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Nutritional potential of the under-utilized indigenous root tuber *Nymphaea petersiana* in Malawi

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Abstract. Demographic, health and micronutrient surveys in Malawi show that the nutritional status of the population is very poor, and has not improved during the last decade. Micronutrient malnutrition is particularly severe. For example, sub-clinical vitamin A deficiency was found in 60% of pre-school children, 38% of school children, 57% of women of childbearing age and 38% of men. Eighty percent (80%) of pre-school children, 27% of non-pregnant women, and 17% of men were anaemic. The national strategy to deal with this problem is multisectoral intervention involving many collaborating partners. One strategy being promoted is the utilization of readily available indigenous crops. One such indigenous crop is a wild water lily root tuber called "nyika" *(Nymphaea petersiana)*, which grows abundantly in the wild in the extensive swamps of the lower Shire river in Chikwawa and Nsanje districts. Nyika tubers are commonly used as sources of food in times of famine and/or poor harvests. Whole cooked tubers are sold in markets as a snack food, or the tubers are peeled, cut into small pieces, dried and ground into flour. The flour is cooked into nsima, the national dish, which is eaten with different types of stews and vegetables. Nutritional analysis of the Nyika tuber revealed it to be a good source of quality protein (about 8%), limiting only in lysine. Although the fat contact is low (1% crude fat), the predominant fatty acids in the tuber were essential fatty acids like oleic (47%), linoleic (37%) and linolenic (7%). More importantly was the high content of iron (100 mg/g of uncooked sun dried tuber). 100 mg of uncooked sun dried tuber supplies approximately 88% of the recommended daily allowance (RDA) of iron in children and 59% of the RDA in women. This amount of iron is higher than the reported values for other common root tubers such as cassava (78 mg/g), potato (7 mg/g) and sweetpotatoes (20 mg/g).

Introduction

Traditional, neglected and under-utilised crops provide a wide range of options to address the complex interface between food insecurity and health problems. They provide resources and mechanisms specific to the conditions and needs of different households and communities in specific agroecological areas. In particular, they contribute to resilient and reliable food security systems. They can improve the diets of poor farmers, they can provide new income-generating options suitable for many households. They also entail an integral approach to food security as cultural values and local food habits are respected and enhanced.

Rural people are often custodians of a rich diversity of crops and crop varieties that are usually neglected and under-utilized. Some of these crops are sometimes despised as "traditional" or "indigenous" in comparison to the handful of economically valuable crops that are relevant in agricultural trade and urban food habits. These traditional, neglected and under-utilised crops are excluded in agricultural policies and programmes, to the extent that their use and dignity have even declined among rural people themselves. Indigenous crops represent strategic crop genetic resources in household food security and nutrition, whilst providing many options for improving rural livelihoods and addressing evolving needs. They comprise crops and crop varieties that have strategic values and potential including advantageous adaptations to local agro-ecological constraints, such as drought, poor soils, and provision of superior nutritional sources. For example many neglected legume crops have high protein levels, and many leafy vegetables are excellent sources of micronutrients. Additionally, production of most of these crops is non-labour intensive.

During the last 2-3 years, drought, floods and insufficient agricultural inputs have resulted into famine in Malawi and other countries in the southern Africa Region. This food insecurity has been exacerbated by the HIV/AIDS pandemic. It is estimated that 16% of the population in the 15-49 years age group in Malawi has HIV. It is also important to recognise that people living with HIV/AIDS (PLWHA), even if they are asymptomatic, may have increased body metabolism, which increases their daily energy, protein and micronutrient requirements. An adequate and balanced diet for PLWHA is considered the only sustainable strategy to improve their health status and that of other people in general. More emphasis should be put on the use of indigenous and locally available foods with excellent amounts of micro-nutrients should be encouraged. Unfortunately many such foods have not been optimally exploited.

One such neglected and under-utilised root tuber is *Nymphaea petersiana,* locally known as Nyika which is an important water tuber in Southern Malawi. It grows abundantly in ponds and along the vast swamps of the Shire river. Nyika tubers are collected from waterways by small boats and canoes. These tubers are especially important as a food reserve during times of famine and poor harvests. Whole cooked tubers are commonly sold in markets as a snack, or the tubers are peeled, cut into small pieces, dried and ground into a powder. The powder is cooked into a thick porridge (locally known as nsima), which is served with different types of stews and/or vegetables. Nyika tubers may be consumed upto four or more times a week.

Although *N.petersiana* tubers are widely consumed in southern Malawi, there is no available data about their nutrient composition or potential for toxicity. This study was therefore, undertaken to determine the tuber's content of selected nutrients and antinutrients and to assess how it might be best utilized in the Malawian diet.

Materials and Methods

Sampling and Processing of Nyika Tubers. Tubers were collected from the Kademera and Chigumukire swamps of the Chikwawa and Nsanje districts of Southern Malawi. Care was taken to select only *N. petersian* tubers for analysis. *N. petersiana* is the only species of waterlilly in Malawi that have floating, sinuate-denated leaves (Flora Zambesiaca Managing Committee, 1960). Harvested tubers were put into cooler boxes with synthetic blue ice blocks and transported to a University of Malawi laboratory for analysis.

In the laboratory, the tubers were divided according to places of collection. Tubers were randomly assigned to one of two processing methods. In one method, the tubers were peeled, cut into small pieces, and sun-dried for 24-48 hours. The sun-dried tuber pieces were ground into powder using a cyclotec 1093 sample mill (Tecator, Hoganas, Sweden) and stored in plastic bottles at -4°C for further analysis. In the second processing method, the tubers were boiled in tap water for 45 minutes, peeled and freeze-dried. The freeze-dried tubers were ground into a powder and stored at -4°C.

Chemical analysis (Proximate composition). The moisture content of boiled/freeze dried (BFD) and uncooked sun-dried (UCSD) tubers was determined by drying in a

Brabender Moisture Tester oven (Brabender Instruments Inc., South Hackensack, NJ) at 135°C for 2 hours according to AOAC approved method 7.007 (AOAC, 1984). The tuber %nitrogen was determined by the Kjeldahl method 2.057 (AOAC, 1984) and converted into crude protein by multiplying by a factor of 6.25. Crude fat was determined by the chloroform-methanol extraction method of Phillips *et al.* (1997). Dietary fiber content was determined using a total dietary fiber assay kit (Sigma catalog no. TDF-100A). Ash content was determined by decomposition in a muffle furnace at 450-620°C for 12 hours (AOAC, 1984). The percentage of total crude carbohydrate was obtained by the difference {100-(% crude protein + % crude fat + % moisture + % ash)} (Muller and Tobin, 1980). A standard reference material SRM 1544, a frozen diet composite, purchased from the National Institute of Standards & Technology (NIST), was run simultaneously with the tuber samples for method validation.

Mineral analysis. Tuber mineral analysis was carried out on wet-ashed tubers according to AOAC method 7.009 (b) (AOAC, 1984). Tubers were analyzed for calcium, zinc, phosphorus, and iron. Calcium and zinc contents were determined by atomic absorption spectrophotometry (ASS) according to AOAC method 7.100 (AOAC, 1984), using a Perkin-Elmer, Model 2100 atomic absorption spectrophotometer. Phosphorus was determined colorimetrically using AOAC method 7.126 (AOAC, 1984). Iron was determined by AAS using a Perkin-Elmer ICP Emission Spectrometer Plasma 400 (Norwalk, Connecticut). A standard reference material (SRM), 1515 Apple Leaves, purchased from NIST was run simultaneously with the samples for method validation.

Amino acid analysis. The UCSD and BFD tuber samples were also subjected to amino acid analysis using AOAC method 982.30E (a, b, c) and F(AOAC, 1990).The amino acids were separated and quantified using a Beckman 6600 automated amino acid analyser, with a Beckman anion exchange column. Norleucine was used as an internal standard.

Fatty acid analysis. Fatty acids content of the tubers was also determined. The gravimetric method of Phillips *et al.* (1997) was used for extraction of total fat from the tuber samples. Extracted lipids were hydrolysed and esterified using 0.5N methanolic base (Supelco catalog no. 33080). The resulting fatty acid methyl esters (FAMES) were analysed by gas chromatography using a Shimadzu GC14A gas chromatograph with model AOC-14 Autoinjector and a Chromatopac C-R4AX processor (Shimadzu, Columbia, MD). FAMES were separated on a SP 2330 capillary column (Supelco, Bellefonte, PA) using temperature programming (from 60-100°C at 10°C/min, hold for 2 min at 100°C, then 10°C/min to 220°C). A set of short-chained C_4-C_{12} and long-chained C_{16}-C_{20} FAME standards were used to compare retention times of these standards to retention times of sample peaks eluted from the column as a means of tentatively identifying fatty acids in the tubers. FAMES were quantified using the internal standard technique, with heptadecanoic acid (17:0) as the internal standard.

Analysis of mono and disaccharides. The mono and disaccharide content of tuber samples was determined by high performance liquid chromatography (HPLC) following the method of Conrad and Palmer (1976) as modified by Johnson and Harris (1987).

Analysis of antinutritional factors. Tuber samples were also assayed for the possible presence of antinutritional factors. These factors were Trypsin and chymotrypsin inhibitors, cyanogenic glucosides, tannins, and phytates. The colorimentric procedure of Bradbury *et al.*(1991) was used to estimate the quantities of cyanogenic glucosides in the tuber samples. The UV-absorbance method of Kollipara and Hymowitz (1992) was used to determine trypsin and chymotrypsin inhibitor activity. The colorimetric procedure of Wheeler and Ferrel (1971) was used to estimate the phytate content of the samples. Tannins were analysed using the method of Burns (1971).

Statistical analysis. Analysis of variance (ANOVA) was used to determine effects of processing method on nutrient amounts and occurrence of antinutrients in the Nyika samples. A randomised complete block design model was chosen for statistical design of this study. Significant processing effects were tested at $P \leq 0.05$. The analysis was done using the SAS statistical package (SAS Inc., Copyright, 1989-1995, Cary, NC).

Results and Discussion

Proximate composition. The proximate composition of UCSD and BFD tubers is presented on both a wet and dry-weight basis in Table 1. UCSD tubers were found, on a dry weight basis, to be significantly higher in crude protein and ash content than BFD tubers. The results suggest that some of the tuber's proteins and minerals may have leached into water during boiling. There were no significant differences in the dietary fiber content of UCSD and BFD tubers, implying that the majority of the dietary fiber is composed of water-insoluble fiber components such as cellulose, hemicelluloses and lignin.

A comparison of the proximate composition of Nyika tubers and some cereals eaten in Africa indicated that except for sorghum (10.7%), the protein content of the tubers (8.0 and 8.1%, on a wet-weight basis, for BFD and UCSD tubers, respectively) was comparable to that of the other cereals including that of the Malawian staple maize (7.9%). The protein content was slightly higher than the protein content of African millets (unspecified) (7.5%), and that of milled and polished rice (7.0%). Since the tuber contains approximately 8% protein, it can, under famine conditions, be substituted for the Malawian staple maize without compromising the protein content of the diet.

A comparison of the proximate composition of Nyika tubers and other tubers eaten in Africa (Table 1) indicates that the tuber has a higher protein content than cassava (1.2 and 1.3% protein for bitter and sweet varieties, respectively), potato (1.7%), sweetpotato (1.6%), yams (3.2 and 1.5%, wild and domesticated, respectively), and the Egyptian *N.lotus* (5.2%). Staple foods with protein contents below 3% (like those of most of the root tubers cited in Table 1) do not meet the protein requirements of humans even when ingested in amounts supplying more than food energy requirements. On the contrary, a diet with an 8-10% protein content meets the protein requirements of adults, provided enough is eaten to supply food

Table 1: Comparison of the proximate composition of Nyika with other African root tubers.

Crop	Protein (%)	Fat (%)	Total CHO (%)	Fiber (%)	Ash (%)
Nyika[a] (BFD)	8.0	1.0	88.4	13.0	1.9
Nyika[a] (UCSD)	8.1	0.8	78.1	12.0	2.2
Cassava[b] bitter (raw)	1.2	0.2	35.7	1.1	0.9
Cassava[b] sweet (dried)	1.3	0.05	86.6	1.8	2.9
Potato[b] (raw)	1.7	0.1	18.9	0.6	1.6
Sweet Potato[b] (raw)	1.6	0.2	28.5	1.0	0.9
Yam[b], wild tuber	3.2	0.1	28.3	0.8	1.1
Yam[b] (African) tuber	1.5	0.1	26.5	0.9	0.9
N. lotus[b], root (Eghyptian)	5.2	0.2	29.5	1.0	1.1

[a]All Nyika values are on a wet-weight basis.
[b]Source: Food composition tables for use in Africa (Leung, 1968).

energy requirements (Cheftel *et al.,* 1985). This means that when eaten in sufficient quantities, *N. petersiana* tubers can meet the protein requirements of an adult.

The UCSD and BFD tubers had a higher fiber content than all the cereals and tubers listed in Table 1. High amounts of dietary fiber could, because of mineral binding to phytates and other dietary fiber components, reduce the bioavailability of zinc, iron and calcium (Zapsalis and Beck, 1985). Other dietary fiber components, specifically condensed tannins, are known to inhibit digestive enzymes, lowering the bioavailability of proteins and starches (Tamir and Alumot, 1969; Romero and Fyan, 1978). Dietary fiber has also been associated with a number of therapeutic effects. Increased consumption of dietary fiber increases excretion of bile acids in humans, which is associated with decreased risk of colon cancer (Haack *et al.,* 1998). A number of plant polyphenolic compounds have been shown to inhibit LDL oxidation (Vinson *et al.,*1995; Kerry and Abbey, 1997). LDL oxidation is thought to be an early precipitating event in the development of cardiovascular disease in humans (Berliner *et al.,* 1995).

Amino acid composition. The nutritional quality of a food protein depends on the kinds and amounts of amino acids it contains, and represents a measure of the efficiency with which the body can utilize the protein. A balanced or high quality protein contains essential amino acids in ratios that are sufficient to meet human needs. This can be determined by comparing the amino acid content of a protein to a FAO/WHO reference pattern (Cheftel *et al.*, 1985). The essential amino acid profiles of UCSD and BFD are shown in Table 2. The profiles indicate that the tuber is well balanced in essential amino acids except for lysine. Based on their lysine content, UCSD and BFD tubers had amino acid scores of 91 and 84, respectively. Knowledge of amino acid scores helps in estimating the complementary value of different proteins in a food mixture. In this case, the amino acid scores would help in deciding what cereals or legumes would complement the tuber if it had to be used as a weaning food, or what side dishes should be eaten with the tuber to complement the limiting amino acid lysine. Two possible choices available to Malawians are groundnuts (peanuts and bambarra nuts). Both are high in lysine and deficient in the sulfur amino acids (CFO, 1970) complementing Nyika proteins which are low in lysine but contain more than the FAO/WHO requirement of cysteine and methionine (Table 2).

Table 2: Comparison of essential amino acid content of *N.petersiana* (Nyika) tubers to a FAO reference pattern.

Amino acid	BFD tubers[a]	UCSD tubers[a]	FAO/WHO (1990) reference pattern[b]
Threonine	39 ± 0.5	39 ± 0.5	34
Cys. & Methionine	45 ± 0.5	46 ± 1.0	25
Valine	52 ± 1.7	52 ± 1.0	35
Leucine	86 ± 0.5	84 ± 1.0	66
Isoleucine	46 ± 2.4	45 ± 0.5	28
Phe.& Tyr.	101 ± 1.3	99 ± 0.5	63
Lysine	49 ± 1.5	53 ± 0.5	58
Tryptophan	26 ± 6.0	29 ± 1.0	11

[a]Reported values are in mg of amino acid/g of crude protein and are means and standard deviation of two determinations.
[b]Based on prechool child data from FAO/WHO/UNU (1985).
Source: FAO/WHO/UNU (1985).

Minerals. Statistical analysis revealed that there were significant differences in the calcium, phosphorus, and iron content of UCSD and BFD tuber samples (Table 3). Approximately 20% of the phosphorus and 80% of the iron in the tubers presumably leached into the cooking water during boiling of BFD tubers. UCSD tubers appear to be an excellent source of iron. According to our calculations, a one hundred gram serving of the UCSD tubers would, on a wet-weight basis, supply approximately 88% of the USRDA for iron for children six months to 10 years of age and 59% of the USRDA for iron for women 11 to 50 years of age (NRC, 1989).

UCSD tubers had a higher iron content on a wet-weight basis (88.0 m/g) than reported values for cassava (78.0 m/g) and sweetpotato (20 m/g). A comparison of the mineral content of *N. petersiana* with other African root tubers (which are also eaten in Malawi) indicated that BFD tubers contain more calcium (1300 m/g) than cassava (480 m/g wild and 1210 m/g domesticated), potatoes (110 m/g), sweetpotatoes (330 m/g), and yams (520 m/g wild and 690 m/g domesticated). Phosphorus content (2200 µg/g and 2600 m/g for BFD and UCSD**,** respectively, on a wet-weight basis) was also higher than the reported values for the root tubers (Leung, 1968).

Other nutrients. Nyika tuber samples were found to contain a preponderance of polyunsaturated (approximately 30% linoleic and 6% linolenic acid) and monosaturated (43% oleic acid) fatty acids (Table 4). Presence of high levels of unsaturated fatty acids is nutritionally desirable. However, unsaturated fatty acids are much more susceptible to oxidation than their saturated counterparts, therefore, exposure of tuber material to oxygen, light, and high temperatures should be minimized during processing and storage to prevent autoxidation of tuber lipids. Despite their high carbohydrate content, BFD and UCSD tubers were found to contain small amounts of the water-soluble sugars fructose (2.4 and 4.8mg/100g), glucose (3.0 and 6.0 mg/100g), and sucrose (0.05 and 0.35 mg/100g). There were significant differences in the content of glucose and fructose sugars between the two processing methods with the UCSD tubers having about twice as much of the two reducing sugars as BFD tubers. This might have been due, once again, to leaching losses of water-soluble mono and disaccharides from BFD tubers.

Antinutrients. The tannin content of *N. petersiana* was 1% and 1.5% in BFD and UCSD samples, respectively, which is lower than that found in sorghum (6.88%) from Uganda (Mukuru *et al.*, 1992). Boiling reduced the tannin content of the tubers which is in agreement with Price *et al.* (1980) who found that they could remove 33% of the tannins in sorghum grain by boiling the grain for 10 minutes. When fed to rats, the biological value (BV) of sorghum grain is significantly reduced by the presence of as little as 0.7% tannin (Muindi and Thomke,

Table 3: Mineral composition of boled/freeze-dried (BFD) and uncooked/sun-dried (UCSD0 *N. petersiana* (Nyika) tubers[a].

Tuber sample	Calcium (inµ/g of tuber)	Phosphorus (in µ/g of tuber)	Zinc (in µ/g of tuber)	Iron (in µ/g of tuber)
BFD tubers (wet-weight basis)	13000±49[a]	2200±27[c]	20±3[a]	20±9[b]
UCSD tubers (wet-weight basis)	928±48[c]	2600±1[b]	22±3[a]	88±43[a]
BFD tubers (dry-weight basis)	1309±48[a]	2215±28[c]	20±3[a]	20±9[b]
UCSD tubers (dry-weight basis)	1040±55[b]	2914±1[a]	25±3[a]	99±48[a]

[a]Reported values are means and standard deviations of eight determinations.

Table 4: Fatty acid composition of boiled/freeze dried (BFD) and uncooked/sun-dried (UCSD) *N. petersiana* (Nyika) tubers[1].

Tuber sample	%Palmitic acid (16:0)	%Stearic acid (18:0)	% Oleic acid (18:1)	%Linoleic acid (18:2)	%Linoleic acid (18:3)
BFD tubers (wet-weight basis)	21.0±0.6[a]	2.0±0.3[a]	42.0±1.0[a]	30.0±1.2[a]	6.0±0.5[a]
UCSD tubers (wet-weight basis)	19.0±1.3[b]	2.2±0.4[a]	43.0±1.9[a]	30.0±2.5[a]	6.5±0.4[a]

[1]Reported values are means and standard deviations of eight determinations.
[a,b]Means in the same column with different superscripts are significantly different at the P = 0.05 level.

1981), which suggests that the tannin content of the tubers is high enough to have significant negative consequences on nutrient bioavailability. Removing the tubers from their outer husks and then boiling them, may be a way of lowering the tannin content sufficiently to prevent such problems.

The phytate content of *N. petersiana* was 3.9µg/g and 5.4µg/g of tubers (BFD and UCSD, respectively, on a wet-weight basis). These values were very low compared to those found by Ferguson *et al*. (1988) for the Malawian staple white maize flour (in mg/100g) (211); cooked maize flour (55); unrefined maize flour (792); cassava (59); and sweetpotato (12). This indicates that the phytate content of *N. petersiana* tubers is so low to be of nutritional concern when compared with other foods commonly eaten in Malawi.

Nyika flour samples were also analysed for hydrogen cyanide, a compound released from cyanogenic compounds during heating. Boiled *N. petersiana* tubers have a slightly bitter taste (like that of some bitter cassava varieties) and this prompted us to analyze the tubers for cyanogenic glucosides common in bitter cassava. Hydrogen cyanide levels in both UCSD and BFD samples were below the limit of detection for the assay (LOD<16 nmoles/L), an indication that cyanogenic glucosides, if present at all in the tuber, are in negligible quantities and of little toxicological concern.

Trypsin and chymotrypsin inhibitor activity were significantyly greater in UCSD than in BFD tuber samples. The trypsin inhibitor (TI) activity in the tubers was reduced by a factor of about seven and the chymotrypsin inhibitor (CI) activity by a factor of five, by boiling (data not presented). Protease inhibitor activity in UCSD tuber samples was significantly greater than in uncooked soybeans that were purchased from a local health food store and assayed simultaneously for TI and CI activity. CI activity in BFD samples was slightly less (50 CIU/g of tuber) than CI activity in uncooked soybeans (52 CIU/g of seed) while TI activity (55 TIU/g of tuber) was less than half of the TI activity of uncooked soybeans (113 TIU/g of seed). These values indicate that although trypsin and chymotrypsin inhibitors were susceptible to moist heat; they were not fully inactivated even when the tubers were immersed in boiling water for 45 minutes.

Conclusions

Based on data presented here, Nyika tubers should be initially sun-dried rather than boiled to retain iron and maximize protein quality. Sun-dried tuber material should however, be consumed only after thorough cooking to ensure that trypsin and chymotrypsin inhibitors are inactivated. Future research should focus on the analysis of the nutrient and antinutrient content of Nyika based snacks and meals as they are prepared and eaten in Malawi. Research efforts should be expanded to include analysis of other essential nutrients and additional antinutrients.

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References

AOAC. 1984. Official Methods of Analysis, 14th Ed. Association of Official Analytical Chemists, Washington, DC.

AOAC. 1990. Official Methods of Analysis, 15th Ed. Association of Official Analytical Chemists, Washington, DC.

Bradbury, J.H., Egan, S.V. and Lynch, M.J. 1991. Analysis of cyanide in cassava using acid hydrolysis of cyanogenic glacosides. J. Sci. Food Afric., 55, 277-290.

Mukuru, S.Z., Butler, L.G., Rogler, J.C., Kirleis, A.W., Ejeta, G., Axtell, J.D. and Mertz, E.T. 1992. Traditional processing of high tannin sorghum grain in Uganda and its effect on tannin, protein digestibility, and rat growth. J. Agric. Food Chem. 40:1172-1175.

Wiersema, J.H. 1987. A Monograph of Nymphaea Subgenus Hydrocallis (Nymphaeacae). Systematic Botany Monographs 16: 1-112.

Zapsalis, C. and Beck, R.A. 1985. Food Chemistry and Nutritional Biochemistry. John Wiley & Sons, New York, NY, p. 1002.