Nutritional and anti-nutritional composition of cassava leaf protein concentrate from six cassava varieties for use in aqua feed

A. Oresegun1, O. A. Fagbenro2, P. Ilona3 and Edah Bernard1*

Abstract: Leaves from six varieties of cassava (*Manihot esculenta* crantz) cultivated in Nigeria were harvested and analysed for their nutritional composition and anti-nutritional factors using standard analytical techniques. The leaves were further processed into cassava leaf protein concentrate. Results obtained indicated highest crude protein levels, *β*-carotene levels and lipid levels of 48.85 ± 0.45, 816.92 ± 8.80 and 13.27 ± 0.06, respectively, in variety 419 and lowest crude protein levels and *β*-carotene levels of 40.19 ± 0.08 and 298 ± 5.74 in variety 326. However, ash, moisture and carbohydrate levels for all six varieties were relatively the same. Mineral compositions for all varieties were also fairly similar. Hydrogen cyanide levels were significantly low (*p* > 0.05) in variety 419 with a value of 0.98 ± 0.05. However, phytate and tannin levels for all six varieties were not significantly different (*p* > 0.05). It can be concluded from this study that cassava variety 419 had the highest potential for use in aqua feed production.

Subjects: Agriculture; Agriculture and Food; Fisheries Science; Oceanography

Keywords: fish feed; cassava leaves; anti-nutritional factors; variety 419; *β*-carotene

ABOUT THE AUTHORS

Nigeria produces more cassava than any other country in the world. It was estimated that in 2010, Nigeria’s production of cassava reached 37.5 million tonnes and that the country has consistently been ranked as the world’s largest producer of cassava since 2005 (FAOSTAT, 2012). Its production requires minimum labour and inputs, and remains the most important food security crop for millions of Nigerians. One hectare of cassava plantation contains between 7,000 and 8,000 stands. Cassava leaf takes about 6–8% of the weight of each stand. Therefore, an established weight of leaves per hectare is 0.69–0.92% MT. This implies some 2.6–3.5 million MT of leaves are unutilized in Nigeria. In addition, their low production cost is emphasized, since the leaves are rather considered as residues and they do not compete with the roots, which are the main commercial product from cassava. This is where value addition comes in.

PUBLIC INTEREST STATEMENT

This research work was carried out to add value to the millions of cassava leaves discarded as waste annually in Nigeria.
1. Introduction
Cassava is extremely reliable to grow, especially on sloping rain-fed soils of low fertility, survives
drought periods and grows well with limited supplies of water. In addition, it is tolerant of acid soils
and yields well on marginal soils without excessive use of costly inputs. These qualities have en-
deared cassava to resource-poor farmers (Phuc et al., 2000).

In Nigeria, considerable amount of cassava leaves are generated annually and readily available as
a by-product at the time of harvesting the roots. While cassava leaf protein is low in sulphur and
amino acids (Kobawila, Louembe, Keleke, Hounhouigan, & Gamba, 2005), the content of most other
essential amino acids is higher than in soya bean meal (Ly & Samkol, 2001). The high protein content
and a relatively good profile of essential amino acids are reasons for believing that cassava leaves
could be a potential protein source for monogastric animals.

Cassava leaves are a significant source of potential alternative protein resource for both humans
and animals (Fasuyi, 2005). The leaves, depending on the varieties, are rich in minerals, proteins,
vitamin and carotenes (Adewusi & Bradbury, 1993). However, it also has some anti-nutritional and
toxic substances. These substances interfere with digestibility and uptake of the nutrients, and they
might present toxic effects depending on the amount in which they are consumed (Wobeto, Corrêa,
Abreu, Santos, & Pereira, 2007).

Cassava and cassava by-products are widely used and represent the basic diet of about 500 mil-
lion people in the world (Chavez et al., 2000). The use of the cassava leaf meal (CLM) may supply a
positive balance for the nutritional quality because they present higher contents of proteins, vita-
mins, minerals and fibre (Corrêa et al., 2004; Ngudi, Kuo, & Lambein, 2003). In addition, their low
production cost is emphasized, since the leaves are rather considered as residues and they do not
compete with the roots, which are the main commercial product from cassava. The main objectives
of this research are to investigate the nutritional composition and anti-nutritional factors of six vari-
eties of cassava leaf and their protein concentrate. This will also provide knowledge on the nutri-
tional potential of cassava and its use in the aqua feed industry (Figure 1).

2. Materials and methods

2.1. Collection and processing of cassava leaves for cassava leaf protein concentrate
Leaves of six cassava varieties namely variety 326, variety 505, variety 419, variety TME 1, variety
1,368 and variety 30,572 were freshly collected from the International Institute for Tropical
Agriculture (IITA) Ibadan Nigeria and transported in plastic bags. The harvested leaves were weighed
and washed to pulping with a leaf pulping machine. This was followed by crushing in water (1:3 w/v
at a pH 8.5) and filtered as described by Fellows (1987). The separated leaf juice was then heated at

Figure 1. Anti-nutritional
factors of six varieties of
cassava.
about 90–100°C for 10 min to coagulate leaf protein concentrate. The coagulated protein fraction was thereafter separated by a method of filtration and subsequently dried at atmospheric temperature. The dried cassava leaf protein concentrate (CLPC) was then milled using a laboratory hammer miller and kept in an airtight container prior to chemical analysis.

2.2. Nutritional composition of cassava leaves
The proximate composition of variety 326, variety 505, variety 419, TME 1, variety 1,368 and variety 30,572 were determined for moisture, ash, crude fat, total carbohydrate and total β-carotene as described by (Association of Official Analytical Chemist Official [AOAC], 2000). Thereafter, the nitrogen was determined by the Micro-Kjeldahl method.

2.3. Determination of mineral content
Mineral composition after wet digestion with a mixture of sulphuric acid, nitric and perchloric acid was determined using the atomic absorption spectrometer (AAS) (Buch Scientific, East Norwalk, CT 06855, USA) for calcium (Ca), magnesium (Mg), manganese (Mn), iron (Fe), copper (Cu) and zinc (Zn), while potassium (K) and sodium (Na) were determined using flame photometry (Table 3).

2.4. Anti-nutritional factor analysis
Variations in the chemical composition and inherent anti-nutritional substances in the different cassava varieties may restrict usage. The principal problems that could undermine its potential include high fibre content and anti-nutrients typified mainly by cyanide, tannin and phytate (Table 2).

2.5. Statistical analysis
The entirely randomized statistical design was used, on a factorial scheme of 6 + 1 + 3, which is, six varieties, a specific plant age and three replicates. Data were analysed by descriptive analysis and one-way analysis of variance (ANOVA). SPSS (version 16.0) statistical software package (SPSS, Chicago, USA) was employed in the analysis. Differences were considered significant at an alpha level of 5%.

3. Results and discussion
The nutritional content and anti-nutritional factors of the six varieties of cassava were investigated. The CLPC was prepared under environmental conditions. The variance analysis and significant differences were observed at 5% probability, by the F-test, in all other parameters studied. Table 1 shows the result of the chemical composition for the different CLPC in percentages owing to their nutritional values with variety 419 recording the highest percentage in crude protein (CP) concentration with a value of 48.85 ± 0.45, while variety 505 had the lowest CP concentration of 40.19 ± 0.42 and differs significantly (p < 0.05) from the rest of the varieties.

Average values for moisture content in the CLPC (g 100 g⁻¹) were highest in variety 505 with a value of 9.55 ± 0.1, while variety 30,572 had the lowest moisture content with a value of 6.27 ± 0.20. The high values of ash content (5.56 ± 0.03) in variety 30,572 indicate a high mineral content in it than in variety 1,368 with an ash content of 3.4 ± 0.01. The values do not correspond with that reported by Frederick (2008).

Total β-carotene levels were equally highest in variety 419 with a value of 816.92 ± 8.80 but show a remarkable decrease in variety TME 1 and variety 30,572 with equal concentrations of 298 ± 5.74. This study demonstrates a considerable variation in the chemical composition among leaves of different cassava varieties which is in agreement with earlier reports (Ly & Samkol, 2001; Ravindran, 1990). The variation may be due to differences in plant development as well as genetic make-up of the different varieties. Table 2 shows the anti-nutritional factors which include cyanide, phytate and tannin concentrations for the different CLPCs.

Toxicity problems that affect nutritive value of cassava leaves can be reduced by traditional preparation methods such as drying, pounding and long periods of boiling (Ajibade, Balogun, Afolabi, &
Kupolati, 2006; Aletor & Adeogun, 1995; Fasuyi, 2005; Lewis & Fenwick, 1987). Sun-drying is an inexpensive effective method of preserving surplus micronutrient-rich foods (Tontisirin, Nantel, & Bhattacharjee, 2002). Variety 1,368 had the highest anti-nutritional factors with cyanide, phytate and tannin concentrations of 6.85 ± 0.0, 1.73 ± 0.0 and 1.55 ± 0.0, respectively. Cyanide concentrations were lowest in variety 419 with a value of 0.98 ± 0.05. Variety 30,572 recorded the lowest phytate and tannin concentrations of 1.14 ± 0.0 and 0.77 ± 0.0 respectively. These levels are within the range reported by (Awoyinka, Abegunde, & Adewusi, 1995 and Câmara & Madruga, 2001) with cyanide levels ranging from 5.3 to 80 mg/100 g dry matter (DM). However, values were not significantly different between groups of varieties (p > 0.05). This variation is probably due to the genetic differences among the cultivars, the plant ages, the leaf maturity and soil fertility.

The CLPC from the analysed six varieties could well be classified as non-toxic because generally their cyanide contents fell below 10 mg 100 g−1 powder (Ikediobi, Onyia, & Eluwah, 1980). The lethal dose (LD) of cyanide oscillates from 0.5 to 3.5 mg kg−1 human body weight (Wogan & Marletta, 1993). However, it is worth emphasizing that the chronic toxicity is due to the consumption of lower cyanide doses at longer timer intervals (Osuntokun, 1981).

This study further revealed a considerable variation in the mineral composition among the six different cassava varieties. Table 3 shows the average mineral contents in DM. Iron content in variety 419 was highest (458.08) compared to variety 505 with a value of 321.32. These results are in line with the result analysed by Kadashi (2005). Iron is required for the synthesis of haemoglobin and myoglobin, which are oxygen carriers in the blood.

In conclusion, among the varieties investigated in this study, variety 419 had the lowest contents for most anti-nutrients (cyanide, phytate and tannin inhibitors) and consequently the highest content for most nutritional values like the CP value, while on the other hand, varieties with most highest anti-nutritional contents tends to exhibit the lowest nutritional contents indicating an inversely
proportional relationship between anti-nutritional factors and nutritional contents of the six varieties. From this study, therefore, variety 419 exhibited highest potential for use both for human consumption, fish feed production and livestock feed production. Though, the other varieties analysed might also be relevant in the feed industry at a non-toxic amount.

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### Competing interests

The authors declare no competing interest.

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### Table 3. Mineral composition of six different varieties of cassava

<table>
<thead>
<tr>
<th>Cassava variety</th>
<th>Ca (%)</th>
<th>Mg (%)</th>
<th>K (%)</th>
<th>Na (ppm)</th>
<th>Mn (ppm)</th>
<th>Fe (ppm)</th>
<th>Cu (ppm)</th>
<th>Zn (ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>326</td>
<td>0.68</td>
<td>0.02</td>
<td>0.13</td>
<td>32.78</td>
<td>76.69</td>
<td>356.96</td>
<td>22.33</td>
<td>36.25</td>
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<tr>
<td>505</td>
<td>0.9</td>
<td>0.03</td>
<td>0.15</td>
<td>33.45</td>
<td>124.21</td>
<td>321.32</td>
<td>18.34</td>
<td>62.06</td>
</tr>
<tr>
<td>TME 1</td>
<td>0.85</td>
<td>0.05</td>
<td>0.14</td>
<td>33.35</td>
<td>152.71</td>
<td>371.05</td>
<td>20.7</td>
<td>56.32</td>
</tr>
<tr>
<td>419</td>
<td>0.71</td>
<td>0.03</td>
<td>0.16</td>
<td>32.75</td>
<td>77.85</td>
<td>458.08</td>
<td>17.03</td>
<td>52.99</td>
</tr>
<tr>
<td>30,572</td>
<td>1.2</td>
<td>0.07</td>
<td>0.12</td>
<td>33.18</td>
<td>141</td>
<td>343.06</td>
<td>23.04</td>
<td>68.95</td>
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</tbody>
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