Comparative Antioxidant Activity of Processed Mango (*Mangifera indica*) and Bush Mango (*Irvingia gabonensis*) Kernels

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**ABSTRACT**
Antioxidant activity of processed mango (*Mangifera indica*) kernel (PMK), processed Bush Mango (*Irvingia gabonensis*) kernel (PBMK) and the mixture of both (MKK) at 50:50 were evaluated using 2, 2-diphenyl -1-picryl hydrazyl (DPPH). The median inhibitory concentrations (IC50) of the three samples showed better correlation with that of reference Quercetin than the reference Vitamin C used. The three samples showed relatively higher radical scavenging effect than the reference samples. The result was significant at p < (0.05), indicative of high level flavonoid and phenolic acid content.

**Keywords:** Flavonoid, phenolic acid, DPPH, antioxidant activity.

**Introduction**
Mango (*Mangifera indica*) plant, a member of Anacardiaceae family yields one of the most celebrated fruits in tropical countries. The tasty fleshy mesocarp of the ripe fruits are eaten raw and manufactured into juice and jellies (Vandrendriessche, 1976). For several decades, the stony endocarp which comprises the shell, testa and kernel have been discarded as waste in Africa, whereas in Asia, interest has mainly been on the lipid component of the kernel because of its potential application in the confectionery industry as a source of cocoa-butter substitute (Rukmini and Vijayaghavan, 1984).

Further studies successfully showed that the kernel was convertible to edible state by processing. These studies bordered on composition, toxicology, and functionality of the kernel before and after processing into flour (Arogba, 1997), the browning activity of its polyphenol oxidase (Arogba et al., 1998), characterization of the tannic constituents (Arogba, 2000a), employing the processed flour in biscuit-formulation (Arogba, 1999), characterization of the biscuit with respect to effect of temperature on moisture adsorption behaviour (Arogba, 2000b), quality/shelf-characteristics of the biscuit (Arogba, 2002), and diversification of the processed flour in preparing some traditional Nigerian dishes (Arogba, 2004).

*Irvingia gabonensis* (Bush Mango) belongs to the family Irvingiaceae. The Irvingia species exist in two varieties: *Irvingia gabonensis* and *Irvingia wombolu*. These species are reported to be gregarious and largely distributed in Africa.

This plant with edible fruits (*I. gabonensis*) is largely used in traditional and modern medicine for the treatment of several illnesses, as well as in industry (Ude et al., 2006). Methanolic extracts of *I. gabonensis* are used in the treatment of bacterial and fungal infections. However, the kernels are used as soup thickener and the juice in wine production which
is attributed to attenuating obesity in relation to treatment of type-II diabetes (Victor et al., 2007).

Raw mango kernel is astringent and the report of Vaghasiya and Chanda (2010) stated that methanolic extract of the seed could be a source of natural antioxidants. However, the earlier work of Arogba (1997) and his subsequent studies indicated above, are yet to assess the effect of the processing methodology employed on radical scavenging potential of the residual processed mango kernel. Furthermore, a similar comparative study between the conventional mango kernel and those growing in the wild as Bush mango (I. gabonensis) has not been undertaken. Therefore, the present study reports on the comparative antioxidant properties of the processed mango kernel (PMK), processed Bush mango kernel (PBMK), and the mixture at 50: 50 % (MKK). The testa on the BMK was left intact in this experiment.

**Materials and Method**

**Materials collection and handling**

Freshly discarded mango seeds of Ikanekpo variety were collected at Anyigba town (Kogi State, Nigeria) and Bush Mango was purchased from Anyigba market in April 2010.

The methodology of Arogba (1997) was replicated to obtain PMK and PBMK samples in this study. The PMK and PBMK were each milled into powdery form. Each composite flour or equal mixture (50: 50) was used in experiments reported below. The testa on the BMK was left intact in this study.

**Reagents**

The main reagents included 2, 2-diphenyl-1-picrylhydrazyl (DPPH) (SIGMA, Germany), vitamin C (Puritan’s Pride Inc., Oakdale, USA), sodium metabisulphite, potassium dichromate, quercetin (SIGMA, Germany), methanol, sodium chloride.

**Preparation of sample extracts**

One gram (1g) of sample was weighed into a beaker containing 50 ml of methanol and placed on orbital shaker for 2 h for shaking. After shaking, the supernatant was carefully decanted to form the stock solution from which serial dilutions were made to obtain other required concentrations.

**Determination of DPPH radical scavenging activity**

The hydrogen or radical scavenging property of the extracts was determined using the stable radical DPPH (2, 2-diphenyl-1-picrrlhydrazyl hydrate) according to the method of Blois (1958) and as described by Brace et al. (2005). When DPPH reacts with an antioxidant compound which can donate hydrogen, it is reduced,

\[ \text{DPPH}^* + \text{RH} \rightarrow \text{DPPH}_2 + \text{R}^* \]

The change in colour from deep violet to light yellow was measured at 517 nm in a UNICAM spectrophotometer.

**Procedure:** To one ml of different concentrations (500, 250, 125, 62.5, 31.25 µg/ml) of the extract or reference was added one ml of 0.3 mM DPPH in methanol. The mixture was vortexed and then incubated in a dark chamber for 30 min after which the absorbance was measured at 517 nm against a DPPH control containing only one ml of methanol in place of the extract. The antioxidant activity (AA) was then calculated using the formula:

\[ \frac{(\text{Ao} - \text{Ac})}{\text{Ao}} \times 100 \]

Where: \( \text{Ao} = \text{Absorbance without extract;} \) \( \text{Ac} = \text{Absorbance with extract} \)

**Statistical analysis**

The results are expressed as mean ± SEM using Graph Pad Prism Graphical-Statistical Package version 5. The difference between groups was analyzed by one-way analysis of variance (ANOVA) followed by Dennett’s test at 5% level of significance \((p < 0.05)\).

**Results**

The drying process to attain constant weights of the samples was achieved within 24 h at 105°C using the conventional air-driven oven and the dry matter content was determined to be 95.2 ± 0.08 %,
Comparative Antioxidant Activity of Processed Mango (Mangifera indica) and Bush Mango (Mangifera indica) and the combination (MKK) respectively.

In separate experiments conducted in our laboratory (unpublished), results showed that the total phenolic content in PMK, PBMK and MKK were 1.67 ± 0.13, 1.15 ± 0.03 and 2.19 ± 0.08 mg GAE/g dry wt respectively with the mixture showing the highest phenolic content. Similarly, the respective total flavonoids content were 1.64 ± 1.02, 0.77 ± 0.23, 1.38 ± 0.18 mg QUE/g dry wt. Notably, PBMK had more free phenolic than flavonoid content.

In the DPPH assay the activity was concentration dependent, i.e. activity increased with increase in concentration. PMK showed the highest activity with the least IC50 value (143.36 ± 0.42 µg/ml), but the mixture showed the highest percentage inhibition of 88.73 ± 0.17 %; statistically their percentage inhibition and IC50 were not significantly different (p > 0.05) in contrast to PBMK with an IC50 value of 177.22 ± 1.05 µg/ml. Vitamin C gave IC50 of 300.22 ± 0.61µg/ml, and Quercetin 184.71 ± 2.5 µg/ml. The samples generally showed more activity compared with the reference samples.

**Discussion**

In recent times, many epidemiological studies have confirmed that intake of exogenous antioxidants is effective in the prevention of a number of human diseases which have been implicated to be due to oxidative stress (Valentao et al., 2002). Consequently, the necessity for the investigation and evaluation of safer antioxidants from natural sources has become imperative. Antioxidants isolated from the plant source have proved to be safer even in chronic administration (Adaramoye et al., 2008). Fruits, vegetables and oil seeds have increasingly been recognized as sources for phenolic antioxidants (Shahidi, 1997, Nack and Sahidi, 1989). Anecdotal evidence suggests that PMK and PBMK are useful in this regard.

The processing of fresh PMK and PBMK into flour gave dry weight matter content values comparable with that reported from similar processing by Arogba (1997). During processing, Arogba (1997) had indicated water solubility of reducing substances such as polyphenols that impart astringent property to the kernels. Thus, the processed (residual) flour was a useful principal ingredient for confectioneries (Arogba, 1999, 2002). The order of antioxidant activity based on IC50s, was MKK > PMK >PBMK (Table 1) (p < 0.05). These results were similar to those of methanol extracts of some medicinal plants (Miliauskas et al., 2004), and the results of Vaghasiya and Chanda (2010) which concluded that *M. indica* kernel flour can be used to provide natural antioxidants and antimicrobials.

Anthocyanin, proanthocyanidine, phenol and flavonoids possess hydroxyl groups which are responsible for free radical scavenging effect. It has been known that flavonoids possess antioxidant activity and, hence, considered as having positive effects on human health and nutrition. Furthermore, flavonoids are antiinflammatory, antitumor, antiviral

### Table 1: Antioxidant activity using DPPH antiradical assay

<table>
<thead>
<tr>
<th>Concentration µg/ml</th>
<th>PMK (%)</th>
<th>PBMK (%)</th>
<th>MKK (%)</th>
<th>Vitamin C (%)</th>
<th>Quercetin (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>500</td>
<td>82.46 ± 0.32</td>
<td>77.7 ± 1.31</td>
<td>88.73 ± 0.17</td>
<td>64.59 ± 0.25</td>
<td>81.17 ± 3.99</td>
</tr>
<tr>
<td>250</td>
<td>68.10 ± 0.45</td>
<td>60.43 ± 0.23</td>
<td>71.80 ± 0.11</td>
<td>51.50 ± 0.78</td>
<td>72.05 ± 2.56</td>
</tr>
<tr>
<td>125</td>
<td>53.06 ± 0.23</td>
<td>54.67 ± 1.24</td>
<td>58.97 ± 0.41</td>
<td>41.77 ± 0.56</td>
<td>61.34 ± 1.44</td>
</tr>
<tr>
<td>62.5</td>
<td>41.46 ± 0.89</td>
<td>48.43 ± 0.12</td>
<td>43.13 ± 0.28</td>
<td>28.82 ± 0.46</td>
<td>29.55 ± 0.98</td>
</tr>
<tr>
<td>31.25</td>
<td>28.50 ± 0.21</td>
<td>20.60 ± 2.35</td>
<td>17.27 ± 1.19</td>
<td>16.83 ± 1.01</td>
<td>7.38 ± 3.55</td>
</tr>
<tr>
<td>IC50</td>
<td>143.36 ± 0.42</td>
<td>177.22 ± 1.05</td>
<td>145.69 ± 0.43</td>
<td>300.22 ± 0.61</td>
<td>184.71 ± 2.50</td>
</tr>
</tbody>
</table>

%: percentage inhibition. All values are expressed as mean ± SEM (n=3)
and antiplatelets (Shahidi and Wanasundara, 1992). The activity shown from the findings of this study shows that they may serve as potential agents of the above listed health benefits.

Phenol acts as free radical chain reaction terminator, thereby acting as antioxidant. Phenol also has a potential of combating oxidative stress, a syndrome causative of some neurodegenerative diseases and cardiovascular diseases. Phenolic compounds comprise a large group of biological active substances. Quercetin, catechin, ferrulic acid, caffeic acid, gallic acid, coumaric acid and rutin are among the most common naturally occurring antioxidant phenolic compounds in foods. Other natural antioxidants include glutathione peroxidase, superoxide dismutase, tocopherols, and vitamin C, and carotenoids (Duvivier et al., 2010). The level of antioxidants as evidenced in this study imply the presence of phenolics.

The mechanism of action of flavonoids are through scavenging or chelating process (Pourmorad et al., 2006); this could attest to the high level of scavenging activity expressed by the mixture and the other two samples (p < 0.05). The results of the IC50 values obtained further implies that PMK, PBMK and MKK showed high antioxidant activity when compared to Vitamin C and Quercetin used as reference antioxidant, and had more phenolic substances.

Conclusion
The study showed that PBMK when potentiated with eqv. concentrations of >125 µg/ml PMK, had a level of AA activity similar to PMK alone, and even with statistically higher (p < 0.05) activity than some other known naturally-occurring phenolic antioxidants.

References


