Physico-chemical and Nutritional Characteristics, and Antimicrobial Activity of Oil Palm Syrup, Raffia Palm Syrup and Honey

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Abstract: The physico-chemical characteristics and antimicrobial activity of oil palm syrup, raffia palm syrup and honey were studied. The materials contained mainly carbohydrate (64.76-68.79%) and water (28.05-31.50). They exhibited similar densities (1.23-1.26 g cm$^{-3}$) and pH (3.51-4.18), and had low ash (0.30-0.50%), protein (0.24-1.04%) and lipid (2.20-3.62%) content. They had modest content of Fe (2.35-3.30 mg/100g), Ca (37.06-79.05 mg/100g), and phenolic compounds (125.93-185.44 mg GAE/100 g), and were rich in potassium (325.12-628.56 mg/100g). They contained non-enzymatic browning products (browning intensity was 0.71 for honey, 0.159 for raffia palm syrup and 0.175 for oil palm syrup). The materials exhibited antimicrobial activity against clinical strains of Pseudomonas aeruginosa, Bacillus cereus, Escherichia coli and Staphylococcus aureus. The antimicrobial activities of dilute solutions (0.1 ml, 0.5% aqueous solutions) of the honey (containing 360 µg dry matter) and syrups (raffia, 360 µg and oil palm, 340 µg dry matter) were similar to that of 10 µg of the antibiotic streptomycin. This indicates that the materials had much lower antibiotic activity (against the test organisms) than streptomycin. The results suggest that in addition to their good mineral and phytochemicals content, the palm syrups could be useful (like honey) as an antimicrobial substance for food preservation and medicinal applications.

Key words: Honey, palm syrups, physical and nutritional characteristics, phytochemicals, antimicrobial activity.

I. Introduction

Honey is an edible sweet substance made by bees using nectar from flowers. Apart from its use as a sweetener, several applications of the material indicate that it also functions as a food preservative. It has been demonstrated that honey serves as a source of natural antioxidants, which are effective in preventing deteriorative oxidative reactions in foods, such as inhibiting browning reactions in fruits and vegetables [1], [2], [3] and preventing lipid oxidation in cooked, ground poultry [4], [5], [6]. Honey has been found effective for increasing serum antioxidant capacity [7] and total plasma reducing capacity [8] in humans. It also exhibits antimicrobial activity due to osmolarity (of concentrated solutions) and the presence of phytochemicals [9], and is used for wound dressing [10].

Palm sap is the white, semi-translucent, sugary sap obtained by tapping the stalk of the immature inflorescent of palm trees, the upper stem, or by tapping the felled trees. The principal sap bearing palms are Borassus spp, Phoenix sylvestris, Phoenix dactylifera (the date palm), Caryota urens, Arenga pinnata, Juboea spectabilis (the Chilean molasses palm), Raphia spp (the raffia palms), Cocos nucifera (the coconut palm), and Elaeis guineensis (the African oil palm) [11], [12], [13], [14]. In the tropics, palm sap is drunk as such (fresh, or pasteurised and bottled), evaporated for palm syrup and sugar, or fermented to alcohol and vinegar. Palm sap is also a source of yeast for bread making. For palm sugar manufacture, fermentation of the sap during collection must be prevented by cooling to low temperature and/or the addition of preservative. The juice is filtered and concentrated in pans on open fires and then filled into suitable containers, where after cooling it hardens to a solid golden brown product with a pleasant taste and smell [13], [15], [16]. The more sophisticated method of sugar manufacture such as vacuum pans and centrifuges are used for manufacture on a larger scale.

In India, a considerable palm sugar industry is based on palmyra (Borassus flabellifer) sap [13] and in Southeast Asia, the syrup is widely used in cooking; its sweetness tempers the flavours of spicy curries, adding its rich, molasses-like flavour to the food [15]. In Nigeria however, sap from the oil palm (Elaeis guineensis) and raffia palms (Raphia spp.) is fermented and drunk as such (fresh, or pasteurised and bottled), or distilled to produce a kind of spirit (gin), which contains mainly ethanol, but also has considerable amounts of methanol and other toxic constituents; consumption of the gin poses a health risk. The use of palm sap for syrup and sugar production could improve income from sap production and utilisation, and discourage its conversion to unwholesome products.
In this study, syrups were prepared from oil palm sap and raffia palm sap. Their physico-chemical properties and antimicrobial activity were compared with those of a commercial honey sample, to find out if they could substitute for honey as a nutritious food additive and antimicrobial agent for food preservation and medicinal application.

II. Materials and methods

2.1. Materials

2.1.1. Palm sap
Oil palm sap and raffia palm sap were obtained from the palm wine bottling unit of the Nigerian Institute for Oil Palm Research near Benin City, Nigeria.

2.1.2. Honey
Honey was obtained from the Benedictine Monastery at Ewu, Edo State, Nigeria.

2.1.3. Preparation of syrups
The syrups were prepared as follows: Oil palm sap or raffia sap (1.5 L) was filtered through muslin cloth and the filtrate was heated in an open pan at a temperature of 100.0°C until it turned viscous and brown; the syrup was cooled and its volume was measured. One and a half litres (1.5 l) of the sap gave 150.0 ml of syrup.

2.1.4. Test organisms
Pure cultures of Pseudomonas aeruginosa and Bacillus cereus were obtained from the Medical Microbiology Unit of the University of Benin Teaching Hospital, Benin City, Nigeria. Escherichia coli and Staphylococcus aureus were obtained from the Microbiology Unit of Benson Idahosa University, Benin City, Nigeria.

2.2. Methods

2.2.1. Physico-chemical characteristics
Nitrogen was determined using the Kjeldahl method [1]. Protein was calculated as: % Protein = total N x 6.5. Calcium was determined by titration with EDTA, potassium by flame photometry, and iron by atomic absorption spectrophotometry [18], [19]. Ash content was determined by ashing in a furnace at 500°C for 24 hr. The ash was left to cool and then weighed. Moisture content was determined by drying to constant weight in a ventilated oven at 50°C. Samples were dissolved in distilled and deionised water and their pH was read. Density of honey and syrups was determined by the use of specific gravity bottles. Determination of total lipid content was by the method of Bligh and Dyer [20].

Total phenolic content (TPC) was determined spectrophotometrically by a modification of the procedure described by Azizah et al [21]. Samples and gallic acid standards, 50, 100, 150 to 500 mg/l (0.1 ml, in duplicate) were mixed with 0.5ml of 0.2N Folin-Ciocalteu reagent and after 8min, 1.5 ml of 7.5% (w/v) sodium carbonate was added. The mixture was kept in the dark for 1 hour and the absorbance was measured at 765 nm. TPC was computed from the gallic acid standard curve and expressed as mg of gallic acid equivalent (GAE)/100 ml of sample. Browning intensity of palm syrups and honey was measured as the absorbance of a 1% solution of honey, raffia palm syrup or oil palm syrup at 420 nm [22], [23].

Palm syrups and honey were screened for the presence of alkaloids, flavonoids, glycosides, saponins, terpenoids and tannins according to Evans [24].

2.2.2. Microbiological Analysis
The pure cultures of Pseudomonas aeruginosa, Bacillus cereus, Escherichia coli and Staphylococcus aureus species were re-identified in the microbiology laboratory of Benson Idahosa University, using cultural, morphological and chemical characteristics [25], [26]. The organisms were inoculated in a test tube containing nutrient broth at 35°C for 16 hr. Standard inoculum density equivalent to McFarland standard No. 0.5 was prepared using a barium sulphate stability standard according to Lenette et al [27] and Bryant [28].

Antimicrobial activity was measured using the agar well diffusion technique [29]. The standardized inoculums were used to swab the surface of solidified nutrient agar plate. Three holes were aseptically made with 7 mm sterile cork borer on the nutrient agar plates and 0.1 ml of each test solution (0.5% honey, palm or raffia syrup) was introduced into holes in the separate plates. Water (0.1ml) and standard streptomycin (10.0 μg) were used as negative and positive controls in determining the sensitivity of each bacterial isolate tested. The plates were incubated at 37 °C for 24 hr and examined for zones of inhibition, which indicate the degree of sensitivity of the test organism [30], [31].

III. Results and Discussion

Oil palm syrup, raffia palm syrup and honey were viscous brown liquids at ambient temperature. Table1 shows their physico-chemical and nutritional characteristics.
Table 1. Physico-chemical and nutritional characteristics of honey and palm syrups.

<table>
<thead>
<tr>
<th></th>
<th>Honey</th>
<th>Raffia palm syrup</th>
<th>Oil palm syrup</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein (N x 6.25, %)</td>
<td>0.43 ± 0.04</td>
<td>0.24 ± 0.06</td>
<td>1.04 ± 0.04</td>
</tr>
<tr>
<td>Ash (%)</td>
<td>0.40 ± 0.02</td>
<td>0.30 ± 0.01</td>
<td>0.50 ± 0.01</td>
</tr>
<tr>
<td>Lipids (%)</td>
<td>3.12 ± 0.02</td>
<td>2.17 ± 0.03</td>
<td>2.20 ± 0.03</td>
</tr>
<tr>
<td>Moisture (%)</td>
<td>28.05 ± 2.83</td>
<td>28.50 ± 4.95</td>
<td>31.50 ± 4.24</td>
</tr>
<tr>
<td>Carbohydrate (by difference) %</td>
<td>68.00</td>
<td>68.79</td>
<td>64.76</td>
</tr>
<tr>
<td>Energy (kcal/100 g)</td>
<td>301.8</td>
<td>295.7</td>
<td>283.0</td>
</tr>
<tr>
<td>Calcium (mg/100 g db)</td>
<td>37.54 ± 3.54</td>
<td>3.8-9.4</td>
<td>37.06 ± 2.80</td>
</tr>
<tr>
<td>Iron (mg/100 g db)</td>
<td>3.30 ± 0.42</td>
<td>22.0-33.0</td>
<td>2.35 ± 0.65</td>
</tr>
<tr>
<td>Potassium (mg/100 g db)</td>
<td>325.12 ± 7.80</td>
<td>32.5</td>
<td>545.75 ± 3.73</td>
</tr>
<tr>
<td>pH</td>
<td>4.18</td>
<td>3.76</td>
<td>3.51</td>
</tr>
<tr>
<td>Density (g/cm³)</td>
<td>1.24</td>
<td>1.23</td>
<td>1.26</td>
</tr>
<tr>
<td>Colour</td>
<td>Dark Brown</td>
<td>Dark Brown</td>
<td>Dark Brown</td>
</tr>
<tr>
<td>Browning intensity (Absorbance of a 1% solution at 420 nm)</td>
<td>0.071</td>
<td>0.159</td>
<td>0.175</td>
</tr>
</tbody>
</table>

*Energy was estimated from proximate composition using the following factors: Protein, 4.0 kcal/ g; carbohydrate, 4.0 kcal/ g; lipid, 9.0 kcal/ g.

*bPercentage of the recommended dietary allowance (RDA) for minerals which may be provided by 100 g of honey or syrup (RDA values: Ca = 400-1000 mg, Fe = 10-15 mg, K = 1000 mg) [32].

Table 2. Total phenolic compounds content and phytochemicals of honey and palm syrups.

<table>
<thead>
<tr>
<th></th>
<th>Honey</th>
<th>Raffia palm syrup</th>
<th>Oil palm syrup</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total phenolic compounds (TPC, mg GAE*/100 g of honey/syrup)</td>
<td>125.93±13.60</td>
<td>170.89±2.87</td>
<td>185.44±10.15</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Glycosides</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Terpenoids</td>
<td>-</td>
<td>+</td>
<td>-</td>
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<tr>
<td>Tannins</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

*GAE: Gallic acid equivalent. +: Present. -: Absent

In recent years, phenolic compounds have been intensively investigated and have been reported to possess many useful properties. They act as antioxidants by preventing oxidation of low density lipoprotein and platelet aggregation, and prevent damage of red blood cells [34]. They also act as metal chelators, antimutagens, anticarcinogens and antimicrobial agents. They are responsible for the red colour, astrignency and bitterness of wine, and contribute to its sensory profile [35]. In wine they are derived from the fruit and vine stems, or by the yeast metabolism. Wild yeasts present in palm sap may also contribute to the synthesis of its phenolic compounds, which may in turn contribute to the sensory profile of palm wine [36]. It has been demonstrated that phenolic compounds content has a strong positive correlation with the antioxidant capacity of honeys [7]. The total phenolic compounds content of honey, raffia palm syrup and oil palm syrup and their phytochemicals are shown in Table 2.
They all contained flavonoids, and their total phenolic content were 125.93±13.60, 170.89±2.87 and 185.44±10.15 mg GAE /100 g respectively. Apart from the phenolic compounds, other classes of phytochemicals of medicinal value were present in the samples (glycosides and saponins in all the materials, and alkaloids only in raffia palm syrup).

During the evaporation of raffia and oil palm sap to produce syrup, the original colour (pale white) gradually turned brown. Browning intensity of raffia and oil palm syrups were 0.159 and 0.175 respectively; that of honey was 0.071. The browning of these materials indicated the presence of Maillard reaction products (MRP) formed from the reaction between the amino acids and sugars present in the oil palm and raffia sap [37], [38] and honey [7]. Several authors have found that MRP have antioxidant activity [39]. They exert antioxidant activity by scavenging oxygen radicals and/or by chelating metals [40], [23], [41]. In addition they exhibit antimicrobial activity. Antimicrobial activity of MRP has been attributed to their inhibition of sugar metabolising enzymes in micro-organisms [42] to their antioxidant activity [43] and to their ability to chelate ions, such as Fe, Zn and Cu, which are essential for growth and survival of pathogenic organisms [44].

Clinical samples of four organisms Escherichia coli, Pseudomonas aeruginosa, Bacillus cereus, and Staphylococcus aureus were tested for growth inhibition by honey, raffia palm syrup and oil palm syrup. A known antibiotic (streptomycin) was used as positive control and distilled water as negative control. Table 3 shows the antimicrobial activity of 0.1 ml aliquots of 0.5% solutions of honey (containing 360 µg dry matter), raffia palm syrup (containing 360 µg dry matter) and oil palm syrup (containing 340 µg dry matter).

### Table 3. Zones of inhibition of bacterial growth (cm) by honey, raffia palm syrup, oil palm syrup, streptomycin and water.

<table>
<thead>
<tr>
<th>Material</th>
<th>Amount</th>
<th>Escherichia coli</th>
<th>Pseudomonas aeruginosa</th>
<th>Bacillus cereus</th>
<th>Staphylococcus aureus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Streptomycin</td>
<td>10µg</td>
<td>0.6</td>
<td>0.6</td>
<td>0.5</td>
<td>0.6</td>
</tr>
<tr>
<td>Honey</td>
<td>360µg</td>
<td>0.7</td>
<td>0.6</td>
<td>0.4</td>
<td>0.6</td>
</tr>
<tr>
<td>Raffia palm syrup</td>
<td>360µg</td>
<td>0.8</td>
<td>0.6</td>
<td>0.4</td>
<td>0.7</td>
</tr>
<tr>
<td>Oil palm syrup</td>
<td>340µg</td>
<td>0.6</td>
<td>0.5</td>
<td>0.6</td>
<td>0.6</td>
</tr>
<tr>
<td>Distilled water</td>
<td></td>
<td>NZ</td>
<td>NZ</td>
<td>NZ</td>
<td>NZ</td>
</tr>
</tbody>
</table>

*Amount of dry matter in 0.1ml of 0.5% solution. *NZ: No zone of inhibition.

Honey gave zones of inhibition of 0.7, 0.6, 0.4 and 0.6 cm against Escherichia coli, Pseudomonas aeruginosa, Bacillus cereus, and Staphylococcus aureus, raffia palm syrup, 0.8, 0.4, 0.6 and 0.7 cm, oil palm 0.6, 0.5, 0.6 and 0.6 cm and streptomycin, 0.6, 0.6, 0.5 and 0.6 respectively. Zones of inhibition of the growth of Escherichia coli, Pseudomonas aeruginosa, Bacillus cereus, and Staphylococcus aureus were therefore similar for 360 µg of raffia palm syrup, 360 µg of honey, 340 µg of oil palm syrup and 10 µg of streptomycin. This shows that on a weight for weight basis, much more honey, oil palm or raffia palm syrup than streptomycin was required in order to obtain similar growth inhibition. Thus streptomycin was a far more potent antibiotic against the organisms than the honey or syrups. However, the syrups and honey, like streptomycin, were active against Gram-positive and Gram-negative organisms.

Sugars, at moderately low concentrations (0.5-2%) act as nutrients; they only inhibit growth when present in media at concentrations of 20 – 40% [45]; this suggests that the sugars present in the very low concentrations of the palm syrups and honey (0.5%) employed in this study might have acted as nutrients, being insufficient to prevent bacterial growth. The limits of hydrogen ion concentration for growth of micro-organisms are from around pH 4.0 to 9.0. Bacteria in general, prefer media of pH near neutrality, and cannot usually tolerate values much below 4 – 5 [45], so the acid pH of honey (4.18) and palm syrups (3.76 for oil palm and 3.51 for raffia palm syrup) would be unfavourable for growth of the test organisms employed in this study. Thus the antimicrobial activity of the honey and palm syrups examined might be due to their acidity, and the phenolic compounds and Maillard reaction products present in them.

### IV. Conclusion

The physical-chemical and nutritional characteristics, and the antimicrobial properties of honey, oil palm syrup, raffia palm syrup and honey were examined. The results indicate that the syrups were similar in composition to honey, were rich in carbohydrates and potassium, and contained modest amounts of calcium and iron. The syrups (like honey) also contained Maillard reaction products and phenolic compounds, alkaloids (in raffia palm sap), saponins and glycosides, and exhibited broad spectrum antimicrobial activity. Their composition suggests that oil palm sap and raffia palm sap based syrups have potential for utilisation as nutritious food ingredient. Their broad spectrum antimicrobial activity suggests that, like honey, they could be used for food preservation, and for medicinal applications such as the treatment of burns.
Acknowledgements

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