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Nutritional and Antimicrobial Properties of *Jatropha tanjorensis* Leaves

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Abstract: Fresh green *Jatropha tanjorensis* leaves were analysed for protein, moisture ash, mineral content and antimicrobial activity. The fresh leaves had a moisture content of 78.77%, a protein content of 2.01% and an ash content of 0.51%. On a fresh weight basis, mineral content was as follows: phosphorus, 77.0μg g^{-1}; selenium $5.4x10^{-3}$μg g^{-1}; iron, 9.4μg g^{-1} and zinc, 6.1μg g^{-1}. An aqueous extract of the leaves inhibited the growth of the gram +ve bacterium *Staphylococcus aureus* and the gram –ve bacterium *Escherichia coli.* The nutritional implications of the results are discussed.

Key words: *Jatropha tanjorensis* leaves · Moisture · Protein · Minerals · *Escherichia coli* · *Staphylococcus aureus*

INTRODUCTION

The plant *Jathopha tanjorensis* is widely grown in Southern Nigeria. Its primary use is for fencing while its secondary uses are as a source of edible leafy vegetable and as medicine (leaves). The leaf extract has hypoglycaemic properties and is taken as medicine for the treatment of diabetic symptoms [1]. It also exhibits low antioxidant and very low hemaglutination titre value, the latter indicating low toxicity on red blood cells [2]. In the present study, the moisture, protein and mineral content of *J. tanjorensis* leaves and the antimicrobial effect of their aqueous extract on the bacteria *S. aureus* and *E. coli* were determined. The nutritional and food processing implications of the results are discussed.

MATERIALS AND METHODS

Materials

Vegetables: The collection of *J. tanjorensis* leaves was done in the rainy season, in the month of May 2007 in the neighbourhood of Benson Idahosa University Campus, Ugbor Road, Benin City, Nigeria. Leaves used for analysis were freshly cut and were dark green in colour.

Reagents: All reagents were analytical grade.

Analytical Methods

Moisture: Shredded fresh vegetable (10g) was dried in a thermostatically controlled ventilated oven at 105°C until constant weight was obtained. The loss in weight was recorded as moisture content [3].

Sample Preparation for Protein, Ash and Mineral Analysis: Leaves were cut into tiny pieces and dried in a ventilated oven at 60°C for 5 days to constant weight. The dried vegetable was ground into powder and stored in airtight bottles for analysis.

Crude Protein: Crude protein was determined by the Kjeldahl method. Dried and pulverised leaf (0.2g) was digested in 2ml concentrated H_2SO_4 in the presence of selenium catalyst, until a clear digest was obtained [3]. The nitrogen content of diluted digest was determined colourimetrically at 630nm according to Charlot [4]. Protein content of leaves was calculated as: Nitrogen content x 6.25.

Ash: For the determination of ash content, dried pulverised vegetable was ashed at 550°C in a muffle furnace.

Minerals: Minerals were obtained by ashing 2.0g dried and ground sample in a muffle furnace at 550°C. The ash was dissolved in 10ml, 20% nitric acid and filtered into a 100ml volumetric flask. The filtrate was made up to the mark with deionised water and the resulting solution was used for the analysis of phosphorus, zinc and iron.

Phosphorus was determined colourimetrically according to Charlot [4]. To mineral solution (10ml) in a

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50 ml volumetric flask was added 0.2ml of 0.5% para-nitrophenol solution. To this was added an ammonia solution (6N). Nitric acid was added until the solution turned colourless. Finally ammonium molybdate/ ammonium vanadate mixed reagent (5ml) was added. The solution was made up to 50ml with distilled water; the flask was stoppered and its contents well mixed. The flask was left for 30min and the absorbance of the solution measured at 400nm. Phosphorus content of the solution was read off a calibration graph prepared using potassium dihydrogen phosphate.

Zinc and iron were determined using an atomic absorption spectrophotometer (Buck Scientific VGP 210) at 630nm [5].

Selenium was determined titrimetrically [6]. To an aqueous extract of the leaves were added 5ml starch solution and 1g potassium iodide. The mixture was substitute stirred for mixed, allowed to stand for 15 seconds and titrated with 0.05N sodium thiosulphate solution. The end point was marked by a change from a dirty or turbid solution to a transparent red colour.

Analyses Were Done in Duplicate

Antimicrobial Activity

Preparation of Crude Water Extract of Leaves: The aqueous extract was prepared by blending 10g fresh leaves in 100ml peptone water (15g peptone in 1l water). The extract was filtered through Whatman No. 1 filter paper and tested for antimicrobial activity.

Preparation of Crude Antibiotic Discs: Sterile Whatman No. 1 paper was punched into 5mm diameter disc sizes. The Whatman discs were placed in MacCartney bottles and sterilised in an autoclave at 120°C for 15 min. The bottles was transferred into a hot air oven at 60°C to dry for 30min. An aqueous extract of the leaves (1.0ml) was transferred into a sterile Bijou bottle containing sterile discs. The sterile crude discs were allowed to soak in extract for 6hr for proper absorption, after which they were removed and allowed to dry [7].

Antimicrobial Assay of Extract: The aqueous extract of *J.tanjorensis* leaves was evaluated *in vitro* for antimicrobial activity against *Staphylococcus aureus* and *Escherichia coli.*

Nutrient agar (7g) was added to 250ml distilled water in a flask. This was stirred and autoclaved at 115°C and then cooled to 50°C. A portion of the medium (20ml) was poured into a sterile Petri dish and allowed to solidify. The sterility of the medium was confirmed by allowing it to stay for 8hr and observing no contamination.

An isolate colony of each test organism was sub-cultured on nutrient broth and incubated at 37°C for 8hr. This was then spread on the entire plate medium to ensure uniform growth. Sterilised crude extract discs were placed on the plates containing the test organisms. The plates were then incubated immediately for 24hr at 37°C [8]. Zones of inhibition were observed using a hand lens for proper magnification and measured. Antimicrobial assays were carried out in quadruplicate.

RESULTS AND DISCUSSION

J. tanjorensis leaves were obtained freshly cut and were dark green in colour. The moisture, protein and ash content of the leaves are given in Table 1. Included for comparison are published results for some locally available leafy vegetables- cassava leaves, cabbage, cowpea leaves and sweet potato leaves.

The dominant constituent of *J. tanjorensis* leaves was water, which accounted for 78.77% of their weight. Fresh leaves had a protein content of 2.01% and an ash content of 0.51%. The leaves had moisture content similar to published values for cassava, cabbage, cowpea and sweet potato leaves. Protein content of the leaves was low and similar to the published value for cabbage, but was lower than published values for cassava, cowpea and sweet potato leaves (van Gastel and van den Wijingaart, 1997, Madamba, 2006).

Overall, foods such as leafy vegetables, with a high water and adequate dietary fibre content provide a low energy density contribution to the meal and create a feeling of satiety. Generally, fresh leafy vegetables have low protein content. This protein is mostly in the form of enzymes, rather than acting as a storage pool, as in grains and nuts [11]. The leaves of cassava, cowpea and sweet potato and cabbage appear to be better sources of dietary protein than *J. tanjorensis* leaves. Thus fresh *J. tanjorensis* leaves may not be an important source of dietary protein. Ash content of leaves was low, indicating a low content of minerals.

Table 1: Moisture, protein and ash content (g/100g) of *J. tanjorensis* leaves

Characteristics	*J.tanjorensis*[a]	Cassava[b]	Cabbage[b]	Cowpea[c]	Sweet potato[b]
Moisture	78.77	80.0	79.0	85.0	83.0
Protein	2.01	6.0	1.4	4.7	4.6
Ash	0.51	-	-	-	-
Colour	Dark green	Dark green	Dark green	Dark green	Dark green

[a]This study. [b]van Gastel and van den Wijingaart [9]. [c]Madamba et al [10].

Table 2: Mineral content (μg g^{-1}) of *J. tanjorensis* leaves

Mineral	*J. tanjorensis*[a]	Cabbage[b]	Cowpea[c]	RDA[b]	%ofRDA
Phosphorus	77.0	540.0	90.0	1300	0.59
Selenium	$5.4x10^{-3}$	-	-	0.055[d]	0.1
Iron	9.4	6.0	19.0	10-15	6.3-9.4
Zinc	6.1	3.0	3.0	15	4.1

[a]This study. [b]RDA in mg [13]. [c]Madamba *et al.* [10]. [d]Wardlaw and Kessel [12].

Table 3: Zones of inhibition (cm) of bacteria by aqueous extract of *J. tanjorensis* leaves

Microorganism	Zone of inhibition
Staphylococcus aureus	1.60
Escherichia coli	1.13

Table 2 shows the phosphorus, selenium, iron and zinc content of *J. tanjorensis* leaves. Included for comparison are values for cassava, cabbage and cowpea leaves and the daily recommended allowances (RDA) for the minerals.

Phosphorus constituted 77.00μg g^{-1} of fresh *J. tanjorensis* leaves (i.e. 0.59% of the RDA for this element was present in 100g of leaves). This was much lower than the published value for cabbage (540.0μg g^{-1}), but similar to that for cowpea (90.0μg g^{-1}). Thus *J. tanjorensis* leaves contained a modest amount of phosphorus.

Selenium constituted 0.0054μg g^{-1} of *J. tanjorensis* leaves. Thus about 0.1% of the RDA for this element (i.e. 0.54μg) was present in 100g of leaves, about one-twentieth of its content in foods considered good sources of the mineral - one boiled egg (11.0μg), half a cup of oatmeal (10.0μg), a slice of whole wheat bread (10.0μg), or a slice of white bread (8.0μg) [12]. Iron content was 9.4μg g^{-1}, (i.e. 6.3-9.5% of the RDA for this mineral was present in 100g of leaves). This was higher than the published value for cabbage (6.0μg g^{-1}), but lower than that for cowpea leaves (19.0μg g^{-1}). This value compares favourably with those of sources considered nutritionally dense in iron, e.g. beef and herring fillets (16.0μg g^{-1} and 8.0μg g^{-1} respectively) [13]. Zinc constituted 6.1 μg g^{-1} of fresh leaves (i.e. 4.1% of the RDA for this element was present in 100g of leaves). This is higher than its content in cabbage (3.0μg g^{-1}), cowpea leaves (3.0μg g^{-1}), milk (3.5μg g^{-1}), cod fillets (4.0μg g^{-1}) and herring fillet (5.0μg g^{-1}) [13, 10]. *J. tanjorensis* leaves therefore appear to be a modest dietary source of the trace element selenium and a good source of iron and zinc.

The effect of an aqueous extract of *J. tanjorensis* on the gram positive bacterium *Staphylococcus aureus* and the gram negative bacterium *Escherichia coli* is presented in Table 3.

Both organisms exhibited sensitivity to the extract, giving zones of inhibition of 1.6cm and 1.13cm for *S. aureus* and *E. coli* respectively.

Leafy vegetables are high moisture, low acid produce, which support the growth of a wide range of microorganisms. Thus great care is needed when processing them, in order to minimize the risk of contamination by bacteria that cause food poisoning, especially those, for example *S. aureus,* which produce heat stable toxins that may not be destroyed by heat treatment such as cooking [13-15]. *S. aureus* and *E. coli* are common food poisoning bacteria. The antimicrobial activity of *J.tanjorensis leaves* could inhibit the growth of these bacteria on the vegetable itself and (by extraction during cooking, if the antimicrobial principle is heat stable) in food, e.g. sauces and stews in which it is an ingredient, thus protecting the consumer from their harmful effect.

CONCLUSION

The moisture, protein, ash, mineral content and antimicrobial properties of *J. tanjorensis* leaves were determined. Fresh *J. tanjorensis* leaves had a high water and low protein content with modest phosphorus content. The trace elements zinc, iron and selenium occurred in concentrations comparable to those found in food regarded as good dietary sources of these minerals. *J.tanjorensis leaves* therefore appear to be a good dietary source of these elements.

Cold aqueous extract of *J. tanjorensis* leaves inhibited the growth of *S. aureus* and *E. coli.*

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