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The Nutritive, Anti-Nutritive and Hepatotoxic Properties of *Trichosanthes anguina* (Snake Tomato) Fruits from Nigeria

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Abstract: Ripe fruits of *Trichosanthes anguina* (snake tomato) were subjected to phytochemical and proximate analyses. Hepatotoxic effects of various feeds compounded with fruits and seeds of the plant were also investigated. Twenty-five male rabbits randomly distributed into 5 groups (A-E) of 5 animals each, were respectively fed grower's mash as the control diet, and raw fruits, cooked fruits, cooked fruits without seeds and cooked seeds respectively for 4 weeks. The levels of serum total (TB) and conjugated (CB) bilirubin, as well as aspartate aminotransferase (AST), alanine aminotransferase (ALT) and alkaline phosphatase (ALP) activities were spectrophotometrically determined weekly. The presence of saponins, flavonoids, cyanogenic and cardiac glycosides as well as tannins was observed. The proximate and vitamin analyses yielded moisture (93.15±0.05%), carbohydrates (3.48±0.04%), proteins (1.85±0.04%), vitamins C (18.9±0.05 mg 100 mL^{-1}) and A (347.0±0.02 ug 100 mL^{-1}). At the end of the feeding period, groups B and E animals significantly lost 102.5±10.61 g and 47.5±17.59 g of their body weight ($p<0.05$), while groups C and D animals, non significantly gained 20.0±8.28 g and 25.0±6.12 g ($p>0.05$) respectively, in relation to the control group. The levels of serum TB, CB, AST, ALT and ALP were all significantly raised in groups B and E animals ($p<0.01$). However, in groups C and D, only ALP activity was significantly raised ($p<0.05$), with TB, AST and ALT levels remaining almost normal. The results indicate that *T. anguina* fruit contains important nutrients and will not be hepatotoxic unless consumed raw or unprocessed. The findings are of nutritional and industrial relevance since the fruit is currently being used in place of tomatoes in many homes in Nigeria.

Key words: *Trichosanthes anguina*, phytochemical and proximate analyses, nutritive and anti-nutritive constituents

Introduction

Snake tomato, also called snake gourd or English tomato, belongs to the *Cucurbitaceae*, a family of flowering plants. A large number of cucurbits are cultivated and most of these are tendril-climbing annual or occasionally perennial, rapid growing herbs (Purseglove, 1991). Some of the cultivated species in Nigeria are watermelon species variously called *Citrillus lanatus* (Thunb.), *Citrillus vulgaris* Schrad, *Colocynthis citrullus* (Linn) O. Ktze. Others are the *egwusi* species, *Cucumeropsis edulis* and *Cucumeropsis mannii*; the popular cucumber (*Cucumis sativus*); the pumpkins (*Cucurbita maxima* and *Cucurbita pepo*); and by far the most important of the lot, fluted pumpkin (*Telfairea occidentalis*) commonly called *ugu* in different areas of Nigeria, and used as vegetable for stews and soups among the different ethnic nationalities of the country (Enwere, 1998). Some cucurbits are cultivated in Nigeria, however, for non-culinary reasons. An important example is *Lagenaria siceraria* (Molina) Standl, the calabash or bottle gourd with its various cultivars, which produce fruits of various shapes and sizes, which serve as containers (calabashes) for storing domestic items. Others serve as basins, drinking cups, ladles and spoons, etc.

Snake tomato (*Trichosanthes anguina* Linn or *Trichosaanthes cucumerina*), on the other hand, is of little or no importance in the several villages in Nigeria where it grows in the wild. However, a few families plant few seeds at the fences to serve as hedges. The plant is well suited to growth in the humid lowland tropics. Most gourd specialists agree that *T. anguina* and *T. cucumerina* are the same species, recommending however, that the name *T. anguina* be reserved for the wild species (ECHO, 2000). Characteristically, snake tomatoes are tendril-climbing annuals, which bear fruits 2-4 months after sowing the seeds. The fruits are generally long, narrow and cylindrical giving the shape of a snake. The fruit appears green when unripe, turning orange-red on ripening. The mature unripe fruit has a bitter taste.

In India and some other parts of the world, the plant is grown principally for the immature fruit that can be cooked and served as a vegetable (ECHO, 2000). In eastern part of Nigeria, it is only the ripe pulp that is assumed to be edible. It is very red in colour and can be used to improve the appearance of food as it can be blended and used to produce a paste for stew which tastes like, and serves the role of tomatoes, hence justifying the name of the plant (Enwere, 1998). The present study was aimed at determining the nutritional and anti-nutritional values of the fruit and its seeds in their native and processed forms.

Materials and Methods

Plant sample collection and preparation: Fresh ripe samples of snake tomato fruits were collected from Oforolla village in Owerri Local Government Area of Imo State, Nigeria, and Dr. S.E. Okeke, a plant taxonomist of the Department of Plant Biology and Biotechnology, Imo State University, Owerri, Nigeria, graciously confirmed the botanical identity. Apparently healthy fruits were sorted, washed to remove dust particles and sun-dried for 4 days before the final drying in an oven at 65°C to a constant weight. The dried fruits were ground into powder and used for the phytochemical screening and Proximate analysis.

Some of the fresh fruits were boiled in water at 100°C until the pericarp became soft. These were separated into cooked fruit, cooked fruit without seeds and cooked seeds only. They were also oven dried to a constant weight and ground. These, together with the dried ground raw fruit, were used to prepare different experimental diets.

Phytochemical studies: The phytochemical screening of the processed fruit powder for the presence of saponins, tannins, alkaloids, flavonoids, cyanogenic and cardiac glycosides were carried out as described by Ojiako and Akubugwo (1997).

Quantitative determinations of oxalates, phytate and tannins were also carried out (AOAC, 1990). Similarly, vitamins A and C were estimated using the methods described by Ojiako and Akubugwo (1997).

Proximate analysis: Crude fat was extracted by the soxhlet method with petroleum ether (40-60°C) for 8 h. Crude protein content was determined by the microkjeldahl method. These, as well as carbohydrate, crude fiber, ash and moisture contents were estimated as described by the Association of Official Analytical Chemists (AOAC, 1990).

Toxicological assessment: Twenty-five male albino rabbits with mean weight, 462.0±42.56 g and mean age, 10±2 weeks, were divided into 5 groups; A, B, C, D and E, and housed in separate individual cages. They were allowed access to water and feed diet (Guinea Feed Nigeria Ltd, Nigeria) *ad libitum* for a week to equilibrate them to laboratory conditions. After this period, the control animals (Group A) were continued on the commercial feed uninterrupted, while the Group B animals were placed on compounded raw fruits of *T. anguina*, Group C on cooked fruits, Group D were on cooked fruits without seeds and Group E animals were placed on cooked seed feed. All experimental feeds were compounded by the blending of 50% commercial feed with 50% *T. anguina* diet. The animals were fed for 28 days with these designated feeds and water *ad libitum*. The weights of the animals were taken immediately before commencement of the feeding experiment and afterwards, at a weekly interval for the remaining period the experiment lasted.

Liver function tests: Blood samples were drawn using a 5 mL syringe through the marginal ear vein of the animals on the day of commencement of the feed regimen and weekly thereafter. The blood samples were dispensed into plain glass containers, allowed to clot and retract, and then were centrifuged and sera separated for the liver function tests.

Total (TB) and conjugated (CB) bilirubin were determined according to the diazo method as described by Balistreri and Shaw (1987). Total bilirubin was determined in the presence of an accelerator caffeine-benzoate, which releases albumin-bound bilirubin. Bilirubin then reacts with diazotized sulphanilic acid to form pink-coloured azobilirubin, which in the presence of alkaline tartarate forms a coloured solution whose concentration is determined spectrophotometrically at 600 nm. Only the conjugated forms of bilirubin will react with the diazo reagent in the absence of the accelerator. Alkaline phosphatase (ALP), and aspartate (AST) and alanine (ALT) aminotransferases activities were assayed by the use of King and Armstrong, and Reitman and Frankel (1957) methods respectively, as described by Balistreri and Shaw (1987).

Statistical analysis: Data collected were statistically analyzed for differences between individual groups by the use of students' t-test and simple percentages. Values for $p<0.05$ were considered statistically significant.

Results and Discussion

The results of the phytochemical screening, proximate and some vitamins as well as anti-nutritional analyses of *T. anguina* fruits are given in Table 1-3 respectively. Saponins, flavonoids and cyanogenic glycosides were the major phytochemicals identified in the fruit extract. Tannins and cardiac glycosides were also found to be slightly present. These phytochemicals exhibit diverse pharmacological and biochemical actions when ingested by animals (Amadi *et al.*, 2006). However, they are usually present in low concentrations in edible fruits and vegetables. Furthermore, food processing such as boiling reduces further the amount of these phytochemicals in plant products (Piorrock *et al.*, 1984). The major nutritional compositions of the fruit were found to include water, carbohydrates, proteins and vitamins C and A. The presence of proteins and carbohydrates does not only arise from the general presence of primary metabolites in biological materials. *T. anguina*, like other curcubits, is known to contain lectins, a complex of carbohydrates and proteins that display remarkable antigenic specificity (Anuradha and

Table 1: Phytochemical profile of *T. anguina*

Phytochemical	Result
Saponins	++
Tannins	+
Alkaloids	-
Cyanogenic glycosides	++
Cardiac glycosides	+
Flavonoids	++

Key: + = Slightly present; ++ = Clearly present; - = Absent

Table 2: Proximate compositions of *T. anguina*

Constituent	%Composition
Protein	1.85±0.04
Fibre	0.81±0.01
Moisture	93.15±0.05
Fat	0.23±0.04
Carbohydrate	3.48±0.04
Total ash	0.48±0.04

Values are mean±standard deviation of duplicate determinations

Table 3: Some vitamins and anti-nutritional compositions of *T. anguina*

Constituent	Composition
Vitamin C (mg 100 mL)	18.90±0.05
Vitamin A (ug 100 mL)	347.0±0.02
Oxalate (%)	0.58±0.12
Phytate (%)	0.11±0.02
Tannins (%)	0.02±0.05

Values are mean±standard deviation of duplicate determinations

Bhide, 1999). Although, the proximate compositions of *T. anguina* fruits show a fairly poor nutrient constitution when compared to other common vegetables such as *Amaranthus hybridus* (Nwaogu *et al.*, 2006) and *Vernonia amygdalina* (Ijeh *et al.*, 1996), the fruit contains adequate concentrations of vitamins C and A. This may justify the name, snake tomato since tomatoes generally have been reported to be excellent sources of vitamins (especially C and E), minerals and carotenoid pigments (sources of vitamin A) (Wright, 2002). Vitamin C has anti-infective properties, promotes wound healing, may boost the immune system and help to ward off infections, while vitamin A helps to maintain good sight and prevents certain diseases of the eye. Both vitamins in tomatoes, together with vitamin E have anti-oxidant properties and may protect against some forms of cancer (Wright, 2002). Meanwhile, the results of this study also revealed the presence, although at low concentrations, of oxalate (0.59%), phytate (0.11%) and tannins (0,02%). These are anti-nutrients, which could be toxic when consumed in an unprocessed food. However, at the present concentrations, they may not constitute major danger (Enechi and Odonwodu, 2003), provided the fruit is cooked before consumption.

The mean change in body weights of the animals during the period of treatment is shown in Fig. 1. Analyses of the cumulative changes in the weights of the animal groups showed that groups B and E animals lost significant ($p<0.05$) weights (102.5±10.61 g and

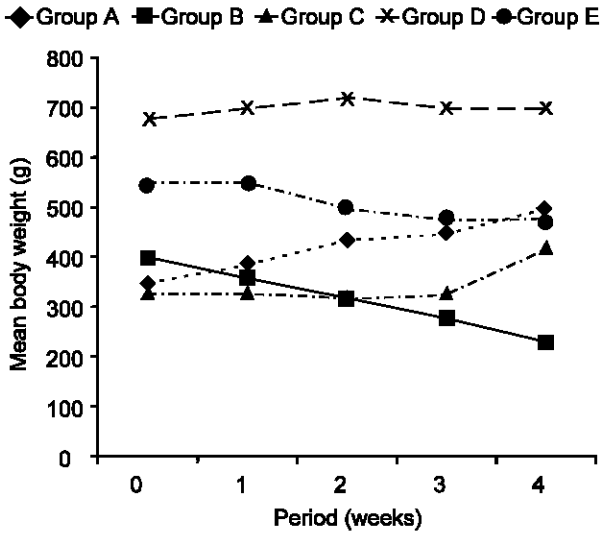

Fig. 1: Changes in mean body weights (g) of the animals

47.50±17.59 g respectively) when compared to the control group ($p<0.05$). On the other hand, groups C and D animals gained some weight (20.0±8.28 g and 25.0±6.12 g respectively) during the treatment period. However, in the later groups the appreciation in mean body weights were not statistically significant when compared with gain in weight observed among the control animals, 94.25±6.03 g ($p>0.05$). The loss in weight observed among the group B animals may be attributed to the direct consumption of the anti-nutrients present in the raw fruit. Similarly, the weight loss observed in the group E animals may indicate increased presence of the anti-nutrients or reduced availability of nutrients in the seeds compared to the other parts of the fruit. Meanwhile, the increase in weight observed amongst the groups C and D further gives credence to the belief that the fruit contains important nutrients and explains why man has continued to cook and serve *T. anguina* fruits as vegetable.

Fig. 2-6 show the changes in the individual liver function parameters with time. Serum total (TB) and conjugated (CB) bilirubin, and aspartate aminotransferase (AST), alanine aminotransferase (ALT), and alkaline phosphatase (ALP) activities were all sequentially raised during the experimental period in groups B and E animals. This translated to rise in levels of TB (67.86% and 61.29%), CB (87.88% and 70.15%), AST (29.87% and 28.57%), ALT (86.41% and 47.21%) and ALP (53.0% and 21.77%) in both groups respectively. A comparative study of the levels of these parameters in the animal groups at the end of the experimental period indicated that all the parameters were significantly increased in the sera of groups B and E animals in comparison with the control group (Table 4). This shows that the raw fruit and the fruit's seeds contain significant anti-nutritional factors, which have hepatocellular

Table 4: Liver function parameters of the animals at the end of experimental period

	Groups				
Parameter	A (n = 5)	B (n = 5)	C (n = 5)	D (n = 5)	E (n = 5)
Total Bilirubin (mg/dl)	0.75±0.71	2.80±0.14**	0.85±0.07	0.70±0.14	1.55±0.21*
Conjugated Bilirubin (mg/dl)	0.50±0.00	1.65±0.20***	0.40±0.00	0.23±0.07**	0.67±0.14*
Aspartate Aminotransferase (U/l)	7.90±0.90	30.20±0.21***	8.50±0.70	8.00±0.01	11.2±1.63**
Alanine Aminotransferase (U/l)	7.30±0.35	36.80±0.46***	5.00±1.71*	6.80±1.06	9.85±0.49***
Alkaline Phosphatase (U/l)	30.5±0.71	100.00±2.82***	35.75±2.50*	47.75±3.35**	67.75±3.18***

Values are mean±standard deviation; *, p<0.05; **, p<0.01; ***, p<0.001 with respect to control group; 'n' indicates number of animals in each group

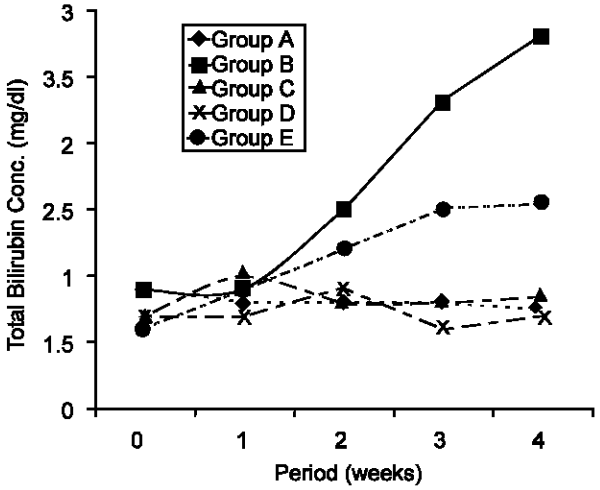

Fig. 2: Changes in serum total bilirubin concentrations (mg/dl) of the rabbits

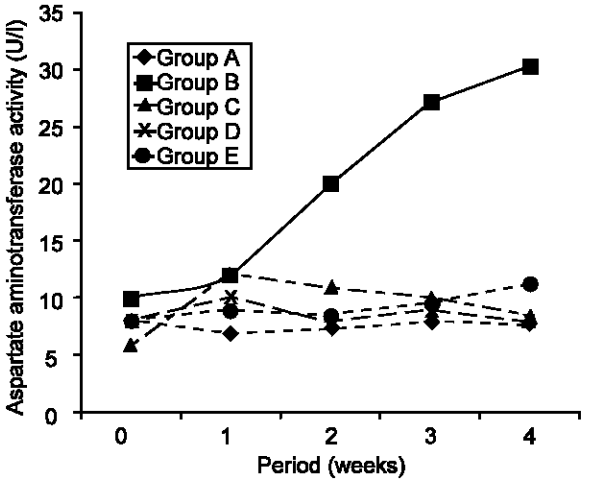

Fig. 4: Changes in serum aspartate aminotransferase activity (U/l) of the rabbits

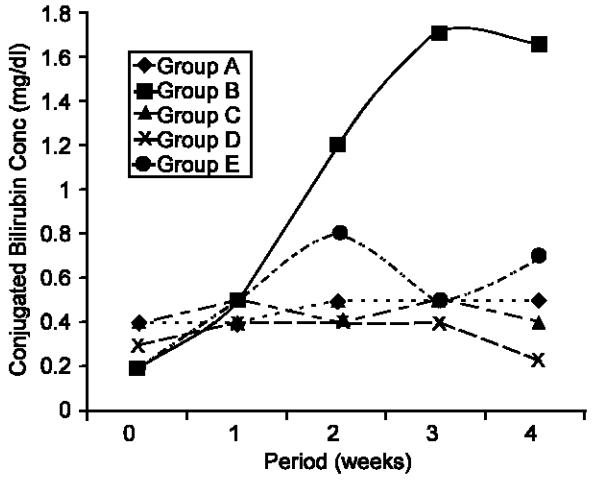

Fig. 3: Changes in serum conjugated bilirubin concentrations (mg/dl) of the rabbits

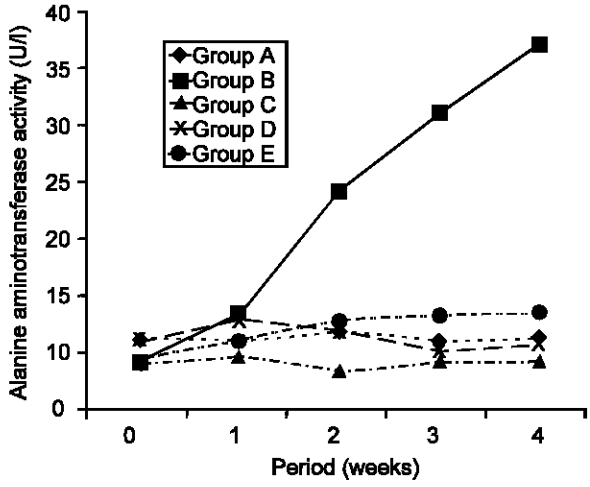

Fig. 5: Changes in serum alanine aminotransferase activity (U/l) of the rabbits

damaging effect. The effect was however, more pronounced with the ingestion of raw fruits. This agrees with earlier reports of toxicity and presence of potential toxins in the fruit (Anuradha and Bhide, 1999).

Serum TB and CB concentrations, as well as AST activity of the animals fed whole fruit (group C) were not significantly different from those of the control animals ($p>0.05$). On the other hand, the group C animals' serum ALT activity was significantly reduced ($p<0.01$), while their ALP activity was increased ($p<0.05$) in

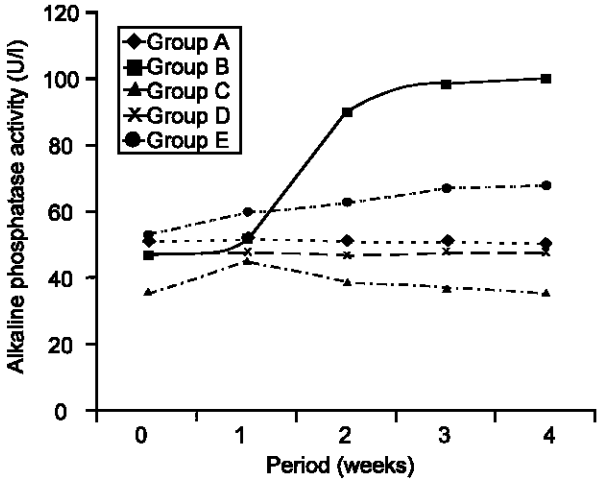

Fig. 6: Changes in serum alkaline phosphatase activity (U/l) of the rabbits

comparison with the controls. The levels of TB, AST and ALT were not significantly reduced ($p>0.05$) in the animals fed fruits whose seeds were removed (group D). Meanwhile, in this same group of animals the levels of CB and ALP were significantly ($p<0.01$) increased in relation to the control group. The slight differences observed in the liver function parameters upon ingestion of cooked whole fruit or fruit without seeds of *T. anguina* in relation to the control diet are not such that should discourage the consumption of the vegetable. The fact that changes in serum TB, AST and ALT were not significant strongly indicates the non-hepatotoxic effect of the vegetable when properly processed. This calls for adequate processing of the fruit before consumption given the significant hepatotoxicity observed with the raw fruits and seeds of the plant. Adequate processing methods have been shown to detoxify several vegetables that are ordinarily toxic in their raw state, including the more popular vegetables, *Telfaria occidentalis* (fluted pumpkin) and *V. amygdalina* (Ojiako and Nwanjo, 2006). This also brings more research points into limelight: what level of processing is necessary, and what will be the effect of such processing on the identified nutrients present in the fruits?

The present study confirms that *T. anguina* fruit contains important nutrients necessary for good human and animal health. But it may be toxic, just like several other fruits and vegetables, if consumed raw. Our findings indicate that when well processed before consumption, the fruit has no hepatotoxic effect, at least in rabbits and considering the level of poverty in Nigeria and the near-zero economic value of this vegetable, its cultivation and consumption should be encouraged.

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