EVALUATION OF PHYTOCHEMICAL CONTENT OF SUN-DRIED AND OVEN-DRIED RIPE PULPS OF Cucurbita pepo L. CULTIVARS AND Citrullus lanatus THUNB

C.D.U. Nwokwu1, A.I. Ukoha2 and C. Imo3
1Department of Biochemistry, Federal University Wukari, Taraba State, Nigeria
2Department of Biochemistry, Abia State University, Uturu, Abia State, Nigeria
*Corresponding author: danielnwokwu@fuwukari.edu.ng; +234-8033189185.

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Abstract: This study investigated the fundamental scientific basis for the use of three common cucurbits (two cultivars of Cucurbita pepo (smooth-rind and coarse-rind varieties) and Citrullus lanatus) by screening and quantifying the percentage of selected phytochemical constituents in the sun-dried and oven-dried fruit pulps. Qualitative analysis revealed the presence of all bioactive constituents in both dried forms of the three fruit samples, but tannins were not detected. Quantitative estimation (%) of the sun-dried pulps [alkaloids (3.84±0.23; 2.83±0.71; 1.97±0.05); saponins (1.24±0.09; 1.58±0.21; 0.6±0.07); and flavonoids (13.60±0.57; 12.78±0.53; 16.75±1.06)] of smooth C. pepo, coarse C. pepo and C. lanatus, respectively, showed a significant (p<0.05) reductive trend upon thermal application [alkaloids (1.69±0.02; 1.47±0.05; 1.15±0.07); saponins (0.84±0.05; 0.98±0.07; 0.37±0.09)] and flavonoids (9.25±1.06; 8.48±1.24; 11.98±0.53), but differences were not significant for saponins (p>0.05). The findings confirm that the three cucurbits are very good sources of thermolabile phytochemicals, with inherent nutritional, medicinal and chemoprotective prospects, thus, propelling pumpkin as a cheaper, and even more beneficial alternative to be massively grown in Nigeria’s Southern tropical forests.

Keywords: Nigeria, oven-dried, phytochemicals, pulp, pumpkin, sun-dried, watermelon

Introduction
Many cucurbits have been shown to contain many bioactive chemical substances that produce definite physiological and biochemical actions in the human body (Trease and Evans, 1989; Ryan and Shattuck, 1994; Hagerman, 2002; Okwu and Omodamiro, 2005). These phytochemical constituents include alkaloids (Trease and Evans, 1989), tannins (Oboh and Akindahunsi, 2003), flavonoids (Barakat et al., 1993), saponins (Hostettmann and Marston, 1995; Sridhar and Bhat, 2007), etc. The medicinal values of plants are dictated by their phytochemical and chemical constituents, prompting the elucidation of their levels in many plants (Cho et al., 2004; Edeoga et al., 2005). The specific compounds present in plants of the Cucurbitaceae family are in the process of being identified (Jang et al., 2001; Kim et al., 2012) and used in studies for alleviation of chronic diseases such as diabetes, cardiovascular events, and some forms of cancer.

Cucurbitaceae is a large family of flowering plants containing about 120 genera and approximately 825 species typically distributed in the tropical climate of Africa and Southeast Asia and poorly represented in temperate regions (Mabberley, 1987; USDA, 2007; Koocheki et al., 2007). In Nigeria, Citrullus lanatus (watermelon) and Cucurbita pepo (pumpkin) are the common cucurbits cultivated in the northern and southern regions, respectively. Pumpkin is called Kabewa, Edelele and Kabewa in the south-east, south-west and northern regions of Nigeria, respectively; whereas water melon remains anglicised, but with slight corruptions in the various vernacular parlances. The oval-shaped C. pepo has two commonly cultivated varieties in Nigeria’s Eastern region – smooth-rind and coarse-rind varieties – presumably a consequence of environmental adaptation, or more remotely, speciation. Many cucurbits have historically been used in both nutrition and medicine. Some are important sources of food, like pumpkins, melon, cucumber, water melon, etc (Kerje and Grum, 2003), while others like the bottle gourd (Lagenariasiceraria) serve non-culinary purposes, basically storage (Dutta, 1964; Burkil, 1985). For instance, C. lanatus is mildly diuretic because of its very high water content (87–92%), has no fat or cholesterol, is an excellent source of vitamins A, B, and C, and contains fiber, potassium, lycopene. Beside its juicy texture, it is rich in useful antioxidants (mainly lycopene) which have been demonstrated to inhibit growth of cancer cells (Perkins-Veazie and Collins, 2004). Watermelon is also a rich source of citrulline (where from it derives its scientific name), an amino acid that can be metabolized to arginine, a conditionally essential amino acid for humans. Arginine is the nitrogenous substrate used in the synthesis of nitric oxide and plays an essential role in cardiovascular and immune functions (Collins et al., 2005). C. lanatus (watermelon), while being a rich reservoir of these phytochemicals, is either too expensive or not easily accessible to majorities of the southern populace in Nigeria because of non-supportive climatic and soil conditions (Atuonwu and Akobundu, 2010).

The seeds of C. pepo are becoming popular for its medicinal properties such as anti-diabetic (Quanhong et al., 2003), anti-fungal (Wang and Ng, 2003), anti-bacterial and anti-inflammatory activities (Caili et al., 2006) and anti-oxidant effects (Nkosi et al., 2006), including expulsion of intestinal worms and parasites and the prevention of kidney stones. Previous proximate analyses of its seeds and flowers have identified its richness in of triterpenoids and carotenoids, protein, carbohydrate and vitamins (USDA, 2002; Sotelo et al., 2007), making the plant a pharmaceutical and nutriceutical candidate. Consumption of pumpkin seeds is common in South-eastern Nigeria without reported adverse effects. The seeds of several species of pumpkin have been used in traditional medicine for centuries. In Africa, the pulp is used as a poultice to treat burns and inflammations and as a cooling compress to treat headache and neuralgia; it has also been applied to tumours and corns (hyperkeratosis). The fruit
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pulp has been used to dehair and soften hides in tanning industries (Caili et al., 2006). Due to the seeming restrictive ecological distribution of the water melon to northern Nigeria, its cheap availability to the ecologically non-supportive South has been a mirage. So the present study is designed to compare the phytoconstituents of the water melon and pumpkin with a view to advocating intensified cultivation of the pumpkin on its favourable habitat (southern region); thus, propelling pumpkin as a cheaper, and perhaps more beneficial alternative for rural dwellers in Nigeria’s South with its non-supportive environment for water melon cultivation. The essence of research work is subsequent translation to community development. To the best of our knowledge, there is limited information on this in existing literature. The present study is therefore aimed at filling this scientific gap by adapting its findings to the socio-economic needs of the rural populations that grow them. The results will be useful for proper awareness on cultivation and use of these fruits throughout the year. This will therefore lead to the amelioration of some nutritional deficiencies and the maintenance of population health in general.

Several researchers have reported on the reductive effects of pure processes on the mineral and phytochemical contents of plants studied (Dupont et al., 2000; Akubugwo et al., 2008). Since the fleshy pumpkin pulp is primarily consumed as a vegetable, either boiled, fried or stuffed, its phytochemical and nutritional content could possibly undergo thermal degradation. Thus, it becomes imperative to establish how the nutritional composition of these fruits can be influenced when subjected to the traditional processing method of cooking. Therefore, this study investigates the fundamental scientific basis for the use of these three common cucurbits (two cultivars of Cucurbita pepo (smooth-rind and coarse-rind varieties) and Citrullus lanatus) by screening and quantifying the percentage of their phytochemical constituents in their near-native (sun-dried) and oven-dried fruit pulps.

Materials and Methods

Materials

All chemical reagents were of analytical grade and provided by Sheda Science and Technology Complex (SHESTCO) in collaboration with their suppliers: Sigma Aldrich Chemical (St. Louis, MO, USA), British Drug House (Poole, Dorset, England) and May and Baker (Essex, United Kingdom), unless otherwise indicated. The benchwork was carried out at the SHESTCO laboratories, Sheda, Abuja, Nigeria, with unrestricted access to all required facilities.

Sample preparation

Two C. pepo cultivars (coarse-rind variety and smooth-rind variety) were procured from a local farmer at Umueze village, Okigwe in Imo State, Nigeria. After identification by a taxonomist in the department of Plant Science and Biotechnology, Abia State University, Uturu, the fruits were conveyed to the Chemistry Advanced Laboratory (CAL) of Sheda Science and Technology Complex (SHESTCO), Abuja. Apparently healthy fruits were selected for the analysis. C. lanatus (light green variety) was purchased at Gwagwalada market, Abuja, and channeled to the same laboratory. The water melon and pumpkin cultivars were washed with clean water, sliced into quarters, peeled and de-seeded. Each of the three cucurbit fruits was divided into two (2) portions. One portion was sun-dried for four (4) days, while another portion was oven-dried at 65°C to a constant weight. The dried fleshy mesocarps and endocarps (pulps) were pulverized into uniform powder using a Waring blender and kept in labeled plastic bags until phytochemical analysis. All samples in this study were analyzed in triplicates.

Phytochemical screening

Prior to quantitative phytochemical determinations, qualitative tests were carried out on the fruit powder samples using standard procedures to identify alkaloids and tannins (Trease and Evans, 1989), saponins (Sofowora, 1993) and flavonoids (Harborne, 1973).

Phytochemical estimation

Alkaloid content was quantified by the gravimetric method (Harborne, 1973); saponin, by combined solvent extraction (Obadoni and Ochuko, 2001) and flavonoids, as described by Boham and Kocipai (1994). Tannin was in trace amounts (where detectable) and so was not included in any quantitative determination.

Statistical analysis

Data were expressed as mean±standard deviation of three independent experiments. Statistical analyses were performed using GraphPad Prism 6.0.1 software (GraphPad Software Inc., San Diego, CA, USA) to determine the significant differences among groups. The differences were considered to be statistically significant at p≤0.05.

Results and Discussion

The phytochemical screening (Table 1) and quantitative estimations (Fig. 1) of the percentage yields of phytoconstituents in the three cucurbits studied showed that the pulps were rich in alkaloids, flavonoids and saponins. Tannin levels were not detected or were present in trace amounts in all representative fruit samples. Since ripe fruits were used in this study, this is concordant with the finding of Price et al. (1978) that tannin content ordinarily decreases with fruit maturation. However, Erukainure et al. (2010) reported a substantial tannin level in watermelon rinds. This is understandable as the rinds of most fruits possess a bitter taste unlike the mostly sweet and edible pulps.

[Values are the means of triplicate determinations on a dry weight basis ± standard deviation. *Flavonoid levels of all representative fruit samples) and alkaloid levels (of smooth C. pepo only) of the sun-dried and oven-dried pulps had significant differences (p<0.05)]

Fig. 1: Percentage phytochemical composition of sun-dried and oven-dried ripe pulps of Cucurbita pepo L. cultivars and Citrullus lanatus thunb
The results indicate appreciable levels of alkaloids and flavonoids. The very high concentrations of flavonoids are understandable in view of the high colour pigmentation (Noststrand, 1995) in the fruits; especially for C. lanatus (16.75%). These secondary metabolites have been associated with antimicrobial activities and numerous physiological activities in mammalian cells in various studies (Sofowora, 1993; Nweze et al., 2004). Flavonoids possess anti-inflammatory, anti-oxidant, anti-allergic, hepatoprotective, anti-thrombic, anti-viral and anti-carcinogenic activities (Sharma et al., 2009).

The sun-dried smooth-rind C. pepo contains more alkaloids (3.84%) in comparison to the coarse-rind cultivar (2.83%) and C. lanatus (1.97%). Alkaloids are beneficial chemicals to plants. They help in repelling predators and parasites. However, when ingested by animals, they affect glucagon, thyroid-stimulating hormone (TSH) and inhibit certain mammalian enzymatic activities (Okaka et al., 1992). All alkaloids, when taken in high doses, are also toxic to humans. However, at lower doses many are useful pharmacologically, e.g. morphine, codeine, nicotine, cocaine, etc. Pure isolated alkaloids and their synthetic derivatives are used as basic medicinal agents for their analgesic, antispasmodic and bactericidal effects (Kwu, 2004).

Saponins are in relatively meagre quantities than other phytochemicals determined in the fruits with the lowest level recorded for oven-dried C. lanatus (0.37%). The sun-dried coarse-rind C. pepo contains more saponin (1.58%) than the smooth-rind variety (1.24%). This may be why it tastes slightly bitterer. The bitter principle, saponin is a well-known anti-nutritional factor that can reduce the uptake of certain nutrients including cholesterol and glucose at the gut through intra-lumenal physicochemical interaction or other yet unidentified activity. This could confer chemo-protection against heart diseases to consumers because of its hypocholesterolemic activity (Price et al., 1987). It may also aid in lessening excess hepatic metabolic burden. There is evidence of the presence of saponins in traditional medicine preparations, with tremendous commercially driven promotion of saponins as nutriceuticals (Asl and Hosseinzadeh, 2008).

According to Rodriguez-Amaya (1993), the phytochemical and nutritional composition of fruits varies somewhat with type and degree of maturity; their ecological distribution and cultivation environments (Park et al., 1997) have also been implicated. Apparently, the variations noticed for the phytoconstituents between the two C. pepo cultivars could be due to speciation, state of maturity, degree of ripeness and/or post-harvest treatment (Lima et al., 2005), since they were both obtained from the same climatic and geographic sites.

### Table 1: Qualitative phytochemical screening of sun-dried and oven-dried ripe pulps of Cucurbita pepo L. cultivars and Citrullus lanatus Thunb

<table>
<thead>
<tr>
<th>Quantified Phytochemicals</th>
<th>Sun-dried Fruit Samples</th>
<th>Oven-dried Fruit Samples</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Pumpkin (smooth variety)</td>
<td>Pumpkin (coarse variety)</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>Tannins</td>
<td>±</td>
<td>±</td>
</tr>
<tr>
<td>Saponins</td>
<td>++</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>+++</td>
<td>+++</td>
</tr>
</tbody>
</table>

(+) = Slightly present; (+++) = Moderately present; (++++) = Heavily present; (±) = Not convincingly detected; (-) = Completely absent

### Table 2: Percentage losses of phytochemicals in the oven-dried fruits*

<table>
<thead>
<tr>
<th>Fruit Samples</th>
<th>Alkaloid (%)</th>
<th>Saponin (%)</th>
<th>Flavonoid (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pumpkin (smooth rind)</td>
<td>55.98</td>
<td>32.26</td>
<td>31.99</td>
</tr>
<tr>
<td>Pumpkin (coarse rind)</td>
<td>48.06</td>
<td>37.97</td>
<td>33.65</td>
</tr>
<tr>
<td>Water melon (light green)</td>
<td>41.62</td>
<td>40.32</td>
<td>47.52</td>
</tr>
</tbody>
</table>

*% Loss = \( \frac{\text{Conc. in Sun-dried sample} - \text{Conc. in Oven-dried sample}}{\text{Conc. of phytochemical in Sun-dried sample}} \times 100 \)

Processing causes some changes in nature and content of phytoconstituents as presented in Table 2. There were significant differences (p < 0.05) between the sun-dried and oven-dried fruits for flavonoids and alkaloids, but not for saponins, for each fruit category. According to Akubugwo et al. (2008), sun-drying is the preferred processing method to retain phytochemical contents. The choice of near-native (sun-dried) forms of the fruits reflects the desired objective of obtaining the approximate crude levels of these constituents vis-à-vis their concentrations in processed (cooked) forms. The average percentage losses of the respective phytochemicals in the oven-dried samples imply that the effect of heat was more pronounced on the alkaloid content (48.55%), followed by flavonoid (37.72%) and least of all, saponin (36.85%). The finding corroborates an earlier report by Akubugwo et al. (2008) that oven-drying drastically reduced phytochemical contents of plants. Elsewhere in literature, we had also demonstrated the degradative effects of boiling on the ascorbic acid content of the three cucurbit fruits under evaluation (Nwokwu et al., 2016). Fagbemi et al. (2005) showed that boiling is most effective in reducing the tannin content, which offers an explanation for the trace amounts obtained for tannin since all samples were subjected to some level of heat processing. Therefore, boiling pumpkin fruits prior to consumption enhances palatability and digestibility, and improves nutrition by destroying certain anti-nutritional factors. The percentage losses in phytochemical content of the ripe pulps of the cucurbits seem to have a more positive correlation with the intensity (60°C/ ~ 12 h for oven-drying) than duration (25 – 30°C/ ~ 96 h for the Abuja sun – Kottek et al. (2006)) of heat exposure, suggesting the employment of less harsh preparation and preservation methods. This also brings...
more research prospects into limelight: what level of processing is necessary, and what will be the effect of such processing on the identified constituents in the fruits? Rui (2003) established that the health benefits of fruits and vegetables are from additive and synergistic combinations of phytochemicals. Severe toxicity has not been reported with the use of Cucurbita extracts. However, cross-reactivity to watermelon, cucumber and pumpkin has occurred, but causality was not proven (De Queiroz-Neto, 1994; Figueredo et al., 2000). Given the outcome of this study, such adverse reaction(s) could have resulted from cumulative over dosage of the active ingredients identified in the fruits. So, balanced consumption of these fruits is strongly recommended. As a better alternative, the outcome of this research provides a sound scientific basis and incentives for intensification of pumpkin cultivation– and it presents as a better alternative, the outcome of this research strongly recommended.

In this study we have shown that ways of expanding the use of available local food sources are increasingly pursued, but knowledge of the nutritive value of such local foodstuffs in their native and processed forms is necessary in order to encourage their increased cultivation and consumption. Knowledge of the nutritive value is essential in supplementing staple carbohydrate foods. With the vast expanse of arable land in Nigeria, there is no gainsaying that intensified cultivation of these cucurbits will boost food security, improve nutrition and foster rural development and support sustainable land care.

Conclusion

In this study we have shown that C. pepo contained as much useful ingredients as C. lanatus and perhaps more, and that heating exerts reasonable reductive effects on the phytochemical content of the fruit pulps. To maximize the benefits from these cucurbits we recommend consumption in their raw forms, or where it is an essential prerequisite, subjected to minimal processing. Due to its ease of cultivation, potential productivity and, above all, its substantial phytochemical content, C. pepo can potentially offer nutritional, medicinal, chemoprotective and economic benefits to its growers and consumers.

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Conflict of Interest

The authors wish to declare that there are no conflicts of interest in this work.

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