bottles and refrigerated until the time of analysis.

Total phenolics (TF), flavonoids, tannins, oxalates concentrations and antioxidant activity (AA) were performed, according to the Analytical Standards of the Adolfo Lutz Institute (2008), in which the determination of TF was performed by the Folin-Ciocalteu. In this case, gallic acid was also used as the standard and $\lambda = 750\text{nm}$. Tannins concentration was determined by the spectrophotometric method, using tannic acid as the standard and $\lambda = 760\text{ nm}$. The data were expressed in mg equivalent of gallic acid/100g and mg equivalent of tannic acid/100g, respectively.

For an evaluation of antioxidant activity ($\lambda = 517 \text{ nm}$), the DPPH method (radical 2,2-diphenyl-1-picrylhydrazyl) was used. As absorbencies, they were placed in the spectrophotometer model “X” (Shimadzu Corporation, São Paulo, Brazil). According to Moon & Shibamoto (2009), the DPPH • method is one of the most used in research to evaluate the antioxidant activities of substances. The principle of this method is based on the generation of a radical by DPPH • and on the ability of the sample to eliminate or neutralize this radical. This reaction is monitored through a UV / visible spectrophotometer (Arnao, 2000). The results of this method are expressed in the IC50, or are defined as an amount of the antioxidant substances present in the reduced sample to decrease or reduce the initial concentration of the radical DPPH • by 50% (Brand-Williams *et al.*, 1995; Chen *et al.*, 2013). Therefore, the lower the sample's IC50 value, the better its antioxidant capacity. All samples were analyzed in triplicate and the results expressed by the mean and standard deviation.

3. Results and Discussion

The bioactive compounds contents (mg/100g) in PANC before HT (without bleaching) ranged from: TF (2.04 ± 0.04 to 37.40 ± 0.46) and Flavonoids (145.96 ± 0.61 to 410.73 ± 1.93), while the Antioxidant Activity (IC50) was from 2.13 ± 0.05 to 7.05 ± 1.36 . The PANC with the highest levels of TF, Flavonoids and Antioxidant activity (AA) were sow thistle, chaya and sow thistle, respectively. Pineapple mint presented the lowest values for TF, Flavonoids and purslane for AA.

Table 1 shows the results of the bioactive compounds concentrations and antioxidant activity of PANC before and after HT on dry base.

Table 1 – Effect of Heat Treatment on Bioactive Compounds and Antioxidant Activity of PANC on a Dry Base.

PANC	HT	TF (mg/100)	Flavonoids (mg/100g)	IC50
Jewels of opar (<i>Tallinum paniculatum</i>)	WW	11.07±0.30	229.32±3.95	4.71±0.58
	B	4.59±0.13	332.1±1.16	1.55±0.38
Purslane (<i>Portulaca oleracea L.</i>)	WW	-	156.47±0.50	7.05±1.36
	B	-	247.70±0.31	0.69±0.23
Spiked Spiralflag Ginger (<i>Costus spicatus</i>)	WW	3.75±0.08	264.90±1.55	6.92±0.38
	B	5.18±0.01	423.05±2.73	2.98±0.21
Chaya (<i>Cnidoscolus aconitifolius</i>)	WW	29.74±0.60	410.73±1.93	5.24±0.15
	B	8.05±1.26	415.91±1.18	4.48±0.28
Pineapple Mint (<i>Mentha suaveolens</i>)	WW	2.04±0.04	145.96±0.61	4.81±0.06
	B	3.99±0.02	193.52±0.46	1.83±0.01
Slender amaranth (<i>Amarantus viridis</i>)	WW	6.28±0.09	305.58±2.08	6.28±0.09
	B	12.55±0.72	281.56±1.20	12.55±0.72
Nettle leaf (<i>Urtica dioica L.</i>)	WW	6.63±0.22	164.23±0.50	6.63±0.22
	B	9.68±0.23	210.67±2.03	9.58±0.77
Sow thistle (<i>Sonchus oleraceus</i>)	WW	37.40±0.46	300.70±0.96	2.13± 0.05
	B	20.87±0.50	214.14±1.19	4.31±0.23

WW - Without Whitening; B – Bleached.

Source: Own authorship, (2020).

Table 1 shows that the HT increased the AA of some PANC (jewels of opar, purslane, spiked spiral-flag ginger, chaya and pineapple mint), and this increase was greater (90.21%) for purslane, followed by of jewels of opar (67.09%) and pineapple mint (61.95%), indicating that HT improved the AA of these PANC. On the other hand, HT reduced the AA of other PANC (slender amaranth, nettle leaf and sow thistle), varying from 44.49 to 79.84%. This variation is possibly associated with the different species of PANC evaluated and, consequently, their physiological structure and chemical composition, as well as the concentration and interaction of antioxidant compounds with other substances, among others.

Considering PANC *in natura*, that is, before HT, sow thistle was the one with the highest AA (IC50 2.13±0.05) and purslane the lowest (IC50 7.05±1.36). The antioxidant activity (AA) of foods may be related to substances that slow down the rate of oxidation, through one or more different mechanisms, such as inhibition of free radicals and metal complexation (Pietta, 2000). Thus, the great interest and importance of the consumption of food sources of these anti-nutritional compounds stands out.

The vegetables cultivation methods can also influence their composition. Studies of vegetables grown in organic and conventional systems were carried out by Machado (2012) - cauliflower (*Brassica oleracea*) and by Arbos *et al.*, (2010) - common chicory, lettuce and arugula. The authors mention that AA was higher in vegetables submitted to organic cultivation when compared to the conventional system.

According to Melo *et al.* (2009), the antioxidant action of a food is strongly influenced by high temperatures, as this process can increase or reduce the antioxidant action. This reduction can occur due to the destruction of bioactive compounds or their conversion into substances with pro-oxidant activity after cooking. On the other hand, the action of HT in the rupture of cell membranes in vegetables can facilitate the extraction of phenolic compounds and increase their concentration (Ismail *et al.*, 2004).

Heat treatments that use less water or have shorter contact time at high temperatures can preserve or even increase the concentration of chemical compounds in vegetables by breaking and contacting cell tissue with heat (slower heating), causing greater extraction chemical compounds of the analyzed material (Reis *et al.*, 2015). In this context, some studies confirm that cooking can increase or decrease the availability of chemical compounds in vegetables, depending on the method and preparation time in which the samples are submitted (Lima *et al.*, 2017; Preti *et al.*, 2017; Reis *et al.*, 2015).

The HT applied in the samples in this research favored the increase of the TF content in the following PANC: Spiked spiral-flag ginger (38.13%), Pineapple mint (95.58%), Slender amaranth (99.84%), Nettle leaf (46, 00%).

Zapata-Vahos *et al.* (2015) evaluated the concentration of phenolic compounds in teas and found that the TF content in aqueous extracts increased significantly between 40 and 90°C, with values ranging between 39.28±1.45 and 85, 62±4.9 mg GAE/g in green tea and 32.66±1.49 and 189.83±11.9 mg GAE/g in black tea. Probably, the higher the water temperature, the greater the amount of extraction of TF compounds. This is due to the effect of temperature on the solubility and diffusion rate of compounds in the solution (Atkins, 2001). Sultana *et al.*, (2008) also found an increase in the content of phenolic compounds after steaming in carrots, peas and spinach.

However, a reduction in TF was observed in PANC samples: jewels of opar (59%), chaya (73%) and sow thistle (45%), with a greater decrease in chaya. As there is a very wide variety of phenolic compounds with different chemical characteristics, this reduction in TF in this study can be attributed to the polarity of these compounds, which were possibly leached in the bleaching water.

-The concentration (mg/100g) of flavonoids in PANC *in natura* (WW) ranged from 145.96±0.61 (pineapple mint) to 410.73±1.93 (chaya).

Among vegetable products, flavonoids are one of the most important phenolic groups and can be found in fruits, vegetables, seeds, flowers and bark, and several of these foods are part of the human diet. More than 4 thousand flavonoid compounds have already been identified (Pereira, 2010).

According to Gobbo-Neto & Lopes (2007) flavonoids are produced in response to environmental conditions and, thus, the differences in flavonoid contents of different vegetables are justified. Its concentration can increase in response to environmental stress, which can be caused by disease, altitude, air pollution, nutrients, climate and ultraviolet radiation (UV) (Gobbo-Neto & Lopes, 2007; Macedo *et al.*, 2013).

Among TF compounds, flavonoids are associated with biological activities, highlighting the antioxidant, anti-inflammatory, anti-tumor, anti-allergic, antiviral action, among others. According to Pereira & Cardoso (2012) these compounds donate hydrogen atoms and, thus, protect tissues from reactions caused by free radicals and lipid peroxidation.

The heat treatment applied, in general, also influenced the increase in the flavonoid content in most PANC: jewels of opar (31%), purslane (37%), spiked spiral-flag ginger (37%), chaya (1.2%), pineapple mint (25%) and nettle leaf (22%). The applied HT used as little water as possible and, thus, it may be that the heating has favored better extraction of the phenolic compounds, avoiding that they were leached in the cooking water. The reduction in the levels of flavonoids occurred only in the samples of slender amaranth (8%) and sow thistle (29%).

Ioku *et al.* (2001) evaluated cooking methods to measure flavonoids in onion and observed that, after submitting the onion under microwave heat, during 1 minute there was a 1.5-fold increase in the total quercetin content, on it, showing that the compounds were extracted more easily, however, when subjected to cooking in water, there was a significant loss of the compounds, justifying that the flavonoids migrated to the water used in cooking.

According to Savi *et al.* (2017), possibly, the presence of fibers in vegetables makes it difficult to extract flavonoids when they are heated. Heating may cause the flavonoids to be trapped in the fiber. Fibers are complex carbohydrates and can be soluble and insoluble. In the body, one of the mechanisms that can explain the action of soluble fibers, would be that they absorb water and form a gel in the intestinal lumen, acting in reducing the absorption of carbohydrates and decreasing the reabsorption of bile acids (Ma & Um, 2016). Thus, as with

carbohydrates and other nutrients, fiber makes flavonoids less available for absorption (Savi *et al.*, 2017).

Table 2 shows the antinutritional compounds in PANC leaves before and after HT.

Table 2 - Antinutritional compounds in PANC leaves before and after HT on Dry Base.

PANC	HT	Tannins (mg/100g)	Oxalates mg/100)
Jewels of opar (JO) (<i>Tallinum paniculatum</i>)	WW B	141.61±3.43 85.76±1.32	2.64±0.30 5.51±0.00
Purslane (P) (<i>Slender amaranth</i>)	WW B	- -	3.31±0.06 4.90±0.26
Spiked Spiralfalg Ginger (<i>Costus spicatus</i>)	WW B	211.74±2.96 179.34±0.96	3.89±0.30 6.91±0.23
Chaya (<i>Cnidoscopus aconitifolius</i>)	WW B	95.00±1.13 24.51±0.69	4.41±0.22 3.20±0.00
Pineapple Mint (PM) (<i>Mentha suaveolens</i>)	WW B	37.24±1.03 45.14±0.90	3.68±0.25 5.37±0.29
Slender amaranth (SA) (<i>Amarantus viridis</i>)	WW B	94.04±0.54 127.59±0.93	2.61±0.13 2.47±0.39
Nettle leaf (NL) (<i>Urtica dioica L.</i>)	WW B	52.07±0.50 71.89±1.92	3.59±0.26 3.06±0.13
Sow thistle (ST) (<i>Sonchus oleraceus</i>)	WW B	4.47±0.46 9.68±0.53	3.39±0.23 2.68±0.27

WW - Without Whitening; B – Bleached.

Source: Own authorship, (2020).

Table 2 shows that HT promoted an increase in the tannin content of the following PANC samples: pineapple mint (21.21%), slender amaranth (33.55%), nettle leaf (38.06%) and sow thistle (11.55%). On the other hand, there was a reduction in the samples of: jewels of opar (39.44%), spiked spiralfalg ginger (15.30%) and chaya (74%). The reduction in tannin levels can be attributed to the degradation or interaction of these with other compounds present in the vegetable or may have been due to its solubility in the bleaching water.

The levels (mg/100g) of tannins in the *in natura* samples, that is, without HT varied from 4.47±0.46 to 211.74±2.96. Sow thistle and spiked spiralfalg ginger had the lowest and highest levels, respectively.

Some phenolic compounds in plants do not show toxicity in normal amounts and conditions. However, polymeric phenols such as tannins (they have the ability to complex and precipitate proteins from aqueous solutions) can act as toxic substances. Tannins are phenolic compounds that have high molecular weight (500-3000 Da) and come from secondary plant

metabolism (Salunkhe *et al.*, 1990). They are also able to bind to amino acids and polysaccharides, causing a reduction in the nutritional value of foods (Guzmán-Maldonado *et al.*, 2000). This tannin binding capacity is possible due to its properties, such as low solubility, large molecules and mobility, providing hydrophobic bonds, hydrogen bonds, ionic and covalent (Battestin *et al.*, 2004).

In a way, the reduction in the tannin content in foods is of great interest, as this anti-nutritional factor has the ability to drastically reduce the digestibility of proteins, in the proportion of 5:1, tannins/protein, as well as precipitate proteins by the action of tannins. (Pino & Lajol, 2003).

Santos (2005) investigated the reduction in the levels of antinutritional factors (nitrates, tannins and oxalates) in broccoli, cauliflower and kale leaves with increased cooking time and demonstrated that HT reduced the concentration of these substances, making these vegetables safer for consumption.

The content (mg/100g) of oxalates in the *in natura* samples (without HT) ranged from 2.61 ± 0.13 (slender amaranth) to 4.41 ± 0.22 (chaya). Oxalic acid is often found in vegetables and has a low toxicity threshold, and the minimum dose considered lethal for adults is around 5g (Benevides *et al.*, 2013). On the other hand, the high amount of oxalate in the urine increases the risk of the formation of calcium oxalate stones in the kidneys, since calcium oxalate is poorly soluble in the urine, and can also cause irritations in the intestinal mucosa (Benevides *et al.*, 2013; Cobayashi, 2004).

~~Differences in oxalate~~ values in foods may be due to different analytical methods used and/or biological variation from various sources, including cultivar, time of harvest and cultivation conditions.

Rocha (2009) determined the levels of oxalic acid in samples of spinach under different acidity conditions and found a variation between 299 mg/g to 251 mg/g depending on the pH of the extracting solution.

The oxalate bioavailability in food is affected by the forms of oxalate salt, methods of processing and cooking food, meal composition, among others (MASSEY, 2007). Benevides *et al.*, (2013) cite that the acceptability of a food in the human diet depends not only on its sanitary, sensory, nutritional quality, cooking and hydration characteristics, but on minimal amounts of anti-nutritional factors, so as not to interfere with bioavailability of its nutrients.

Evaluating the effect of HT on oxalate concentrations, a reduction was observed in the following PANC: chaya (27.44%), slender amaranth (5.36%), nettle leaf (14.96%) and sow

thistle (21.00%) and increase in the others, ranging from 45.92 to 77.63% with emphasis on jewels of opar.

The location of oxalate in the vegetable may influence its variation after processing. According to Siener *et al.* (2005), oxalate is distributed throughout the plant tissue, however its content is higher in leaves and stems.

Regarding the effect of HT, Santos (2005) mentions that bleaching has been shown to be efficient in reducing oxalic acid in vegetable leaves. Chai & Liebman (2005) also evaluated the effect of cooking methods on vegetables on the oxalate concentration and mention that boiling markedly reduced the soluble oxalate content (30-87%) and was more effective than steam (5-53 %). According to the authors, the losses of insoluble oxalate during cooking varied widely, ranging from 0 to 74%. As soluble sources of oxalate appear to be better absorbed than insoluble sources, employing cooking methods that significantly reduce soluble oxalate can be an effective strategy to decrease oxaluria in individuals predisposed to the development of kidney stones.

Yadav & Sehgal (2003) also observed losses of this antinutrient in spinach (*S. oleracea*) after 5 minutes of bleaching, equivalent to a 36% reduction, where before bleaching this vegetable had 12.59 ± 0.09 mg/g after treatment thermal 8.01 ± 0.13 mg/g. It is suggested that HT, as bleaching, would be efficient in reducing anti-nutritional factors in vegetables.

According to Lopes *et al.* (2009) the ideal value of oxalic acid in the diet would be 50 to 200 mg/day. In this context, the values found in this research are well below this limit, indicating that the PANC analyzed have little amount of oxalates, being an alternative for including these leaves in human food.

4. Conclusion

It is concluded that PANC has very divergent concentrations of bioactive compounds and AA. In general, the applied HT favored an increase in the concentration of bioactive compounds and AA in the PANC, which was not the case for antinutritional factors. Thus, due to the great difference in physical and chemical characteristics between PANC species, it is suggested that more research be carried out to determine the nutritional value, bioactive compounds, antioxidant properties and anti-nutritional factors, as well as the relationships between the compounds present in these plants. unconventional food. It is noteworthy that the

use PANC in human nutrition is an important strategy to expand the supply of nutrients, diversify the diet and contribute to the food security of the population in the world.

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