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NUTRITIONAL QUALITY, BIOACTIVE COMPOUNDS AND ANTIOXIDANT ACTIVITY OF SELECTED AFRICAN INDIGENOUS LEAFY VEGETABLES AS INFLUENCED BY MATURITY AND MINIMAL PROCESSING

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ABSTRACT

The African indigenous vegetables (AIVs) are excellent sources of β-carotene, vitamin C, iron as well as protein, minerals, fiber and bioactive compounds. In the recent past, AIVs have gained commercial importance as a result of increased awareness of their nutritional and health benefits and are now produce in both formal and informal marketing channels. One of the challenges in production, marketing and consumption of AIVs is that they are highly perishable and there is inadequate capacity for their storage in fresh state. This is because most storage techniques require low temperatures, which are nonexistent for AIVs in Kenya. Minimal processing can enable AIVs produced in far flung locations to be stabilized and transported to the markets in the urban centres. However, this can affect the color, texture, flavor, and nutritional quality of AIVs. This study aimed at examining the influence of harvest maturity and minimal processing techniques on the nutritional, phytochemical and anti-oxidant capacity in stinging nettle, amaranth and black nightshade. The results indicated significant differences between treatments and stages of maturity. Results further show that the highest contents of β-carotene in fresh state, at young stage was 47.82 mg/100g in amaranth and mature stage was 71.22 mg/100g in black night shade. For vitamin C, the highest content was 142.06 mg/100g in stinging nettle at young stage while amaranth had the highest content of vitamin C at mature stage as 193.52 mg/100g. The highest phenol content in fresh state was in black night shade at 1.09 g/100g and 1.29 g/100g at young stage and mature stage respectively. Among the processed, the highest content of vitamin C was seen in Freeze-Dried Unsliced Unblanched black nightshade at both young and mature stage as 86.64mg/100g and 111.14mg/100g respectively. For β-carotene, the highest content was reported on Freeze-Dried Unsliced Blanched in amaranth as 30.24mg/100g at young stage and mature stage had 57.12mg/100g in black nightshade.

Key words: Minimal processing, blanching, drying, African indigenous vegetable, maturity

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INTRODUCTION

African Indigenous Vegetables (AIVs) are gaining prominence as an excellent source of vitamins A and C, iron as well as protein, minerals and fiber. In addition, AIVs have been found to be high in antioxidants and other health related phytochemicals [1, 2].

The AIVs have gained commercial importance over the past 15 years as a result of the enormous growth in demand and market [3]. The AIVs are now retailed in supermarket chains and other lucrative markets, resulting in better incomes. To respond to this increase in demand, there has been a tremendous increase in production of AIVs in the country. For instance, 200 AIV varieties have been documented [4] and many of these are either cultivated or gathered from the wild [5].

High postharvest losses are incurred during harvesting, transport and retailing due to lack of adequate capacity to maintain cold chains in these AIVs [6]. The high perishability is mainly attributed to the high moisture content. In addition, large portions are lost after harvesting due to poor handling and marketing conditions. In Africa and Kenya in particular, a significant portion is wasted during the in-season abundance [6, 7].

Use of processing technologies can increase the shelf life of AIVs by maintaining quality, improve safety and thus prevent losses. Minimal processing of AIVs has the potential to create new market opportunities with employment at various levels and maximize returns from fresh produce. Minimalprocessing, however, can affect the color, texture, flavor, and nutritional quality. Hence, there is need to determine the effect of minimal processing on the nutritional and phytochemical qualities of some AIVs.

The objective of this study was to evaluate the influence of harvest maturity and minimal processing on the nutritional and phytochemical contents, and anti-oxidant capacity ofstinging nettle, amaranth and black nightshade. The AIVs; amaranth and black nightshade, were selected because of their higher consumption and availability in the market whereas stinging nettle was selected for comparison due to its traditional consumption as an AIV.

MATERIALS AND METHODS

Plant material

The study was carried out on *Amaranthus dubius* (Amaranth), *Solanumscabrum* (Black nightshade) and *Urtica dioica* (stinging nettle). Amaranth and black nightshade seeds were purchased from Simlaw seeds company, Kenya and grown in a randomized complete block design in an open-air field at the Jomo Kenyatta University of Agriculture and Technology (Juja, Kenya), experimental research farm. The seeds were directly sown into furrows at an inter-row spacing of 40cm. Two weeks after germination, the plants were thinned to a spacing of 15cm between the plants. During planting, compost manure was sprinkled at the rate of 10 tonnes/hectare. Planting was done during the month of December 2016 - February 2017. Stinging nettle was sourced from a commercial farmer in Juja farm.

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Harvesting and sample preparation

Leaves of the AIVs were harvested at two stages: young stage (5weeks) and mature stage (10weeks) after planting. After harvesting, AIVs were washed, with potable water, and allowed to drain the excess water, then divided into four portions for minimal processing treatments. Treatments involved blanching and unblanching, slicing and unslicing of leaf samples then subjecting them to either solar or freeze-drying. Fresh samples (no treatment) were used as a control. Slicing of the samples involved cutting the fresh leaves into small pieces of dimensions approximately 0.3cm × 0.5cm thickness. Blanching was done on the sliced and unsliced samples using hot water at 95±1°C for 30seconds with the ratio of 1:7 vegetables to water (g/ml) according to the method by Tanongkankit *et al*. [8]. The blanched samples were removed and immediately dipped in ice cold water (4°C) to stop any enzymatic activity. The blanched samples were then left in a wire mesh bucket to drain water. These samples were then solar dried or freeze-dried together with the unblanched samples.

Drying processes

Solar-drying

The processed AIV samples were spread in a single layer in 40 x 60 cm rectangular chambers. The solar drier structure measured 185 cm wide by 273 cm long by 255 cm high with door dimensions measuring 60 cm wide by 180 cm high. The top part of the structure was semicircular in shape with a radius of 50 cm and was entirely covered with a polyvinyl chloride (PVC) material. The PVC material filters radiation which can destroy light sensitive nutrients in the dried samples [9]. The drying chamber temperature ranged between 42 and 63°C while that of the solar dryer's leaf collector was between 40 and 73°C. Moisture content (M.C.) of the AIV leaves was determined during and after drying. Drying was finalized when the vegetables were brittle and the dried samples were stored in zip lock bags at -20°C for further analysis. Experiment was carried out in replicates and all results expressed in dry weight (dw) except antioxidant activity which was expressed in fresh weight (fw) basis.

Freeze-drying

A freeze-drier (Alpha1-4 LD plus-Martin Christ Model-101541; Germany) was used. Processed samples were placed in airtight ziplock bags and frozen in a freezer at -21°C for 72hrs. Before placing in a freeze-drier, zip lock bags were perforated to attain several vents. These allowed good balance of pressure and temperature inside and outside the bags during drying. The initial and final drying were carried out at temperature and pressure conditions recommended by the drier manufacturer, which were -41°C, 0.11 mbar, -47°C, 0.055 mbar, respectively for 48hrs.

Determination of moisture content

The moisture content was determined according to method 984.25 [10].

Determination of Crude fiber

Two grams of the sample, initial weight(w0) of vegetable was weighed into a conical flask and 200mL of 1.25% sulphuric acid, added and the solution was boiled for 1hour. The content was then filtered using a glass wool and washed with hot water. The residue

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was transferred to a 500mL conical flask and 200mL of 1.25%NaOHwas added. The solution was boiled for 1hour and filtered using a glass wool. The residue was then washed with 7mL each of hot water, 1%HCL, methanol and petroleum ether and air dried for about 30minutes. The glass filter was then dried in an oven at 105°C for 1hour, and the first weight (w1) recorded. The glass filter was then put in a muffle furnace at 600°C for 1hour, and left to cool at room temperature then second weight (w2) recorded. The crude fiber was then calculated as follows:

$$crude\ fiber = \frac{W1 - W2}{W0} * 100$$

Where
w0 - initial weight of the sample
w1 – weight of the extracted fiber before ashing
w2 – weight of the fiber after ashing

Determination of Beta carotene

Two grams of AIV samples were weighed and extracted with about 10mL acetone by grounding thoroughly in a mortar and pestle. The acetone extract was then transferred to 100mL volumetric flask and the residue extracted again, with about 10mL acetone. This was repeated until the residue no longer gave orange color to acetone. The combined extract was made to 100mL mark. Twentyfive mL of the extract was evaporated to dryness on a rotary vacuum evaporator and the residue dissolved in about 1ml petroleum ether. The solution was introduced into chromatographic column that was packed with cotton wool and silica gel and eluted with about (10Ml) petroleum ether and collected up to 25mL. The absorbance of the solution was determined at 440nm using UV-vis spectrophotometer (Shimadzu model UV-1601 PC, Kyoto, Japan) and plotted against their corresponding standard concentrations.

Determination of Vitamin C content

Five grams of the sample was weighed and extracted with 0.8% metaphosphoric acid by grinding in a mortar and pestle. The extract was then made to 20 mL with 0.8% metaphosphoric acid and centrifuged at 10000 rpm for 10 minutes. The supernatant was filtered and diluted with 10 mL of 0.8% metaphosphoric acid. This was then filtered using cotton wool and micro-filtered through 0.45 µ filter and 20µL injected into the HPLC. High-performance liquid chromatography analysis was done using Shimadzu (10A model; Tokyo, Japan) and a UV-Vis detector. The mobile phase was 0.8%metaphosphoric acid, at 1.1mL/min flow rate and wavelength of 266.0 nm.

Extraction of total phenols, total flavonoids and antioxidants

Five grams of samples were weighed into amber-colored bottles containing 50 mL of analytical grade methanol and vortexed for 3 hr. The solution was incubated in darkness for 48–72 hr at room temperature. The extracts were centrifuged for10 min at 13,000×*g*/relative centrifugal force (RCF) and supernatants used to determine the total phenolic content and antioxidant capacity.

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Determination of total phenolic content

Total phenolic content was determined by the Folin–Ciocalteu colorimetric method [11] with gallic acid as the standard. Two milliliters of 10% (v/v) Folin Ciocalteu reagent and 4 mL of 0.7 mol/L sodium carbonate were added to 1 mL of prepared sample extract. The mixture was vortexed and allowed to stand at room temperature for 2 hrs. The absorbance was measured at 765 nm using UV-Vis spectrophotometer (Shimadzu UV–1240), and results were expressed as gallic acid equivalent (GAE), milligrams/100 g of dry matter.

Determination of total flavonoid content

Colorimetric method was used for determination of flavonoids as described by Jagadish *et al*. [12] with slight modification. To a 10 ml volumetric flask, 1 ml of plant extract was taken and 3mL of 5% sodium nitrite solution was added. After 3 minutes, 3 mL of 10% aluminum chloride was added to the mixture, which was kept at room temperature for 5 more minutes, followed by the addition of 2ml of 1M sodium hydroxide. The mixture was vigorously shaken for 5 min and the volume made up to 10ml with water. Absorbance was measured at 415nm using UV-Vis spectrophotometer (Shimadzu model UV-1601 PC, Kyoto, Japan). The total flavonoid was quantified using quercetin standard and results presented as quercetin equivalent (QE)/g.

Determination of antioxidant activity

The free radical-scavenging activity was determined using diphenylpicrylhydrazyl radical (DPPH) according to Ayoola *et al*. [13]. The following concentrations of the extract were prepared, 0.01, 0.05, 0.1, 0.5, 1.0, 2.0 and 5mg/ml in methanol in cuvette placed in the spectrophotometer. One milliliter of the extract was placed in a test tube; 3 mL of methanol was added followed by 0.5 mL of 1 mM DPPH in methanol. The mixture was shaken vigorously and left to stand for 5 min. Vitamin C was used as the antioxidant standard at the same concentration as the extract. The absorbance of the resulting solution was measured at 517 nm with a UV-vis spectrophotometer. All tests were run in triplicate and the radical scavenging activity was then calculated using the following formula;

$$\%\ Inhibition\ of\ DPPH = \frac{(A_B - A_A)}{A_B} \times 100$$

Where:

A_B = absorption of the blank sample;

A_A = absorption of the extract.

Statistical analysis

Data were subjected to analysis of ANOVA using Stata version 12 software (Stata Corp.) while means were separated using Duncan test at 0.05 significance level.

RESULTS AND DISCUSSION

The results of moisture content (M.C) of young and mature leaves are presented in Tables 1 and 2, respectively. From the results, fresh sample of black nightshade that was unsliced blanched had the highest M.C of 89.34% compared to the other AlVs at young stage. After drying, the sliced treatments of solar dried stinging nettle retained the highest

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M.C of 7.14%. Moisture content of the vegetables declined with the maturation of the leaves. Similar findings have been observed by Florkowski *et al*. [14] who reported that *Cleome gynandra* had a decrease in M.C with maturation. This may be due to structural changes as leaf grows older probably due to transpiration and starch hydrolysis. The blanched samples had higher M.C as compared to the unblanched samples, possibly due to disruption of the leaf tissue cells, facilitating degradation and solubilization of watersoluble components [15]. This, therefore, leads to softening of leaf tissues, hence higher rates in removal of water.

In this study, the M.C of freeze-dried samples ranged between 1.31-7.57%, while solar dried samples had slightly higher M.C. suggesting that freeze-drying led to removal of more wateras compared to solar drying.

Result for fiber content of the three AIVs (Table 3) shows that there was no significant difference ($P > 0.05$) between the treatments. Results show that, fresh black nightshade had the highest fiber content of 12.89% and 9.53% for mature stage and young stage, respectively. On the other hand, the lowest percentage of fiber content (6.28 %.) was observed in sliced blanched solar dried samples of amaranth.

There was a significant difference ($P \leq 0.05$) between the fiber content of the leaves at the two stages of harvest. The fresh mature leaf samples had significantly higher values as compared to fresh young leaf samples in the study. This may be due to fiber material being more elaborate in mature leaf organs than in young leaf [16]. For the blanched samples, there was no significance difference ($P \geq 0.05$) in fiber content for both solar dried and freeze-dried samples. This could be due to the stability of fiber component found in the vegetables. In addition, no significant difference ($P \geq 0.05$) was observed in fiber content between the sliced and unsliced samples, blanched and unblanched samples and between the solar and freeze-dried samples.

The β-carotene results for young and mature stage AIV samples are presented in Table 4. From the results, the fresh samples of amaranth had a higher β-carotene content of 47.82mg/100g at young stage, while the blacknightshade had the highest β-carotene content at mature stage (71.22mg/100g). On the other hand, the lowest content was observed in the young and mature stage of stinging nettle in solar dried sliced unblanched with β-carotene content of 12.33mg/100g and 18.29mg/100gdw, respectively.

In this study, mature leaves were found to have significantly higher ($P < 0.05$) β-carotene content compared to the young leaves, suggesting that β-carotene increases with the maturation of the leaves. The blanched solar and freeze-dried samples had higher beta carotene content as compared to the unblanched samples. In this case, there was a significance difference ($p \leq 0.05$) between the unsliced unblanched and sliced unblanched samples of solar and freeze-dried treatments. On the other hand, both solar and freeze-dried samples of unblanched treatments were significantly lower as compared to the blanched treatments. As reported by Rickman *et al*. [17], thermal processing causes isomerization of all the naturally predominant trans-β-carotene to cis form due to presence of conjugated double bond hence higher β-carotene content in blanched samples. Slicing of the samples significantly affected the β-carotene content whereby

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lower amount was observed in sliced treatments compared to unsliced samples. The reduction of the β-carotene during slicing may be due to increase in surface area which could have promoted the oxidation of β-carotene [18]. The highest β-carotene of the three AIVs on solar drying was reported in unsliced blanched leaves, followed by sliced blanched, unsliced unblanched and the lowest was observed in sliced unblanched leaves. On the other hand, solar dried samples had significantly lower β-carotene in all the vegetables ($P<0.05$) as compared to freeze-dried samples, probably due to degradation of some of the compounds by solar radiation [19].

The fresh samples of stinging nettle had higher vitamin C content (142.06mg/100g) table 5, at young stage, whereas amaranth had the highest vitamin C content at mature stage (193.52 mg/100g). Lowest vitamin C content, on the other hand, was observed in sliced blanched solar dried samples of young stage amaranth and mature stage stinging nettle with values of 11.87 mg/100g and 30.66 mg/100g, respectively.

Vitamin C was shown to increase with the maturation of the leaves. These findings concur with other studies in broccoli leaves [20] and spinach leaves [21]. Blanching was observed to significantly affect the vitamin C content ($P < 0.05$). Blanched samples had lower vitamin C compared to the unblanched samples. The results of blanching agree with the findings of a study by Volden *et al*. [22] which showed that blanching affects the vitamin C content. Their study reported a loss of 41% of vitamin C content in blanched cauliflower as compared to the unblanched [22]. Since vitamin C is heat labile, much of it was lost during blanching and heat treatment [22]. In addition, Liu *et al*. [23] observed that the blanching temperature inactivates most of vitamin C enzymes thus inhibiting their accumulation. Slicing of the samples also affected the vitamin C content whereby, the unsliced blanched had significantly higher content as compared to sliced blanched samples. Besides, the sliced unblanched samples showed significantly higher content as compared to sliced blanched. It has been reported that slicing of the vegetables increases the surface area, therefore altering the availability of vitamin C [24]. There was a significant difference on the vitamin C content in solar and freeze-dried samples. Freeze-dried samples had a higher content than solar dried samples.

The total flavonoid results for AIV samples are shown in Table 6. These results indicate that black nightshade at young and mature stage had higher flavonoid content of 1.2g/100g and 1.52g/100g (qe), respectively as compared to the dried samples. This was followed by unsliced unblanched freeze-dried samples of black nightshade which had the highest flavonoid content of 0.74 g/100g and 1.41 g/100g (QE) in young and mature stage, respectively. The lowest flavonoids were observed in sliced blanched solar dried samples of both young and mature stage stinging nettle with values of 0.23 g/100g and 0.36 g/100g, respectively.

All mature AIV leaves had significantly higher ($P < 0.05$) flavonoid as compared to the young leaves, therefore, suggesting that total flavonoid content increases with plant maturity. These results concur with the findings of Pandjaitan *et al*. [25] who observed increased flavonoid content in mature spinach leaves compared to immature leaves. Blanching affected the flavonoid content whereby the blanched samples had significantly ($P < 0.05$) lower content of flavonoids in the three AIVs as compared to the unblanched

samples. This suggests that blanching affected the chemical components especially the flavonoid, which likely leached into the blanching water. Slicing of the samples significantly ($P < 0.05$) affected the flavonoids. Unsliced samples had significantly higher flavonoid as compared to the sliced samples. Sliced unblanched samples had significantly higher flavonoids as compared to sliced blanched but significantly lower as compared to unsliced unblanched. According to Dos Reis *et al*. [24], slicing of vegetables alters the bioavailability of bioactive compounds such as flavonoids.

The total phenol contents had the same trend as the flavonoids (table 7) with fresh samples of young and mature stage black nightshade reporting higher contents of 1.09 g/100g and 1.29 g/100g, respectively as compared to dried samples. Unsliced unblanched freeze-dried samples of black nightshade on the other hand reported a higher value of 0.51g/100g and 1.07g/100g in young and mature stage, respectively. Sliced blanched solar dried samples had the lowest value of total phenols with stinging nettle being the most affected at both stages.

The total phenols in the studied AIVs ranged between 0.05 -1.29 g/100g. However, the values were significantly lower than the ranges of 3.23 g/100g - 11.7 g/100g reported by Zainol [26]. Phenol content increased with maturity as higher concentration was observed in mature leaves than the young leaves. Similar results were observed by Igbal and Bhanger [27] who reported increase in polyphenols concentration as leaf matures. Total phenols were affected by blanching and the drying methods. Blanched samples had significantly lower phenols than the unblanched samples. Sliced samples reported low levels of phenols as compared to the unsliced. Dos Reis *et al*. [24] reported that chopping alters the bioavailability of bioactive compounds such as carotenoids, polyphenols and flavonoids. Aditha *et al*. [28] also found out that raw amaranth extract had higher total phenolic content as compared to blanched counterpart. Similarly, Amin *et al*. [29] reported a loss of 71% of total phenolic content in blanched Amaranth. According to Aditha *et al*. [28], blanching of vegetables leads to oxidation of the compounds specifically, phenolics thus affecting their concentration. In addition, since phenolic compounds are known to occur in soluble forms and in combination with cell wall components in plants [30], the high temperature of the blanching may also lead to the disruption of the cell walls and the breakdown of the phenolics. This leads to leaching of these compounds into the blanching water. On the other hand, fresh and freeze-dried samples had significantly higher phenols than the solar dried samples.

The antioxidant activity results for AIVs are shown in Table 8. The IC_{50} values were high in fresh black nightshade at 2.4mg/ml and 1.11mg/mL in amaranth at mature stage. On the other hand, the IC_{50} values of the blanched samples were significantly higher ($p \leq 0.05$) as compared to the unblanched samples. The IC_{50} values of stinging nettle and amaranth for unsliced blanched solar dried similarly to unsliced blanched freeze-dried samples were significantly lower (2.48mg/ml and 2.4mg/ml (fresh weight respectively) as compared to amaranth of sliced blanched solar dried (2.5mg/ml) but significantly higher as compared to sliced unblanched solar dried (2.35mg/ml) of amaranth.

Significant changes in antioxidant activity were observed for the different growth stages, treatments and drying methods. The IC_{50} values are inversely proportional to the

antioxidant activity where the higher the IC_{50} the lower the antioxidant activity and vice versa. Therefore, from the results, it shows that there is a significant increase in antioxidant activity as the AIVs leaves matures. There was a significant difference ($P < 0.05$) on the antioxidant activity of the blanched samples of both solar and freeze-dried treatments. The antioxidant activity of the blanched samples was significantly lower (2.5mg/ml) ($P < 0.05$) as compared to the unblanched samples (2.01 mg/ml). Similarly, for the solar and freeze-dried samples, the antioxidant activity of unsliced blanched (2.48 mg/ml) was significantly higher as compared to sliced blanched (2.5 mg/ml) but significantly lower as compared to sliced unblanched (2.3 mg/ml). On the other hand, there was a significant difference ($P < 0.05$) on the unblanched samples. Higher antioxidant activity was observed in freeze and solar dried unsliced unblanched (0.02 mg/ml) and lowest in sliced unblanched (2.35 mg/ml). Lower antioxidant activity on the blanched samples might have been due to leaching of soluble antioxidants into the blanching water. Besides, slicing also had significant effect on the antioxidant activity since lower antioxidant activity was reported for the sliced samples as compared to the unsliced samples. The results of study concur with findings by Sreelatha [31] and Pandjaitan [25] which found out that antioxidants are affected by stage of maturity in*Moringa oleifera* and spinach leaves, respectively.

CONCLUSION

From the study, it is evident that the AIV maturity as well as the different processing techniques affects their nutritional and phytochemical composition. The fiber, vitamin C, β-carotene, phenols, flavonoids and antioxidant activity increased with the maturity of the leaves. Freeze-drying retained vitamins C, β-carotene, phenols, flavonoids and antioxidant activity better than solar dried. The fiber content of AIV leaves were not affected by the drying method or even processing like slicing and blanching. On the other hand, blanching of the three ALVs; stinging nettle, amaranth and black nightshade was shown to affect the nutritional and phytochemical quality where in all the parameters it decreased during blanching, unlike β-carotene where its content was higher as compared to unblanched samples. For the sliced samples, all the parameters were affected, resulting to lower content on the AlVs.

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Table 1: Effect of processing on moisture % of young stinging nettle, amaranth and black nightshade

	Percentage moisture content								
	Stinging Nettle			Amaranth			Black nightshade		
Treatments	Fresh	Solar Dried	Freeze-Dried	Fresh	Solar Dried	Freeze-Dried	Fresh	Solar Dried	Freeze-Dried
Fresh	76.60± 0.27^{bA}			80.44± 0.71^{bB}			87.28± 0.79^{bC}		
UU	76.60± 0.27^{bF}	6.76±0 .31^{aE}	5.50±0. 13^{aD}	80.44± 0.71^{bG}	4.7±0. 19^{bC}	3.49±0. 02^{bB}	87.28± 0.79^{bH}	3.3±0. 08^{bB}	1.35±0. 03^{aA}
UB	80.25± 0.01^{dF}	6.96±0 .01^{aE}	5.27±1. 03^{aD}	83.43± 0.33^{dG}	4.73±0 .21^{bC}	3.62±0. 10^{bB}	89.34± 0.68^{cH}	3.83±0 .16^{bB}	1.62±0. 19^{aA}
SU	75.39± 0.19^{aE}	7.04±0 .17^{bD}	5.26±0. 07^{aC}	79.04± 0.13^{aF}	5.82±0 .05^{cC}	3.34±0. 37^{bB}	86.64± 0.07^{aG}	2.99±1 .05^{aB}	1.26±0. 11^{aA}
SB	78.68± 0.23^{cE}	7.14±0 .21^{bD}	5.10±0. 27^{aC}	82.53± 0.89^{cF}	2.26±0 .42^{aA}	2.76±0. 92^{aA}	88.00± 0.05^{cG}	3.18±0 .17^{bB}	2.04±0. 09^{bA}

Values are given as percentage moisture means of three replicates ± SD. Means with different superscript uppercase letters across the row and lower case within the column are significantly different ($P < 0.05$). Fresh - no processing, UU - unsliced unblanched, UB - unsliced blanched, SU - sliced unblanched, SB -sliced blanched

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Table 2: Effect of processing on moisture % of mature stinging nettle, amaranth and black nightshade

	Percentage moisture content								
	stinging nettle			Amaranth			Black nightshade		
Treatments	**Fresh**	**Solar Dried**	**Freeze-Dried**	**Fresh**	**Solar Dried**	**Freeze-Dried**	**Fresh**	**Solar Dried**	**Freeze-Dried**
Fresh	74.95± 0.97^{bA}			78.22± 0.66^{bB}			85.78± 0.27^{bC}		
UU	74.95± 0.97^{bF}	7.08±1 .29^{aE}	4.32±0. 17^{aC}	78.22± 0.66^{bG}	6.09±0 .47^{bD}	2.31±0. 51^{bB}	85.78± 0.27^{bH}	1.74±0 .41^{aA}	2.05±0. 02^{bB}
UB	75.88± 0.27^{cE}	7.14±0 .61^{aD}	5.66±0. 03^{bC}	81.2±0. 39^{dF}	2.37±0 .37^{aB}	1.53±0. 01^{aA}	87.22± 0.31^{dG}	2.2±0. 09^{bB}	1.97±0. 03^{aA}
SU	71.66± 1.01^{aE}	7.57±0 .91^{aD}	4.76±0. 19^{aC}	75.01± 0.07^{aF}	2.03±0 .99^{aB}	1.31±0. 09^{aA}	82.97± 0.89^{aG}	4.6±0. 23^{dC}	1.5±0.2 9^{aA}
SB	76.31± 0.87^{dF}	7.06±0 .11^{aE}	4.59±0. 31^{aD}	80.05± 1.03^{cG}	2.52±0 .77^{aB}	1.42±0. 11^{aA}	86.31± 0.07^{cH}	3.01±1 .03^{cC}	1.54±0. 81^{aA}

Values are given as percentage moisture content means of three replicates ± SD. Means with different superscript uppercase letters across the row and lower case within the column are significantly different ($P < 0.05$). Fresh - no processing, UU - unsliced unblanched, UB - unsliced blanched, SU - sliced unblanched, SB -sliced blanched

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Table 3: Effect of harvest maturity and processing on % crude fiber of young and mature stinging nettle, Amaranth and Black nightshade (on dry weight basis)

	Stinging nettle		Amaranth		Black nightshade	
Fiber	**Young**	**Mature**	**Young**	**Mature**	**Young**	**Mature**
SDUU	9.07 ± 0.65^{cB}	11.98 ± 0.53^{bB}	6.25 ± 0.57^{bA}	10.69 ± 0.26^{aB}	7.48 ± 0.36^{aA}	11.6 ± 0.38^{aB}
SDUB	8.44 ± 0.22^{bA}	11.18 ± 0.19^{bB}	6.09 ± 0.32^{bA}	10.85 ± 0.25^{aB}	7.52 ± 0.24^{aA}	12.05 ± 0.46^{bC}
SDSU	8.32 ± 0.48^{bA}	11.05 ± 0.74^{bB}	5.96 ± 0.5^{aA}	10.45 ± 0.18^{aB}	7.55 ± 0.12^{aA}	11.77 ± 0.17^{aB}
SDSB	7.51 ± 0.54^{aA}	10.01 ± 0.88^{aB}	6.4 ± 0.63^{bA}	10.03 ± 0.66^{aB}	7.14 ± 0.12^{aA}	10.76 ± 0.51^{aB}
Fresh	9.53 ± 0.17^{cA}	12.48 ± 1.13^{cC}	7.51 ± 0.3^{cA}	10.96 ± 0.67^{aB}	7.59 ± 0.66^{aA}	12.77 ± 0.17^{bC}
FDUU	9.17 ± 0.16^{cB}	11.55 ± 0.67^{bC}	6.63 ± 0.51^{bA}	11.43 ± 0.73^{bC}	7.62 ± 0.67^{aA}	12.18 ± 0.23^{bC}
FDUB	8.93 ± 0.19^{bA}	11.15 ± 0.91^{bB}	6.5 ± 0.55^{bA}	10.84 ± 0.55^{aB}	7.2 ± 0.65^{aA}	12.89 ± 0.25^{bC}
FDSU	8.35 ± 0.13^{bA}	11.17 ± 0.41^{bB}	6.28 ± 0.47^{bA}	10.91 ± 0.22^{aB}	7.79 ± 0.51^{aA}	11.29 ± 0.56^{aB}
FDSB	8.15 ± 0.14^{bA}	10.84 ± 0.9^{aB}	6.36 ± 0.24^{bA}	10.38 ± 0.54^{aB}	7.69 ± 0.14^{aA}	12.41 ± 0.17^{bC}

Values are given as (%) of three replicates ± SD. Means with different superscript uppercase letters across the row (per parameter) and lower case within the column are significantly different ($P < 0.05$). SDUU-solar dried unsliced unblanched, SDUB-solar dried unsliced blanched, SDSU-solar dried sliced unblanched, SDSB-solar dried sliced blanched. FDUU-freeze-dried unsliced unblanched, FDUB-freeze-dried unsliced blanched, FDSU-freeze dried sliced unblanched, FDSB-freeze-dried sliced blanched and fresh-fresh sample (no processing)

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Table 4: Effect of harvest maturity and processing on beta-carotene of young and mature stinging nettle, Amaranth and Black nightshade (on dry weight basis)

	Stinging Nettle		Amaranth		Black nightshade	
Beta-Carotene	**Young**	**Mature**	**Young**	**Mature**	**Young**	**Mature**
SDUU	17.08±1.59^{bA}	27.15±1.31^{dB}	23.31±1.53^{bB}	40.29±1.52^{cC}	22.24±1.43^{bB}	47.64±1.62^{cC}
SDUB	26.86±1.61^{dA}	18.29±1.14^{aB}	27.25±1.06^{fA}	45.74±1.71^{fC}	27.27±2.74^{eA}	54.82±2.49^{gD}
SDSU	12.33±1.56^{aA}	30.96±0.91^{fA}	18.36±1.67^{aB}	34.52±0.94^{aC}	20.18±0.8^{aB}	41.15±1.66^{aD}
SDSB	22.66±1.49^{cA}	20.21±2.47^{bB}	25.87±1.63^{dB}	42.46±1.71^{dC}	25.5±2.45^{dB}	50.92±1.48^{eD}
Fresh	33.67±2.48^{fA}	41.45±1.56^{gB}	47.82±1.32^{hB}	64.35±1.46^{hC}	33.94±2.32^{iA}	71.22±0.87^{iC}
FDUU	21.55±1.52^{cA}	24.44±2.51^{cA}	26.18±0.87^{eA}	42.63±2.18^{dB}	24.74±1.29^{cA}	49.15±1.55^{dB}
FDUB	26.66±1.72^{dA}	29.27±1.4^{eA}	30.24±1.35^{gA}	44.07±1.89^{eB}	29.19±2.4f^{A}	57.12±1.23^{hC}
FDSU	16.22±1.34^{bA}	20.57±1.57^{bB}	24.97±2.2^{cB}	38.22±2.4^{bC}	22.41±1.57^{bB}	46.7±0.96^{bC}
FDSB	28.67±2.39^{eA}	34.07±0.92^{cAB}	27.55±2.4^{eB}	48.04±2.18^{gB}	27.26±2.79^{eA}	53.36±2.62^{fB}

Values are given as means (mg/100g) of three replicates ± SD. Means with different superscript uppercase letters across the row (per parameter) and lower case within the column are significantly different ($P < 0.05$). SDUU-solar dried unsliced unblanched, SDUB-solar dried unsliced blanched, SDSU-solar dried sliced unblanched, SDSB-solar dried sliced blanched. FDUU-freeze-dried unsliced unblanched, FDUB-freeze-dried unsliced blanched, FDSU-freeze-dried sliced unblanched, FDSB-freeze-dried sliced blanched and fresh-fresh sample (no processing)

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Table 5: Effect of harvest maturity and processing on vitamin C of young and mature stinging nettle, amaranth and black nightshade (on dry weight basis)

	Stinging Nettle		Amaranth		Black nightshade	
Vitamin C	**Young**	**Mature**	**Young**	**Mature**	**Young**	**Mature**
SDUU	36.86±2.66^{dA}	50.67±4.61^{fB}	53.41±3.11^{gB}	67.7±4.95^{dC}	84.74±2.99^{gD}	103.66±3.04^{fE}
SDUB	21.65±3.96^{bA}	37.46±2.52^{bB}	33.94±3.96^{cB}	43.29±2.94^{bB}	34.44±3.02^{bB}	89.35±2.97^{cC}
SDSU	29.26±2.8^{cA}	43.91±3.45^{dB}	44.2±2.93^{eB}	59.67±2.47^{cC}	78.97±2.66^{eD}	96.52±2.72^{dE}
SDSB	17.69±3.83^{aA}	30.66±3.12^{aB}	11.87±2.24^{aA}	37.37±2.96^{aB}	20.52±3.01^{aA}	66.82±2.91^{aC}
Fresh	142.06±2.71^{hB}	181.48±3.22^{iC}	102.14±2.53^{iA}	193.52±3.04^{hC}	124.64±3.57^{iAB}	177.97±3.73^{hC}
FDUU	53.98±2.32^{gA}	68.23±3.58h^{AB}	55.07±3.4^{hA}	98.46±2.99^{gB}	86.64±4.37^{hB}	111.14±2.41^{gC}
FDUB	35.44±2.17^{dB}	47.87±3.52^{eA}	41.76±2.68^{dA}	68.93±6.44^{dD}	72.23±2.43^{dB}	95.35±2.81^{dC}
FDSU	45.17±2.22^{fA}	61.82±2.33^{gB}	14.62±2.56^{bA}	94.98±2.27^{fC}	52.95±2.64^{cC}	101.48±3.9^{eD}
FDSB	40.61±2.08^{eA}	39.42±2.2^{cB}	49.13±2.09^{fA}	80.63±3.11^{eBC}	82.78±3.23^{fC}	74.89±3.66^{bD}

Values are given as means (mg/100g) of three replicates ± SD. Means with different superscript uppercase letters across the row (per parameter) and lower case within the column are significantly different ($P < 0.05$). SDUU-solar dried unsliced unblanched, SDUB-solar dried unsliced blanched, SDSU-solar dried sliced unblanched, SDSB-solar dried sliced blanched. FDUU-freeze-dried unsliced unblanched, FDUB-freeze-dried unsliced blanched, FDSU-freeze-dried sliced unblanched, FDSB-freeze-dried sliced blanched and fresh-fresh sample (no processing)

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Table 6: Effect of harvest maturity and processing on total flavonoid content of young and mature stinging nettle, amaranth and black nightshade (on dry weight basis)

	Stinging Nettle		Amaranth		Black nightshade	
Flavonoids	**Young**	**Mature**	**Young**	**Mature**	**Young**	**Mature**
SDUU	0.46 ± 0.03^{gA}	0.48 ± 0.01^{fA}	0.47 ± 0.02^{fA}	1.16 ± 0.02^{fC}	0.65 ± 0.02^{dB}	0.65 ± 0.02^{dB}
SDUB	0.28 ± 0.02^{bA}	0.4 ± 0.03^{bB}	0.3 ± 0.02^{bA}	0.77 ± 0.02^{aC}	0.56 ± 0.04^{bB}	0.56 ± 0.04^{bB}
SDSU	0.4 ± 0.03^{eA}	0.45 ± 0.02^{dA}	0.39 ± 0.02^{dA}	0.82 ± 0.04^{cC}	0.59 ± 0.03^{cB}	0.59 ± 0.03^{cB}
SDSB	0.23 ± 0.02^{aA}	0.36 ± 0.04^{aA}	0.23 ± 0.03^{aA}	0.73 ± 0.02^{iC}	1.2 ± 0.02^{gC}	1.2 ± 0.02^{gC}
Fresh	0.3 ± 0.03^{cA}	0.78 ± 0.05^{iB}	0.8 ± 0.03^{hB}	1.49 ± 0.04^{hC}	0.52 ± 0.03^{aB}	0.52 ± 0.03^{aB}
FDUU	0.52 ± 0.02^{hA}	0.55 ± 0.02^{hA}	0.31 ± 0.02^{cA}	1.23 ± 0.04^{gC}	0.74 ± 0.02^{fB}	0.74 ± 0.02^{fB}
FDUB	0.35 ± 0.03^{dA}	0.47 ± 0.02^{eA}	0.39 ± 0.02^{dA}	0.78 ± 0.02^{bC}	0.65 ± 0.02^{dB}	0.65 ± 0.02^{dB}
FDSU	0.44 ± 0.02^{fA}	0.5 ± 0.03^{gA}	0.44 ± 0.03^{eA}	0.89 ± 0.02^{eB}	0.69 ± 0.04^{eB}	0.69 ± 0.04^{eB}
FDSB	0.63 ± 0.03^{iA}	0.43 ± 0.03^{cA}	0.51 ± 0.03^{gA}	0.86 ± 0.03^{dC}	0.59 ± 0.02^{bB}	0.59 ± 0.02^{bB}

Values are given as means (g/100g) of three replicates ± SD. Means with different superscript uppercase letters across the row (per parameter) and lower case within the column are significantly different ($P < 0.05$). SDUU-solar dried unsliced unblanched, SDUB-solar dried unsliced blanched, SDSU-solar dried sliced unblanched, SDSB-solar dried sliced blanched. FDUU-freeze-dried unsliced unblanched, FDUB-freeze-dried unsliced blanched, FDSU-freeze-dried sliced unblanched, FDSB-freeze-dried sliced blanched and fresh-fresh sample (no processing)

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Table 7: Effect of harvest maturity and processing on total phenol content of young and mature stinging nettle, amaranth and black nightshade (dry weight basis)

	Stinging Nettle		Amaranth		Black nightshade	
phenols	**Young**	**Mature**	**Young**	**Mature**	**Young**	**Mature**
SDUU	0.27 ± 0.02^{fA}	1.01 ± 0.04^{hB}	0.37 ± 0.03^{fB}	0.56 ± 0.06^{eE}	0.45 ± 0.05^{fB}	1.03 ± 0.02^{gC}
SDUB	0.09 ± 0.03^{bA}	0.19 ± 0.02^{aA}	0.21 ± 0.08^{bB}	0.33 ± 0.03^{bC}	0.21 ± 0.05^{bB}	0.62 ± 0.02^{cD}
SDSU	0.24 ± 0.04^{eA}	0.41 ± 0.05^{dB}	0.34 ± 0.03^{eA}	0.47 ± 0.04^{dB}	0.33 ± 0.09^{dA}	0.94 ± 0.07^{eC}
SDSB	0.05 ± 0.02^{aA}	0.3 ± 0.02^{bC}	0.19 ± 0.02^{aA}	0.27 ± 0.03^{aB}	0.11 ± 0.03^{aA}	0.39 ± 0.03^{aB}
Fresh	0.89 ± 0.05^{iA}	0.49 ± 0.04^{fB}	0.85 ± 0.04^{iA}	$1.01\pm0.06g^{B}$	1.09 ± 0.03^{hB}	1.29 ± 0.08^{iC}
FDUU	0.43 ± 0.04^{hA}	0.59 ± 0.02^{gB}	0.47 ± 0.04^{hA}	0.63 ± 0.03^{fB}	0.51 ± 0.03^{gB}	1.07 ± 0.04^{hC}
FDUB	0.17 ± 0.02^{dA}	0.44 ± 0.03^{eB}	0.3 ± 0.03^{dB}	0.39 ± 0.02^{cB}	$0.29\pm0.02c^{A}$	0.71 ± 0.04^{dC}
FDSU	0.34 ± 0.01^{gA}	0.49 ± 0.03^{fB}	0.25 ± 0.02^{cA}	0.56 ± 0.06^{eB}	0.21 ± 0.03^{bA}	1.01 ± 0.06^{fC}
FDSB	0.11 ± 0.03^{cA}	0.36 ± 0.02^{cB}	0.39 ± 0.02^{gA}	0.33 ± 0.02^{bB}	0.44 ± 0.07^{eB}	0.45 ± 0.03^{bC}

Values are given as means (g/100g) of three replicates ± SD. Means with different superscript uppercase letters across the row (per parameter) and lower case within the column are significantly different ($P < 0.05$). SDUU-solar dried unsliced unblanched, SDUB-solar dried unsliced blanched, SDSU-solar dried sliced unblanched, SDSB-solar dried sliced blanched. FDUU-freeze-dried unsliced unblanched, FDUB-freeze-dried unsliced blanched, FDSU-freeze-dried sliced unblanched, FDSB-freeze-dried sliced blanched and fresh-fresh sample (no processing)

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Table 8: Effect of harvest maturity and processing on antioxidant activity of stinging nettle, amaranth and black nightshade (mg/ml fresh weight basis for fresh samples and dried weight basis for dried samples)

	Young			Mature		
	Stinging Nettle	**Amaranth**	**Black nightshade**	**Stinging Nettle**	**Amaranth**	**Black nightshade**
SDUU	2.01 ± 0.00^{dD}	2.1 ± 0.00^{bE}	1.42 ± 0.01^{bC}	0.07 ± 0.01^{bA}	2.18 ± 0.02^{dF}	1.39 ± 0.09^{dB}
SDUB	2.48 ± 0.22^{gD}	2.48 ± 0.00^{fD}	2.38 ± 0.00^{fA}	2.15 ± 0.00^{gB}	2.48 ± 0.01^{gD}	2.4 ± 0.00^{hC}
SDSU	2.3 ± 0.01^{eD}	2.35 ± 0.01^{dE}	2.23 ± 0.00^{eC}	0.34 ± 0.04^{dA}	2.3 ± 0.01^{eD}	1.53 ± 0.01^{eB}
SDSB	2.5 ± 0.09^{hC}	2.5 ± 0.00^{gC}	2.5 ± 0.04^{hC}	2.48 ± 0.02^{hB}	2.4 ± 0.00^{fA}	2.49 ± 0.16^{hB}
Fresh	2.34 ± 0.07^{fE}	2.17 ± 0.04^{cD}	2.4 ± 0.00^{gF}	0.78 ± 0.00^{eB}	1.11 ± 0.23^{aC}	0.12 ± 0.00^{aA}
FDUU	0.02 ± 0.07^{aA}	1.91 ± 0.00^{aF}	0.88 ± 0.01^{aC}	0.98 ± 0.03^{fD}	1.38 ± 0.97^{bE}	0.29 ± 0.00^{bB}
FDUB	2.32 ± 0.04^{eD}	2.4 ± 0.21^{eE}	2 ± 0.11^{dC}	$0.08\pm0.00c^{A}$	2.4 ± 0.12^{fE}	1.98 ± 0.00^{fB}
FDSU	0.99 ± 0.00^{cB}	2.11 ± 0.10^{bF}	1.51 ± 0.01^{cD}	0.04 ± 0.09^{aA}	1.72 ± 0.00^{cE}	1.11 ± 0.01^{cC}
FDSB	2.44 ± 0.01^{eC}	2.5 ± 0.03^{gE}	2.49 ± 0.00^{hD}	0.09 ± 0.01^{cA}	2.48 ± 0.01^{eD}	2.29 ± 0.03^{gB}

Values are given as means IC_{50} (mg/ml) ± SD. Means with different superscript uppercase letters across the row and lower case letters within the column are significantly different ($P < 0.05$). IC_{50} values (the concentration which scavenges 50% of the DPPH radicals). SDUU-solar dried unsliced unblanched, SDUB-solar dried unsliced blanched, SDSU-solar dried sliced unblanched, SDSB-solar dried sliced blanched. FDUU-freeze-dried unsliced unblanched, FDUB-freeze-dried unsliced blanched, FDSU-freeze-dried sliced unblanched, FDSB-freeze-dried sliced blanched and fresh-fresh sample (no processing)

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