



Assessment of Nutritional and Anti-Microbial Qualities of Pawpaw (Carica Papaya) Seed Obtained From Zungeru Niger State, Nigeria

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ABSTRACT

The proximate, mineral and antimicrobial activity pawpaw (*Carica papaya*) was carried out; this sample seed was obtained from discarded plants and prepared for use by decocting, sun drying and grinding into powder. Using petroleum ether; of boiling range 40-60°C, their fats were extracted, the protein content, ash content, crude fibre, moisture, carbohydrate as well as the mineral contents were determined using standard methods. The fats yield of 26.01±0.00%, crude protein 8.75±0.01%, crude fibre 14.01±0.02%, carbohydrate content 36.25±0.01% and calories 414.09±0.02 kcal/100g was obtained for the sample. The mineral compositions determination of the sample showed that 13.70±0.12, 5.46±0.003, 26.58±0.02 and 9.06±0.30 mg/100g for potassium, calcium, iron and copper respectively. The antimicrobial activity of the methanol extract of *C. papaya* seed was determined. The MIC value for the methanol extract was between 0.875 and 1.75 mg/ml, while that of MBC was between 1.75 and 2.5 mg/ml.

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Introduction

Waste generated from food industries is a source of an untapped energy which is mostly dumped inland fills whereby it releases greenhouse gases into an atmosphere. It is very difficult to treat and recycle food waste due to its composition. Food industry produces large volumes of wastes, both solids and liquid, resulting from the production, preparation and consumption of food. These wastes lead to increasing waste disposal and can pose severe pollution problems and represent a loss of valuable biomass and nutrients (Vasso and Winfried, 2007). Waste utilization in fruits and vegetable processing industries is the one of the important and challengeable job around the world. It is anticipated that the discarded fruits as well as weight materials could be utilized for further industrial purposes (Atul, 2010). Considering the challenges in the area of food industry, efforts are to be made to optimize processing technologies to minimize the amount of waste. Research focusing on the potential application of natural antimicrobial compounds in food, cosmetic and pharmaceutical industries has gained considerable importance owing to the growing number of antibiotic resistant pathogens (Rao and Rao 2007). The emergence of antibiotic resistance has been linked to the overuse of antibiotics which helps the bacteria to adapt to the drugs through alteration in their genetic makeup (Paterson et al. 2004). In the recent years, apart from medicinal plants, fruits, which are a rich source of bioactive compounds, have become popular subjects for such investigations.

Possible products that can be considered from fruit waste are as candied peel, oils, pectin, reformed fruit pieces, enzymes, wine and vinegar (Singh, 2007). Each of the above uses for fruit waste requires: a good knowledge of the potential market for products and of the quality standards required, a careful assessment of the economics of production,

a certain amount of additional production knowledge, a certain amount of additional capital investment in equipment, a fairly large amount of waste to make utilisation worthwhile. Besides, it finds numerous applications in pharmaceutical preparations, pastes and cosmetics. All these combined efforts of fruit waste minimization during the production process, environmentally friendly preservation of the fruit peel, and utilization of fruit waste by-products would substantially reduce the amount of fruit waste, as well as boost the environmental aspect of fruit processing industry (Pap, 2004).

Pawpaw (*Carica papaya* L.) belongs to the family of Caricaceae with many species such as *Asimina reticulata*, *Asimina incarna*, *Asimina longifolia* and *Asimina parviflora* etc. It is a deciduous, often narrowly conical tree growing from 12 to around 20 feet. Its trees are prone to producing root suckers a few centimetres from the trunk and when these are permitted to grow, the single clone pawpaw patch comes into being (Peterson and Neal, 1991).

Materials and Methods

Sample Collection

The seeds of pawpaw (*Carica papaya*) used in the course of this work was obtained from Zungeru in Niger state of Nigeria. The fruit were separated from the seed. The seed were washed with clean water, dry and ground into powder form using electric grinder. The grinded samples were store in a well labeled air -tight container at ambient temperature for further analysis.

Methods

Moisture Content

2g of the sample was put into the crucible, dried in an oven at 105°C overnight. The dried samples were cooled in a dessicator for 30 minutes and weighed to a constant weight.

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The percentage loss in weight was expressed as percentage moisture content (AOAC, 1999). This was repeated twice.

Ash Content

2.00g of the grounded sample was placed in a crucible and ashed in a muffle furnace at 6000C for 3 hours. The hot crucibles was cooled in a dessicator and weighted. The percentage residual weighed was expressed as ash content (AOAC, 1999).

Crude lipid content determination

From the pulverized sample, 2.00 g was used for determining the crude lipid by extracting the lipid from it for 5 h with (60 to 80°C) petroleum ether in a soxhlet extractor (AOAC, 2006). Triplicate samples were extracted to obtain triplicate values that were later averaged.

Protein determination

Total protein was determined by the Kjeldahl method. 0.5 g of the sample was weighed in triplicate into a filter paper and put into a Kjeldahl flask, 8 to 10 cm³ of concentrated H₂SO₄ were added and then digested in a fume cupboard until the solution became colorless. Distillation was carried out with about 10 cm³ of 40% NaOH solution. The condenser tip was dipped into a conical flask containing 5 cm³ of 4% boric acid in a mixed indicator till the boric acid solution turned green. Titration was done in the receiver flask with 0.01 M HCl until the solution turned red (AOAC, 2006).

Determination of crude fibre

From the pounded sample, 2.00 g were used in triplicates for estimating the crude fibre by acid and alkaline digestion methods using 20% H₂SO₄ and 20% NaOH solutions (AOAC, 2006).

Carbohydrate Determination

The carbohydrate content was calculated using following: Available carbohydrate (%), = 100 - [protein (%) + Moisture (%) + Ash (%) + Fibre (%) + Fat (%)].

Elemental analysis

The sample (0.5 g each) was put into Kjeldahl digestion flask to which 24cm³ of a mixture of concentrated nitric acid (HNO₃), conc. H₂SO₄ and 60% HClO₄ (9:2:1v/v) was added. The flask was allowed to stand over-night to prevent excess foaming (Sahrawat et al., 2002). The flask was put on a heating block and digested to a clear solution, cooled and the content filtered into a 50 cm³ volumetric flask. The solution was then diluted to the volume with distilled water. Blank solution was prepared in similar manner without sample being added. The solution was used for the mineral analysis.

The mineral contents (calcium, magnesium, iron, zinc, copper, manganese and lead) were analysed using AAS. Sodium and potassium were analysed using atomic emission spectrometry and phosphorus was determined by colorimetry using Vanadomolybdate (blue) method (AOAC, 2000).

MIC and MBC

Eight clinically isolated pathogenic bacterial strains that included five gram negative bacteria (*Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Proteus vulgaris*, *Salmonella typhi*) and three gram positive bacteria (*Streptococcus pyogenes*, *Staphylococcus aureus*, *Enterococcus faecalis*) were used for the present investigation. The antimicrobial activity was tested using the agar well diffusion method (Okeke et al. 2001).

Extracts with significant inhibition zones based on agar well diffusion method were evaluated for their MIC using two fold broth dilution method (Chattopadhyay et al. 1998). 1 ml of each fruit peel extract was prepared at a concentration of 2.5 mg/ml and was serially diluted two fold in sterile tubes

containing 1 ml of Mueller Hinton broth to achieve concentrations of 2.5, 1.75, 0.875 and 0.437 mg ml⁻¹. Bacterial inoculum (0.1 ml of 10⁸ c.f.u ml⁻¹) were pipetted into the tubes and kept for incubation at 37°C for 24 hours. The lowest concentration tube that did not show any visible growth was considered as MIC. 0.1 ml of each of the inoculated broths were transferred and spread onto fresh Mueller Hinton agar medium plates, and the tube containing the lowest concentration of the extract which did not show any visible growth on the plates was considered as MBC. All the assays were performed in triplicates.

Statistical Analysis

Data generated in triplicates were expressed as mean ± standard deviation using SPSS version 16 statistical packages.

Results and Discussion

Table 1. Proximate analysis of *C. papayaseed* (mg/100g).

S/N	Parameters	Value
1	Ash	4.00±0.00
2	Crude protein	8.75±0.01
3	Moisture	11.00±0.01
4	Crude fat	26.01±0.00
5	Crude fibre	14.01±0.20
6	Carbohydrate	36.25±0.01
7	Calories (Kcal/100g)	414.09±0.02

Values are mean±SD of three determinations

Table 2. Mineral composition of *C. papayaseed* (mg/100g).

SN	Parameters	Value
1	Calcium	5.46±0.03
2	Potassium	13.70±0.01
3	Magnesium	3.43±0.21
4	Sodium	9.00±0.10
5	Phosphorus	9.00±0.12
6	Iron	26.58±0.02
7	Zinc	7.06±0.10
8	copper	9.06±0.030

Values are mean±SD of three determinations

Table 3. MIC and MBC Concentration of Methanol extract of *C. papaya Seed*.

S/N	Microorganism	MIC (mg/ml)	MBC (mg/ml)
1	<i>Escherichia coli</i>	0.875	1.75
2	<i>Klebsiella pneumoniae</i>	1.75	2.5
3	<i>Pseudomonas aeruginosa</i>	1.75	2.5
4	<i>Proteus vulgaris</i>	0.875	1.75
5	<i>Salmonella typhi</i>	0.875	1.75
6	<i>Streptococcus pyogenes</i>	0.875	1.75
7	<i>Staphylococcus aureus</i>	0.875	1.75
8	<i>Enterococcus faecalis</i>	0.875	2.5

Table 1 shows the result of proximate composition of *C. papaya*. Fats play a vital role in maintaining health skin and hair, insulating body organs against shock, maintaining body temperature and promoting health cell function. It is also essential nutrients as they increase the pleasant to taste of food by absorbing and retaining their flavours (Omotoso, 2006). The crude fat of the studied seed was 26.01±0.00%. The values of the fat yield of this sample can make the industrial practice of the fat recovery from this sample a profitable venture and will reduce the level of waste that is obtained from juice making industries especially those using *C. papaya*. The value obtained in this work was low compared to that recorded by Anwar et al., (2008) on grape seeds (46.2%). This high value indicated that the seeds are a good source of oil. The crude protein content of the sample was 8.75±0.01. The protein content recorded in this work was low to reported protein content of orange, grape and white roselle seeds which were found to be 20.20, 21.40 and 22.70% respectively by Gerner and Poiters, (2008).

The protein content in this sample showed that the sample can be regarded as good sources of protein hence the cake can be modified into protein concentrate feeds for livestock. The level of protein in the sample indicates that they contribute significantly to the daily protein requirement of 23-56g for humans as stipulated by the NRC, (1980). The seed of the studied plant was found to have ash content of $4.00 \pm 0.00\%$. This sample with the highest ash content had the highest probability of being the one with the highest mineral contents, as the ash content of grape was taken as a rough measure of the mineral contents of the food material (Anwar et al., 2008). The result of the ash content of studied seed was similar with that of citrus seeds (4.60%) reported by Anwar et al., (2008). The moisture content of the $11.00 \pm 0.01\%$ was recorded for *C. papaya*. The value was high compared to other fruit seed like citrus seed. This indicated that the *C. papaya* cannot be preserved for a reasonable period of time without the risk of microbial deterioration and spoilage. The long shelf-life promised here is an added advantage over other sources of protein like beef, egg and fish which are easily prone to spoilage if proper care is not given to them. The crude fiber content of $14.01 \pm 0.20\%$ was recorded for the studied seed. The value agreed with 5.0-58% reported by Anwar et al., (2008). The fibre content of *C. papaya* seed was higher than that of orange seeds (11.0%) and grape seeds (7.50%) but lower than that of red roselle (28.50%). The physiological role of crude fibre in the body is to maintain an internal distension for proper peristaltic movement of the intestinal tract (Oduoret al., 2008).

From Table 2, the metallic compositions of *C. papaya* seed was determined. Calcium contents of *C. papaya* seed was 5.46 ± 0.03 mg/100g. This concentration was however, in agreement with what was obtained for the melon seeds reported by Anwar et al. (2008). The zinc and magnesium contents from the table tended to have the least of metallic composition, but the value agreed with that of citrus seeds 1.00-9.00 mg/100g reported by Brown et al., (1993). From the table, the potassium level of *C. papaya* seed was about 13.70 ± 0.01 mg/100g. This level was lower than the reported for the melon seeds 25.0 mg/100g (Bird, 1990) but higher than that of the citrus seeds 1.0 mg/100g reported by Anwar et al., (2008). In the case of iron content in the plant, it was observed that *C. papaya* seed has high content of iron. However, the iron content of melon seeds was lower than the value obtained from this work. It was also observed that copper content obtained from this studied was $9.06 \pm 0.30\%$. However this value was lower than 11.0-19.0 mg/100g reported by Anthony, (1986) for the melon seeds. Generally, from table 2.0 above, it was observed that, the plant seed was rich in copper, sodium, potassium and iron contents while calcium, zinc and magnesium were not as much in the present studied.

The MIC is interpreted as the lowest concentration that inhibits visible microbial growth and expressed in terms of mg/ml, whereas the minimum bactericidal concentration (MBC) is interpreted as the lowest concentration that can completely remove the microorganism. The MIC and MBC were determined for the extracts showing very high potential antimicrobial activity, the methanol extract of *C. papaya* seed. As seen from Table 3, the MIC values for the methanol extract was between 0.875 and 1.75 mg/ml, while that of MBC was between 1.75 and 2.5 mg/ml. The *C. papaya* seed extract showed the same value for MIC and MBC this finding supports the present antimicrobial study which reports very high antimicrobial activity observed for *C. papaya* seed extract.

MIC and MBC values for *C. papaya* seed extract were observed to be more against all the tested bacteria. The MIC and MBC values of *C. papaya* seed extract against these bacteria was observed to be higher too, thus indicating the high resistance of these bacteria.

Conclusion

From the results of this work, it is strongly recommended that the industrial production and commercialization of the *C. papaya* seed should be given adequate attention in order to supplement the conventional seed fats/oils like cornflower, groundnut, linseed and citrus seeds to provide more sources of edible and industrial oils. This will also reduce waste hence a useful tool for economic development since wealth will actually be produced from waste. In addition, they can be used in feed formulations.

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