

Original Research Article
**BLANCHING AND JUICING EFFECT ON
 FLAVONOIDS CONTENTS IN COMMONLY
 CONSUMED LEAFY VEGETABLES IN SOUTH
 WEST NIGERIA**

ABSTRACT

Aims: This study investigated the flavonoids level, and effect of blanching and juicing on commonly consumed vegetables in South West Nigeria.

Place and Duration of Study: Chemical sciences, Redeemers University, January to March, 2013.

Methodology: Ten green leafy vegetables were obtained in four major markets in South West Nigeria. Each vegetable were thoroughly mixed and rinsed then divided into three groups, which are fresh, blanched and juice groups with four replicates per group. Flavonoids content were analyzed using standard laboratory methods.

Results: Blanched Hibiscus esculenta contain the highest flavonoids content. However, in most vegetables blanching and juicing present similar effects by increasing flavonoids content ($p < 0.05$) in Amaranthus spp, Crassocephalum ruben, Amaranthus viridis and Manihot esculenta and decreasing it in Colocasia esculenta and Corchorus olitorius when compared with their corresponding fresh vegetables except in Ipomoea batatas that was stable.

Conclusion: Flavonoids content in juice extract of Hibiscus esculenta was low while significantly higher ($p < 0.05$) in blanched. Though, both processing methods alter flavonoids content in similar manner, however, increase in flavonoids was more pronounced in blanching.

Keywords: processing, vegetable, blanching, juicing, antioxidant and flavonoid

1. INTRODUCTION

Nutrients and other phytochemical constituents in plants tissues have attracted alteration of scientists involved in food science and technology partly due to their antioxidants, anti-inflammatory, antimutagenic, anticancer, antibacterial and antiviral potential mediated by their enzymes irreversible binding and inhibitory capacities [1]. More so, considerable evidences have shown that adequate consumption of fruit and vegetable rich diets have

substantial role in reducing the risk of chronic diseases [2] including cardiovascular disease, ageing, age-related diseases as well as diabetes [3]. These plants based health benefits are mediated by the synthesis of certain secondary metabolites such as terpenoids, alkaloids and polyphenols [4]. The protective effect of flavonoids has been proven in-vitro and in-vivo [8]. They inhibit the oxidation of lipids and some enzymes activities including influence of the formation and transformation of peroxy radicals [9] as well as chelation of transition metal ions [10].

Evidences abound of the protective role of dietary flavonoids against coronary heart disease and central nervous system disorder like dementia [11]. Due to the antioxidants potentials of flavonoids, frequent as well as long term consumption of wine, beverages, foods, vegetables and other flavonoids rich plants products have served good antiosteoporotic, antithrombogenic, antiatherosclerotic supplements. More so, flavonoids protect against DNA free radicals, modulate inflammation as well as platelet aggregation [12]. However, it metabolites was not without complications in people. For instance, consumption of catechin generate such metabolite that binds tightly to erythrocyte membranes and form new antigenic sites which resulted in the development of autoantibodies that may cause hemolytic anemia [13]. Flavonoids have been reported to influence the metabolism of ascorbic acid [14, 15].

Vegetables present one of the most common and cheapest rich sources of flavonoids [16]. They however undergo varieties of processing methods which include sun-drying, boiling, freezing, salting, blanching and juicing. These processes could improve their palatability and bioavailability relative to phytochemicals and nutrients, and preservation as well as removing microbes and other toxins [17]. In spite of the ample documentation of various processing methods and their effects, there is dearth report as regards the content and effect of processing methods on flavonoids constituents of some local vegetables commonly consumed in South Western Nigeria. Therefore, this study investigated the flavonoids level, and effect of blanching and juicing on commonly consumed vegetables in South West Nigeria.

2. MATERIAL AND METHODS

Vegetables used for this research work were sourced from major markets in Ago-iwoye, Ikenne and sagamu, in Ogun state and Ketu in Lagos state, Nigeria. The weight of the samples ranged between 1 to 5kg; identified at the herbarium of the plant science and zoology department, OlabisiOnabanjo University.

2.1. Sample preparation

The vegetables were destalked to remove the inedible part; afterwards samples of each specimen (two from each market) were mixed together and divided into four. Each group was further grouped into three subgroups: Fresh, juiced and blanched.

2.1.1. Blanching

This process was done by putting 200g of vegetables in 500ml of boiled water and it was allowed to stay for five minutes. The vegetables were removed and then drained before analysis.

2.1.2. Juicing

This was done by using master chef juice extractor (model no: mc-J2101). The juice and pulp were collected separately and the pulp was discarded.

2.2. Sample analysis

2.2.1. Quantitative analysis of Flavonoids

Flavonoid in the test sample was determined by the acid hydrolysis of spectrophotometric method. 0.5 g of processed plant sample was mixed with 5 ml of dilute HCl and boiled for 30 min. The boiled extract was allowed to cool and filtered. 1 ml of the filtrate was added to 5 ml of ethyl acetate and 5 mls of 1% NH₃. This was then measured at wavelength between 420 - 520nm for the absorbance. (Harborne, 1976).

2.2.2. Moisture content

The moisture content of 10g of each sample was determined. This was done by taking 10g of each sample from each replicate (4 samples) into a 200ml crucible and then it was dried in oven at a temperature of 105oC for 24hour.

3. RESULTS AND DISCUSSION

3.1. RESULTS

Table 1: Flavonoids content of fresh leafy vegetable

Botanical names	English/ Local names	Flavonoids contents (mg/100g dry wt.)	Moisture contents
<i>Amaranthus spp.</i>	Joy weed/ Ebiden	112.03±15.47 ^{c,d}	85.20±0.41 ^{c,d}
<i>Crassocephalum ruben</i>	Ebolo	54.70±6.19 ^a	87.13±0.22 ^{d,e}
<i>Amaranthus viridis</i>	Amaranth / Tete	73.40±6.65 ^{a,b,c}	83.52±0.08 ^{a,b}
<i>Ipomoea batatas</i>	Sweet potatoes leaf / Ewe odukun	66.27±12.87 ^{a,b}	86.13±0.25 ^{c,d}
<i>Manihot esculenta</i>	Cassava leaf/ Ewe paki	81.16±6.80 ^{a,b,c}	85.08±1.33 ^c
<i>Piper guineensis</i>	Uziza	98.13±6.50 ^{b,c}	85.33±0.65 ^c
<i>Colocasia esculenta</i>	Cocoyam leaf/ ewe koko	86.56±5.03 ^{a,b,c}	85.52±0.58 ^c
<i>Corchorus olitorius</i>	Jute plant / Ewedu	138.57±30.75 ^d	82.98±0.60 ^c
<i>Basella rubra</i>	Amunututu	104.71±0.68 ^{b,c,d}	91.50±0.00 ^f

Hibiscus esculenta Okra leaf / Ewe ila 222.36±8.29^e 85.73±0.43s^e

Results presented are mean ± SEM (n = 4); values in the same column with the same superscript are not significantly different from each other (P > 0.05).

Table 2: Flavonoids content of blanched leafy vegetable

Botanical names	English/ Local names	Flavonoid contents (mg/100g dry wt.)	Moisture contents
<i>Amaranthus spp.</i>	Joy weed/ Ebiden	366.44±47.70 ^b	86.10±0.49 ^c
<i>Crassocephalum ruben</i>	Ebolo	505.04±22.40 ^c	89.00±0.08 ^d
<i>Amaranthus viridis</i>	Amaranth / Tete	870.13±115.19 ^d	87.65±1.76 ^d
<i>Ipomoea batatas</i>	Sweet potatoes leaf / Ewe odukun	61.17±8.92 ^a	85.93±0.43 ^{b,c}
<i>Manihot esculenta</i>	Cassava leaf/ Ewe paki	354.78±12.66 ^b	78.13±0.61 ^a
<i>Piper guineensis</i>	Uziza	58.63±5.47 ^a	87.78±0.35 ^{c,d}
<i>Colocasia esculenta</i>	Cocoyam leaf/ ewe koko	66.56±2.07 ^a	85.88±0.69 ^{b,c}
<i>Corchorus olitorius</i>	Jute plant / Ewedu	86.41±20.33 ^a	87.03±0.41 ^{c,d}
<i>Basella rubra</i>	Amunutu	36.36±11.28 ^a	83.35±1.58 ^b
<i>Hibiscus esculenta</i>	Okra leaf / Ewe ila	131.55±18.13 ^a	83.30±0.31 ^b

Results presented are mean ± SEM (n = 4); values in the same column with the same superscript are not significantly different from each other (P > 0.05).

Table 3: Flavonoids content of leafy vegetable juice extract.

Botanical names	English/ Local names	Tannin contents (mg/100g dry wt.)	Moisture contents
<i>Amaranthus spp.</i>	Joy weed/ Ebiden	163.47±15.12 ^{a,b}	95.23±0.46 ^{b,c,d}
<i>Crassocephalum ruben</i>	Ebolo	226.92±35.05 ^b	96.33±0.39 ^{c,d}
<i>Amaranthus viridis</i>	Amaranth / Tete	119.90±15.51 ^{a,b}	93.35±0.09 ^b
<i>Ipomoea batatas</i>	Sweet potatoes leaf / Ewe odukun	195.91±37.94 ^{a,b}	95.70±1.02 ^{c,d}
<i>Manihot esculenta</i>	Cassava leaf/ Ewe paki	216.32±68.43 ^b	95.20±1.34 ^{b,c,d}
<i>Piper guineensis</i>	Uziza	195.49±28.57 ^{a,b}	95.07±0.52 ^{b,c}
<i>Colocasia esculenta</i>	Cocoyam leaf/ ewe koko	56.75±5.07 ^a	86.68±0.70 ^a
<i>Corchorus olitorius</i>	Jute plant / Ewedu	113.07±12.95 ^{a,b}	93.50±0.79 ^{d,e}
<i>Basella rubra</i>	Amunutu funfun	254.13±24.74 ^b	97.35±0.19 ^{d,e}
<i>Hibiscus esculenta</i>	Okra leaf / Ewe ila	1050.10±113.03 ^c	98.93±0.05 ^e

Results presented are mean ± SEM (n = 4); values in the same column with the same superscript are not significantly different from each other (P > 0.05).

104 **Table 4: Percentage difference of flavonoid from fresh leafy vegetable**

Botanical names	English/ Local names	Percentage difference in blanched (%)	Percentage difference in Juice (%)
<i>Amaranthus spp.</i>	Joy weed/ Ebiden	227.09	45.92
<i>Crassocephalumruben</i>	Ebolo	823.29	314.84
<i>Amaranthusviridis</i>	Amaranth / Tete	1085.46	63.35
<i>Ipomoea batatas</i>	Sweet potatoes leaf / Ewe odukun	(7.70)	195.62
<i>Manihotesculenta</i>	Cassava leaf/ Ewe paki	337.14	166.54
<i>Piper guineesis</i>	Uziza	(40.25)	99.22
<i>Colocasiaesculenta</i>	Cocoyam leaf/ ewe koko	(23.11)	(34.44)
<i>Corchorusoliterus</i>	Jute plant / Ewedu	(37.64)	(18.40)
<i>Baselarubra</i>	Amunututu	(65.28)	142.70
<i>Hibiscus esculenta</i>	Okra leaf / Ewe ila	(40.84)	372.25

105 Values in brackets are negatives (-)

106 Table 1 shows flavonoids content in fresh vegetables, highest flavonoids content was
 107 observed in *Hibiscus esculenta* (222.36±8.29mg/100g dry weight) and its significantly higher
 108 (p<0.05) than other vegetables. The decreased order of flavonoids content was observed
 109 from *Hibiscus esculenta*, *Corchorusoliterus*, *Amaranthus spp.*, *Baselarubra*, *Piper guineesis*,
 110 *Colocasiaesculenta*, *Manihotesculenta*, *Amaranthusviridis*, *Ipomoeabatatasto*
 111 *Crassocephalumruben*. No significant (p>0.05) difference was observed between
 112 *Corchorusoliterus*, *Amaranthus spp.*, *Baselarubra*. Also, no significant difference (p>0.05)
 113 was noticed among *Piper guineesis*, *Colocasiaesculenta*, *Manihotesculenta*,
 114 *Amaranthusviridis*, *Ipomoea batatas*, *Crassocephalumruben* and *Crassocephalumruben*.

115 Shown in table 2 are flavonoids content in blanched vegetables; *Amaranthusviridis*
 116 (870.13±115.19mg/100g dry weight) contains the highest flavonoids and it is significantly
 117 (p<0.05) higher than others. A decreasing order of flavonoids content was observed from
 118 *Amaranthusviridis*, *Crassocephalumruben*, *Amaranthus spp.*, *Manihotesculenta*, *Hibiscus*
 119 *esculenta*, *Corchorusoliterus*, *Colocasiaesculenta*, *Ipomoea batatas*, *Piper guineesis* and
 120 *Baselarubra*. Though, *Crassocephalumrubenis* significantly lower than *Amaranthusviridis* but
 121 it is significantly higher than (p<0.05) others. No significant difference (p>0.05) was observed
 122 between *Amaranthus spp.* and *Manihotesculenta*. Also, no significant differences (p>0.05)
 123 were observed between *Hibiscus esculenta*, *Corchorusoliterus*, *Colocasiaesculenta*,
 124 *Ipomoea batatas*, *Piper guineesis* and *Baselarubra*.

125 Table 3 revealed flavonoids content in juice extracts of vegetables; *Hibiscus esculenta*
 126 contains the highest (1050.10±113.03 mg/100g dry wt). A decrease order was observed
 127 from *Hibiscus esculenta*, *Baselarubra*, *Crassocephalumruben*, *Manihotesculenta*, *Ipomoea*
 128 *batatas*, *Piper guineesis*, *Amaranthus spp.*, *Amaranthusviridis*, *Corchorusoliterus* and

129 *Colocasia esculenta*. Though, no significant ($p>0.05$) difference was observed among the
130 vegetables except *Hibiscus esculenta* which was significantly ($p<0.05$) higher.

131

132 3.1. DISCUSSION

133 Distribution of flavonoids in plants kingdom as well as plants tissues differs. Though, the
134 structure and the concentration of flavonoids have been strongly associated with varying
135 factors ranging from biotic factors like genetics and predation to abiotic factors such as
136 temperature, pollution, soil factors and weather condition [4]. The syntheses of flavonoids in
137 plants are primarily meant for protection against ultraviolet radiation [4], self-defense against
138 predation and parasitism [18] as well as repair of damaged tissues [4]. Antioxidant capacity
139 of flavonoids in fruits and vegetables may be influenced by these aforementioned factors.
140 During hormonal biosynthesis, flavonoids apparently modulate colour and palatability of
141 several plant tissues [19]. This present study revealed a range of concentration of flavonoids
142 in fresh, blanched and juice extracts of green leafy vegetables commonly consumed in
143 south west Nigeria. The flavonoids content in fresh vegetables were observed to range
144 between 1050.10 ± 113.03 mg/100g dry weight in *Hibiscus esculenta* and 56.75 ± 5.07
145 mg/100g dry weight in *Colocasia esculenta*. Variation in fresh vegetable has been linked to
146 factors like genetic variation [20]. The protective ability of flavonoids against organic toxin
147 from microbes [21] and inorganic toxic agents from the environment is a key factor in the
148 adaptive features of plants. Flavonoids achieves this task by forming complexes with other
149 molecules and ions such as free radicals and other oxidizing intermediates, chelation of iron
150 or copper ions and inhibition of certain proteins including enzymes like oxidases [22].

151 Blanching, a brief exposure of vegetables to high temperature alters the constituents
152 of plants tissues [16]. This includes substances such as oxalic acid which increased in sweet
153 potatoes but decreased in cabbage [23]. It may also destroy other contaminants [23]. From
154 the reports obtained, blanched *Amaranthus viridis* contained highest flavonoids content while
155 lowest (870.13 ± 115.19 mg/ 100g dry weight), as observed in tables 2, while the lowest was
156 recorded in *Basella rubra* (36.36 ± 11.28 mg/ 100g dry weight). A significant increase in
157 flavonoids content was observed in four of the vegetables while others displayed decrease
158 ($p<0.05$) when compared with the fresh of the corresponding vegetables except *Ipomoea*
159 *batatas* which showed no significant ($p<0.05$) change. Though flavonoids are altered by
160 change in temperature but the thermal effect varies with respect to the structure.

161 The varying effect of blanching process observed in our results was similar to that
162 observed by Ganiyu (2005) where blanching significantly increased total phenol content in
163 green leafy vegetables except in *Amaranthus cruentus* and *Vernonia amygdalina* [24].
164 Quercetin and kaempferol contents of strawberry were reported to be stable at temperature
165 up to 90°C while naringin, rutin, quercetin and naringenin contents in grapefruit reduced [25,
166 4]. But, temperature of about 70°C allows substantial increase in flavonoid content in apple
167 [26]. In addition, roasting at 130°C for 33 minutes increased phenol content of cashew and
168 peanuts [27]. Though, there are different factors that can influence thermal effects on
169 flavonoids contents in plants tissues but the structure of flavonoids seemed to play a prime
170 role [4].

171 Juicing, mechanically concentrates plants tissues constituents such as vitamins,
172 minerals and other phytochemicals. It also improves bioavailability of these constituents. In
173 this study, juicing increased flavonoids contents in all the vegetables except in
174 *Colocasia esculenta* and *Basella rubra*. Study has established that cutting which is a

mechanical force similar to juicing increased flavonol content in fresh cut-potatoes [28]. This could be that plants tissues tend to re-organize due to mechanical force exerted during juicing as injury thereby producing more flavonoids [28].

Therefore, blanching and juicing alter the constituent and concentrations of flavonoids in green leafy vegetables in similar manner either by increasing the flavonoids content as seen in *Amaranthus* spp, *Crassocephalum ruben*, *Amaranthus viridis* and *Manihot esculenta* or decreasing as observed in *Colocasia esculenta* and *Corchorus oliterus*. These two processing methods may also affect the flavonoids content of certain green leafy vegetable in an inconsistent manner. This was observed in *Hibiscus esculenta*, *Basella rubra*, *Piper guineensis* and *Ipomoea batatas*. The possible reason for the increase in flavonoids' content in juice extracts of vegetables could be in part due to the release of certain flavonoids caused by mechanical force exerted during the process [28] and concentration of such. Hydrogen bonding contributes to the bonds exercised by flavonoids to produce complexes with other molecules which are susceptible to thermal and mechanical effects respectively. Furthermore, increase in temperature is inversely proportional to the availability of oxygen thereby increasing free flavonoids such as quercetin and rutin [29].

4. CONCLUSION

Flavonoids are valued for their pharmacological activities and nutritional benefits in both humans and other animals. The anti-inflammatory, antithrombotic and iron binding properties of flavonoids cannot be overemphasized. The two processing methods employed in this study revealed similar effect on almost all the vegetables. Although, it has been established that concentration of flavonoids is not absolutely dependent on its antioxidant properties [30] however, temperature initiates a synergy between flavonoids concentration and its antioxidant activities. Therefore, blanching could increase antioxidant properties, in the process as well increasing anti-nutrient components which in turn could lead to health complications such as anemia, poor wound healing and poor fetal developments in subjects susceptible to such effect. In this study flavonoids contents in majority of juice extracts increased compared with the fresh. Though, fewer vegetables in the blanched increased in flavonoids contents however these few with increased flavonoids content was much pronounced. Therefore, it is suggested based on this findings that juicing and balancing help in increasing flavonoids (a beneficial antioxidant) contents in vegetables, however much caution must be exercised in the intake of such juiced or balanced vegetable because of the concentrating effect of this processing methods which may affect anti-nutrients alike.

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