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Effect of boiling on bioactive compounds and radical scavenging activity of anthocyanin-rich vegetables: Red amaranth and red skin potato

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Abstract

This study was conducted to determine the optimum boiling time resulting in maximum retention of the bioactive compounds and radical scavenging activity. For this analysis, two most commonly consumed anthocyanin-rich vegetables, red amaranth (*Amaranthus tricolor*) and red skin potato (*Solanum tuberosum*) were exposed to boiling for 0, 1, 5, 10, and 20 minutes. Then the effects of boiling times on physicochemical, bioactive compounds, and antioxidant activity by DPPH radical scavenging ability were analyzed. In physicochemical analysis, ash content was decreased significantly, and the pH value increased significantly as the boiling time increased (p<0.05). The % of WSI was increased significantly as the boiling time for red amaranth as well as red skin potato (p<0.05) increased. The total polyphenol content (TPC), total anthocyanin content (TAC), total carotenoid content (TCC), and radical scavenging activity (RSA) of red amaranth and red skin potatoes were gradually reduced with the increasing of boiling time. The first - order kinetic model showed the good fit for the loss of total phenolic, carotenoids, anthocyanin, and DPPH radical scavenging activity of red amaranth and red skin potatoes with 0.86 - 0.98 coefficient of determination (R²). The findings could encourage both the household boiling and the food industry to recommend specific boiling time to maintain vegetables' antioxidant properties.

Keywords: Antioxidant; D-value; Rate of loss; Red amaranth; Red skin potato

1. Introduction

"Eating more vegetables" is an ancient and reasonable advice for a healthy diet. Vegetables play an important role in preventing the development of cardiovascular diseases, diseases associated with aging, obesity, cancers and improving memory of humans [1]. The cancer and other action to prevent disease are supposedly due to the fact that vegetables contain not only abundant nutritional components, but also a large amount of antioxidants. In addition, vegetables are very low in calories and are usually consumed in fresh condition, as well as after processing and cooking. Most vegetables cooked prior to consumption, including amaranth and red skin potato. Cooking adds pleasant sensory characteristics to the final product, as well as being more digestible and microbiologically safer to eat. However, depending on the process methods, vegetable species and shapes, the effect may be either positive or negative [2]. Culinary processes induce significant changes in foods such as water loss, changes in the total fat content and fatty acid profile, degradation of thermo - labile compounds, and formation of other compounds due to heat - induced chemical reactions [3]. Phenolic compounds can also be affected by thermal processes and, consequently the antioxidant capacity of consumed vegetables too [4]. In recent years, increasing consumers' awareness towards bioactive components and

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their potential health benefits, has forced them to search for vegetables which contain more functional bioactive compounds.

Most phenolic compounds are well known to be water - soluble and usually lost during boiling. The possible reason for this is easier release of phenolic from soft matrix and/or inactivation of polyphenol oxidase, which may be responsible for loss of phenolic in raw plants [5]. It is necessary to consider the losses of these nutrients while developing new products and/or processes. Knowledge of kinetics including reaction order and rate constant, especially during thermal processing, is very vital to predict the losses / gain. There are still limited data on the effects of boiling on nutritional, antioxidant properties and kinetic model of boiled vegetables. A more integrated analysis is needed to obtain insight into the effects of boiling on bioactive phytochemicals and antioxidant capacity of commonly consumed vegetables. The aim of this study is therefore to investigate the effects of water boiling time on physicochemical, bioactive compounds and antioxidant activity of red amaranth and red skin potato.

2. Material and methods

2.1. Sample

Red amaranth (*Amaranthus tricolor L.*) and red skin potato (*Solanum tuberosum*) were obtained from the local market (Sylhet, Bangladesh). Within 24 hours the samples were prepared in the common manner (e.g., hard stems and blemishes removed) and then washed, wiped, cut into almost equal small pieces, and mixed well [6].

2.2. Boiling method

The boiling of vegetable was performed using the method described by Kao et al. [6]. Two hundred milliliters of water was brought to boil in a 500 mL beaker. The beaker was covered to prevent water loss due to evaporation. Ten grams each of red amaranth and red skin potato were boiled separately for 0, 1, 5, 10, and 20 minutes respectively. The samples boiled for 0 minute were taken as control. After boiling the samples were drained and cooled rapidly in cold water. Then the samples were vacuum packed in polyethylene bags and store at -20 °C.

2.3. Physicochemical characterization

Moisture, ash, pH, and water solubility index (WSI) were determined in all samples. To evaluate the moisture content, a gravimetric assay was conducted [7]. Samples (~5 g) were dried in an oven at 105 °C, followed by regular weighing up to a constant weight. Results were expressed as percentage (%). The mineral content was assessed by incineration at 550 °C and results were expressed as percentage (%) [7]. A pH meter (Model No. PH500) was used to measure the pH of the samples. The WSI were determined according to the Yagci and Gogus [8]. A total of 10 g of sample was boiled into 200 mL of distilled water. This solution was transferred to experimental tubes, centrifuged for 5 min at 3000 rpm, and allowed for 30 minutes to settle completely. An aliquot of 25 mL of the supernatant was transferred to pre-weighed petri dishes and immediately oven-dried at 105 °C for 5 h. The water solubility index (WSI) was calculated as the weight difference by following the equation (1) below-

WSI % = (Weight of dissolved solid in supernatant/weight of dry solids) x 100 (1)

2.4. Determination of bioactive compounds

2.4.1. Preparation of extracts

Ten grams of samples were taken in a centrifuge tube with 40 mL 60 % ethanol. The mixture was homogenized for 1 minute under nitrogen flow. Then centrifuged at 1000 rpm for 5 minutes at room temperature and the supernatant was collected. The precipitate was re-extracted by adding 20 mL 60% ethanol, homogenized for further 1 minute, and centrifuged at 1000 rpm for 5 minutes. This ethanol extraction was repeated four times, and the resulting supernatants were combined and dried under vacuum, at a temperature below 30 °C. The residue was then re-dissolved by ultrasonic agitation to a final volume of 20 mL in 60% ethanol, and extracts were stored at -18 °C until analysis [5].

2.4.2. Determination of total phenolic content (TPC)

The total phenolic content was measured following the method described by Lemos et al. [9] with some modifications. In short, a sample solution aliquot (500 μ L) was mixed with 500 μ L of distilled water and 1 mL of Folin – Ciocalteu reagent, maintaining the mixture for 5 minutes. After adding 5 mL of sodium carbonate (5 %), the volume with distilled water was adjusted to 10 mL. At room temperature, the reaction mixture was incubated for 1 h. The absorbance was measured at 765 nm with a spectrophotometer (UV-2550, Shimadzu, Japan). Quantification was based on the gallic acid

standard curve (y = 0.0004x+0.0013, $R^2 = 0.998$). The results were expressed as micrograms gallic acid equivalent per gram of fresh sample weight.

2.4.3. Determination of total anthocyanin content (TAC)

The total anthocyanin content was determined by pH differential method following the procedure reported by Lemos et al. [9] with some modifications. In short, 1000 g of boiled sample in 20 mL of 95 % ethanol was re-suspended: 1 M HCl (3:1), shaken in the dark for 24 hours, filtered and evaporated. The volume of the solution was then adjusted to 25 mL with 0.01% HCl. The absorption of each sample was measured against a blank at both pH < 1.0 and pH 4.5 using a spectrophotometer at 700 nm and 520 nm. The total contents of anthocyanin were calculated using Eq. (2) and was expressed as mg cyanindin-3- glucoside/100 g fresh weight , where V was the diluent volume, A was the absorbance (calculated from Eq. (3), M was the molecular weight of a reference pigment (cyanidin-3-glucoside)-449.2 g/mol, and ϵ_1 is the molar absorptivity (29,600). Results were expressed as milligrams of cyanidine-3-glucoside equivalent (CGE) 100⁻¹ g fresh weight.

$$A = (A_{520} - A_{700}) pH_1 - (A_{520} - A_{700}) pH_{4.5}$$
(2)
C (mg/g) = V x n x M/ ϵ_1 x m (3)

2.4.4. Determination of total carotenoids content (TCC)

Total carotenoids content was determined by spectrophotometric method according to Vinha et al. [10]. Before quantification, each vegetable (raw and boiled) was submitted for previous extraction. Five grams of homogenized sample were added to 50 mL ethanol (60 %), wrapped in aluminum foil and shaken at room temperature for 30 min. After filtration, 3 mL of supernatant was collected, and absorbance was measured at 445 nm. The total carotenoid content (mg/100 g of fresh sample) was determined according to the following equation (4):

Total carotenoids (mg/100g) =
$$(A \times y \times 10^{6})/(A1\% \text{ cm} \times 1000 \times \text{w})$$
 (4)

Where, A represents the absorbance of the extract at 445 nm; y is the volume of extract (mL); A1% cm represents the extinction coefficient of carotenoids (A1 % cm = 2592), and w is the sample weight (g).

2.4.5. Determination of antioxidant activity by DPPH•radical-scavenging activity

A Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay was performed according to the method adopted by Olugbami et al. [11]. In a 96-well flat - bottomed micro - plate on ice, a 20 μ L aliquot of the extract was added to 20 μ L of distilled water. After 200 μ L of 118.3 mg/L DPPH radical solution was added, the mixture was mixed thoroughly. Ethanol was used to produce the DPPH radical solution. After the plates were stored in the dark for 30 min on ice, the absorbance was measured using a plate reader (1510, Thermo Fisher, USA) at 515 nm. A control containing 20 μ L of ethanol (no extract) was also included in each plate. The DPPH radical scavenging activity was calculated using Eq. (5).

DPPH radical scavenging activity (%) =
$$\left(\frac{Abs_{control} - Abs_{sample}}{Abs_{control}}\right) \times 100$$
 (5)

2.5. Kinetic analysis

The data obtained from the degradation of total phenolic content, total carotenoid content, total anthocyanin content, and DPPH radical scavenging activity of red amaranth and red skin potato was subjected to first order kinetic analysis [12] using the following first orders kinetic Eq. (6)

$$C = C_0 \exp(-kt)$$
(6)

Where, k is the first order rate constant, t is boiling time, C_0 is initial concentration, C_t is their concentrations after heating for time (t).

The thermal resistance time or decimal reduction time (D values), the time required to reduce the nutrient concentration by 90 %, was related to reaction rate constants following equation (7)-

$$D = \frac{2.303}{k}$$
(7)

2.6. Statistical Analysis

The results were shown as the mean \pm standard deviation of three parallel measurements. Differences between variables were tested for significance using analysis of variance. All data were the mean values of three replicates and the data were analyzed for significance at the p < 0.05 level. Non - linear regression analysis equipped kinetic models with Origin pro 8.5 statistical software.

3. Results and discussion

3.1. Effect of boiling on physicochemical properties of red amaranth and red skin potato

Physicochemical properties of red amaranth and red skin potato during different boiling time are presented in Table 1 and 2 respectively. The result shows that the moisture content in red amaranth and red potato was not changed significantly with increasing the boiling times. Whereas the ash content in both samples was significantly reduced by increasing the boiling time. These results are consistent with the findings reported for the boiling and leaching of potatoes by Bethke and Jansky [13], who also found reductions in phosphorus, magnesium, sulfur, zinc, manganese and iron during boiling in 6 potato cultivars tubers. The pH value increased by increasing the boiling time for both red amaranth and red potato (p<0.05).

Boiling Time (min)	Moisture (%)	Content	Ash Content (%)	рН	Water Solubility
					Index (% w31)
0	86.62±1.90ª		1.3 ± 0.01^{a}	6.08 ± 0.04^{d}	-
1	92.23 ± 2.20^{a}		0.95 ± 0.01^{b}	6.85±0.01 ^c	1.69 ± 0.27^{d}
5	91.32 ± 2.40^{a}		0.92±0.01 ^c	7.45 ± 0.05^{b}	3.45 ±0.92°
10	$90.46^{\pm}2.57^{a}$		0.90 ± 0.01^{d}	7.45 ± 0.07^{b}	6.61 ±1.45 ^b
20	90.27 ± 2.64^{a}		0.85±0.01 ^e	7.50 ± 0.04^{a}	16.69 ± 3.13^{a}

Table 1 Physicochemical properties of red amaranth

All values are of means of triplicate determination expressed on wet weight basis ± standard deviation. Mean values in the following row sharing a common letter are not statistically significant (p<0.05).

This may occur due to the release of ascorbic acid from cell disintegration and along with its destruction during boiling. The % of water solubility index increased with increasing boiling time in both red amaranth and red skin potatoes. This finding may be due to the water leaching of soluble sugars and organic acids.

able 2 Physicochemical properties of red skin potato

Boiling Time (min)	Moisture Content (%)	Ash Content (%)	рН	Water Solubility Index (% WSI)
0	81.54±2.35 ^A	$1.19 \pm 0.05^{\text{A}}$	$6.00 \pm 1.00^{\text{D}}$	
1	85.23±2.20 ^A	0.98 ± 0.05^{B}	6.58±1.11 ^A	0.70 ± 0.02^{D}
5	85.30±2.30 ^A	0.89±0.03 ^c	6.51 ± 1.92^{B}	1.35±0.10 ^c
10	84.33±2.35 ^A	$0.85 \pm 0.01^{\text{D}}$	6.48±2.01 ^{CB}	2.27 ± 0.50^{B}
20	84.50±2.40 ^A	0.80 ± 0.04^{E}	6.50 ± 1.51^{B}	4.67±1.05 ^A

All values are of means of triplicate determination expressed on wet weight basis ± standard deviation. Mean values in the following row sharing a common letter are not statistically significant

(p<0.05).

3.2. Effect of boiling on bioactive compounds of red amaranth and red skin potato

Bioactive compounds are extra - nutritional constituents, available mainly in fruits and vegetables that provide additional health benefits to humans. During cooking by boiling at the domestic level, due to thermal degradation, dilution and leaching into the water used for treatment, the composition and content of various bioactive components are very likely to be affected [14-15]. Losses of these nutrients must be taken into account while developing new products and/or processes. Kinetic models are used for fast quality assessment and prediction; in addition, they can be employed to predict the influence of processing on critical quality parameters. In this study, we investigated the effect of boiling in the % of loss and first order of degradation kinetics of total polyphenol content (TPC), total anthocyanin content (TAC), total carotenoid content (TCC), and DPPH radical scavenging activity (RSA) of red amaranth and red skin potato.

3.2.1. The effect of boiling on total phenolic content (TPC)

The effect of boiling time on total phenolic content of raw, and boiled red amaranth and red skin potato is shown in Figure 1 and the % of losses is shown in Table 3. In the present study, the TPC in red amaranth was found to decrease significantly by increasing the boiling time and the loss at 20 minutes of boiling was about 38.21±2.01 % of its initial level. Domestic boiling opens the cell matrix, facilitates the extractability and bio-accessibility of total phytochemicals, promotes the release of bound phytochemicals and forms soluble low - molecular weight phytochemicals that can be easily degraded by the heating process and leads to a decrease in total phenolic content [16].



Figure 1 Total phenolic content of red amaranth and red skin potato at the different boiling time

The first order kinetic model for the degradation of bioactive compounds during boiling treatment was determined in agreement with other studies [17]. A graph was plotted between boiling time and C_t/C_0 values in order to evaluate the relationship between boiling time and degradation of TPC compounds [Figure 5 (a)]. From the 1st order kinetics model it can be seen that the loss rate was about 0.025 mg / min and the thermal resistance time (D) value was 92.12 min.

The R^2 values obtained from the graph were 0.961 indicated a good correlation between the predicted value and the experimental values and also indicated a strong correlation between the boiling time and the TPC content in red amaranth.

Similarly, the raw red skin potato contained TPC 583.65 \pm 2.40 mg GAE/100 g and gradually decreased to 390.65 \pm 4.05 mg GAE/100 g at 20 minutes of boiling, resulting in losses of 33.06 \pm 1.94 % (Table 3). The findings are lower with the results reported by Perla, Holm, & Jayanty [18], who found that boiling reduced the total phenolic content by about 62.02 %. In the kinetic study, it was found that the D - value is 115.15 min in red skin potato and the loss rate is 0.020 mg / min (Table 3). The R² value is 0.98 indicate a strong relation of TPC with time (Table 3). Increases in cooking time and temperature can exacerbate the loss of phenolic compounds due to thermal degradation or the transfer of hydrophilic phenolics from red amaranth and red skin potatoes to water [16]. Lemos et al. [9] also reported that cooking had a detrimental effect on the total phenolic levels in purple majesty potatoes.

Table 3 The percentage (%) of loss, kinetic parameter estimated (k) and statistical values of first-order kinetic modelof bioactive compounds and antioxidant activity

Daramotor	Name	Time (min)	The % of loss	Degradation kinetics			
				D	k	R ²	
		0	-				
	Red amaranth	1	1.10 ± 0.02				
		5	17.74±1.27	92.12	0.025	0.961	
		10	22.08±2.10				
Total phenolic content		20	38.21±2.01				
i otai pilenone content	Red skin potato	0	-				
		1	2.40±0.90				
		5	7.82±1.03	115.15	0.020	0.981	
		10	21.81±1.21				
		20	33.06±1.94				
	Red amaranth	0	-				
		1	3.78±0.57 ^c				
		5	13.35±1.67°	100.13	0.023	0.972	
		10	23.62 ± 2.0^{b}				
Tatal anthe granin contant		20	34.92±1.9 ^a				
Total anthocyanin content		0	-				
		1	4.05 ± 0.72^{d}				
	Red skin potato	5	21.90±1.23 ^c	76.76	0.030	0.086	
		10	32.12±2.78 ^b				
		20	37.82±2.31ª				
		0	-				
		1	3.03±0.02 ^c				
	Red amaranth	5	5.88±0.03°	329.00	0.008	0.953	
		10	9.09±0.19 ^b				
m , 1 , 1 ,		20	15.65±0.28 ^a				
Total carotenoids content	Red skin potato	0	-				
		1	1.33±0.01°				
		5	2.81±0.01°	329.00	0.007	0.989	
		10	7.98±0.51 ^b				
		20	14.64±0.19 ^a				
	Red amaranth	0	-				
		1	1.80±0.57 ^d				
		5	14.49±1.67°	85.29	0.027	0.968	
		10	28.01±2.0 ^b				
DPPH radical scavenging		20	38.36±1.90ª				
activity		0	-				
	Red skin potato	1	3.56 ± 0.72^{d}				
		5	7.68±1.23°	143.93	0.016	0.903	
		10	18.06±2.78 ^b				
		20	26.39±2.31ª				

3.2.2. Effect of boiling time on total anthocyanin content (TAC)

Anthocyanins are water - soluble colored pigments belonging to the phenolic group. The pigments are in glycosylated forms. Anthocyanins responsible for colors, red, purple, and blue, are present in fruits and vegetables. Anthocyanins are considered one of the most important pigment and health - promoting compounds in red amaranth and red skin potatoes. The compounds, however, are easily dissolved in water and broken down by thermal treatment [19]. Several boiling times were observed to induce distinct losses of total anthocyanin (Figure 2). The present study revealed that when prolonging their boiling time, the anthocyanin content was greatly reduced in both red amaranth and red skin potato. The anthocyanin content in raw red amaranth was 89.25±1.96 mg CGE/100 g which was gradually reduced to 57.89±2.17 mg CGE/100 g after 20 minutes of boiling, which is about 34.92±1.9 % loss (Table 3).



Figure 2 Total anthocyanin content of red amaranth and red skin potato at the different boiling time

In case of kinetic study, the rate of loss was observed 0.023 mg/min and D-value was 100.13. Similarly, the anthocyanin content in red skin potato gradually decreased from 198.30±8.63 (raw/0 min boiled) to 123.30±15.16 mg CGE/100 g at 20 minutes of boiling. At 20 minutes of boiling, the % of loss in red skin potato was 37.82±2.31 %. The loss rate in the red skin potato was estimated at 0.030 mg CGE / minute and D - value was 76.76. The higher R2 values indicated that the losses of TAC during the boiling period followed the first order kinetics. The experimental and predicted data are presented in the Figure 5. Tian et al. [20] reported that the anthocyanin of purple skinned potato was reduced during boiling and which was about the losses of 14.66 %. However, the extent of anthocyanin loss reported by these authors was different, which may have been due to the different cooking times, temperatures and pretreatments used in the different studies. The findings of the present study is in accordance with Ioannou et al. [21] who were reported that anthocyanin could be lost due to combined effect of thermal degradation and the soaking in the boiled water.

3.2.3. Effect of boiling time on total carotenoid content (TCC)

Carotenoids are isoprenoid molecules that are widespread in nature and have a wide range of functions due to their biological antioxidants (i.e. protecting against age - related macular degeneration and cataracts and reducing diseases, including cardiovascular diseases and cancer). It is a class of lipophilic compounds, less susceptible to leaching and less sensitive to heat. The changes in TCC during boiling at different boiling times are shown in Figure 3. In this study, the total carotenoid content in red amaranth (p<0.05) was not significantly changed until 10 minutes of boiling, but at 20 minutes TCC was reduced to 1.67 mg/100 g, which was about 15.56 % loss (Table 3). The TCC was also found to be significantly reduced in red skin potato after 10 minutes of boiling. At 20 minutes of boiling, the percentage of loss in both vegetables was 14 - 15 %.



Figure 3 Total carotenoids contents of red amaranth and red potato at the different boiling duration

This can be attributed to disruption of cell walls with food processing procedures, facilitating their release from proteins. But this behavior can vary with the food matrix, as these bioactive compounds are reported to increase and decrease. Unlike phenolics and anthocyanins, there are few data available on the effects of boiling time on carotenoids in red amaranth and red skin potato. In a study with five tropical leafy vegetables from Africa, Djuikwo et al. [22] also recorded losses of total carotenoids from 5 to 20 % after 10 minutes boiling. Kao et al. evaluated the effect of boiling in various carotenoid-rich green leafy vegetables; including Thai basil leaves and cilantro, and noted that total carotenoids content reached the maximum after boiling those vegetables for 5 minutes and 10 minutes [6]. A negative effect on the total carotenoids contents of the vegetables was noticed with more boiling time. The first order kinetic study showed that the rate of TCC loss was almost same 0.008 and 0.007 mg/min in red amaranth and red skin potato respectively. The time of thermal resistance was found 329.00 min in both vegetables. The predicted value and experimental values of the TCC and strong correlations (higher R² values 0.95 and 0.98) as shown in the Figure 5(c) demonstrated that the losses of total carotenoid vs boiling time followed the first order kinetics.

3.3. Effect of boiling on DPPH Radical Scavenging Activity (RSA)

In this study the total antioxidant activity was measured by DPPH Radical Scavenging Activity (RSA). The RSA levels in raw red amaranth and red skin potato and its changes after boiling is summarizes in Figure 4. RSA was found to have gradually reduced in both boiled red amaranth and red skin potato. In the boiled tissues of red amaranth after boiling for 5 min RSA decreased to 41.72±0.89 % which was 14.49±1.52 % loss from its initial level. Further significant loss of RSA to 38.36±2.94 % was observed when the boiling time was extended to 20 minutes (Table 3). Similar trend was found in red skin potato, which significantly increased by increasing the boiling time.



Figure 4 DPPH Radical Scavenging Activity (RSA) in red amaranth and red skin potato

The rate of degradation of TAA was estimated by first order kinetic model (Figure 5d). The result showed that the loss rate of red amaranth and red skin potato was 0.027/min and 016/min respectively. The thermal resistance of red

amaranth and red skin potato was 85.29 and 143.93 min respectively (Table 3). Puupponen-Pimia et al. reported that the cauliflower DPPH index decreased by 23 % during blanching in water [23]. The loss of antioxidant activity in cooked tissue during blanching was due to the large surface area of the vegetables in contact with the water [24]. Antioxidant levels were reported to decrease after aqua thermal treatment of broccoli [25] and selected cruciferous vegetables [26].



Figure 5 First order kinetics of (a) Total phenolic content, (b) Total anthocyanin content, (c) Total carotenoid content, and (d) DPPH radical scavenging activity (RSA)

4. Conclusion

The effects of boiling on physicochemical, antioxidants and degradation rate during different boiling time of red amaranth and red skin potatoes were evaluated. The present study indicated that total polyphenol, anthocyanin, and antioxidant activity of red amaranth and red skin potatoes were significantly reduced when prolonging the boiling time, whereas total carotenoid contents was less affected by boiling. The first order kinetic model was determined as the most appropriate model to represents the degradation of polyphenols, anthocyanin, carotenoids and total antioxidant activity of red amaranth and red skin potatoes during boiling. The results of our investigation demonstrated that it is vital to use less boiling time to minimize the loss of polyphenols, anthocyanin and total antioxidant activity. We concluded that the assessment of the effects of boiling time on the bioactive compounds and antioxidant activity of red amaranth and red skin potatoes helps in dietary survey evaluating, planning intake and also used for making recommendations on the food processing techniques to be chosen for preserving the most functional and bioactive compounds of vegetables.

Compliance with ethical standards

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Disclosure of conflict of interest

Author's has no conflict of interest.

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