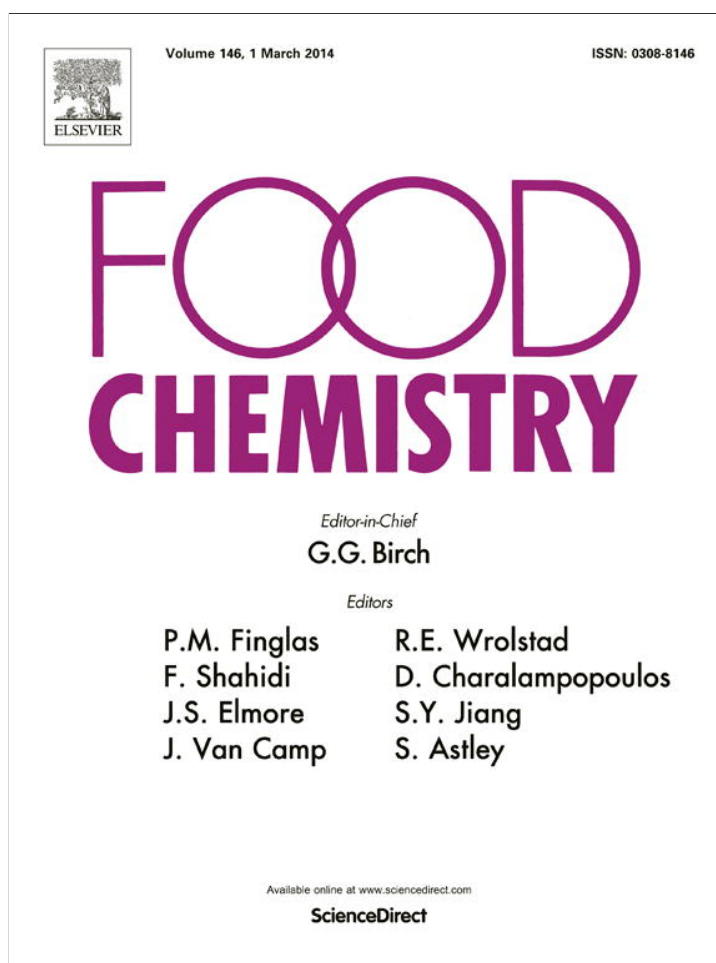




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## Effect of delayed harvesting and pre-treatment methods on the antinutritional contents of trifoliate yam flour

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## ABSTRACT

Effects of delayed harvesting and pre-treatment methods on the anti-nutritional contents of trifoliate yam flour were examined. Trifoliate yam tubers were washed, peeled, sliced and subjected to pre-treatment methods, such as soaking, pre-cooking and blanching/soaking. The phenols, phytate, oxalate, tannin and alkaloid profiles of the flours were evaluated and the values of phenols, tannin, oxalate and phytate contents were 0.02–0.32, 0.04–0.53, 0.11–4.32 and 0.20–1.05 mg/100 g, respectively. The predominant alkaloids in trifoliate yam flour were dioscorine and dihydrodioscorine. The white trifoliate yam flour had higher levels of anti-nutrients than the yellow trifoliate yam flour. Alkaloid contents of trifoliate yam flour increased slightly with delayed harvesting periods. Blanching/soaking method drastically reduced the anti-nutrient contents of trifoliate yam flour than other methods.

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## 1. Introduction

*Dioscorea dumetorum* Pax (family *Dioscoreaceae*) is commonly known as bitter yam or cluster yam. It originated wildly throughout Africa, predominantly in the tropics (Nimenibo–Uadia, 2003) and occurs in both wild and cultivated forms. Yam tubers are known to contain different toxic substances that affect both human and animals when they are consumed, despite their high nutritional values (Polycarp, Afoakwa, Budu, & Otoo, 2012). The antinutritional factors in yam include phenol, tannin, oxalate, phytate and alkaloids. The bitter tastes in some yam tubers have been attributed to high tannin content and the major disadvantage of phenolic compound is the browning reactions that cause undesirable darkening of the tuber when cut and exposed to the atmosphere (Ngoddy & Onuoha, 1985). The degree of browning is proportional to the amount of polyphenol oxidase activity (Ngoddy & Onuoha, 1985). Oxalate occurs primarily in form of soluble oxalate, insoluble calcium oxalate (raphides), or a combination of the two forms (Libert & Franceschi, 1987). The intense irritation of the skin and mucous membrane (e.g. in the mouth) when they come in contact with mucilage is due to the presence of calcium oxalate crystals (raphide) which are thought to be the most obvious toxicant in yams (Sakai, 1983). Phytic acids is a hexaphosphate derivative of

inositol, it is insoluble and it is not absorbed by humans intestinal mucous because of lack of endogenous enzymes system, such as phytase in humans and monogastric animals (Osagie 1992). Phytates bind minerals in the gastrointestinal tract, making dietary minerals unavailable for absorption and utilization by the body (Plaami, 1997). The alkaloids level may vary depending on cultivated species and cultural practices. At low level they impact bitterness to the tuber tissue and at high level, as found in some wild species, they can cause burning sensation in the mouth and throat, vomiting and diarrhea (Oke, 1985). Oke (1985) reported further that severe cases of alkaloid poisoning can cause general paralysis of the central nervous system, which can also result in seizures and convulsion. Alkaloids are water soluble and must be disipitated by boiling or soaking in water. These alkaloids are used in the pharmaceutical industries for the production of steroids, heart stimulant, shampoo, a cure for snake bites, insecticides, etc (Oke, 1985). The antinutritional contents of food are significantly affected by age, cultivar, the geographic locality of the plant and storage condition after harvesting (Polycarp et al., 2012; Yang & Lin, 2008). The antinutritional composition of trifoliate yam tubers contributed greatly to the under-utilization of the yam coupled with the hardening process undergone after harvesting. The tubers are not harvested and stored like other yam species but are left in the ground after maturity until they are needed as food. The effects of delayed harvesting and pre-treatments methods on the antinutritional contents of trifoliate yam have not been widely studied. However, various methods of reducing anti-nutritional contents of

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yam have been studied (Bhandari & Kawabata, 2006; Ezeocha, Ojmelukwe, & Onwuka, 2012; Medoua, Mbome, Agbor-Egbe, & Mbofung, 2007; Shanthakumari, Mohan, & Britto, 2008; Wanasundera & Ravindran, 1992). Therefore, the present paper reports the effect of delayed harvesting and pretreatment methods on the anti-nutritional composition of trifoliate yam flour. This study is necessary to give information on the levels of anti-nutrients at different harvesting periods and how to reduce them for human consumption. This information would aid processing of trifoliate yam into flour thereby improving the utilization of the yam and reducing wastage of the crop during its season.

## 2. Materials and methods

### 2.1. Materials

The trifoliate yam sets of two cultivars (white and yellow) were collected from Esa-Oke farm settlements, Osun State, Nigeria.

### 2.2. Samples preparation

Trifoliate yam setts weighing 850–900 g were planted in mounds with 10 per row and spacing of  $1 \times 1$  m on a plot of land. Thirty samples were planted per each cultivar. The planting was done on 20th March 2010 and sprouting of some of the yam setts occurred on 26th of April 2010. These trifoliate yam setts were marked and used for the study. The plot of land was kept weed-free manually at a month interval after planting. There was no application of chemicals of any kind applied either as fertilizer, pest control or herbicides. The yam tubers were harvested in a month interval starting from November 26th 2010 to March 26th 2011. The experiment was repeated in November 2011 to March 2012.

### 2.3. Preparation of raw flour

The freshly harvested yam tubers were washed, drained and peeled. The peeled tubers were sliced and dried in the hot air oven at 60 °C for 48 h. The dried chips were milled into flour with hammer mill and sieved with 600 µm sieve size. The flour samples were sealed in polythene bag.

### 2.4. Preparation of soaked raw flour

The freshly harvested yam tubers were washed, drained and peeled. The peeled tubers were diced, soaked in water for 1 h and dried in the hot air oven at 60 °C for 48 h. The dried chips were milled into flour with hammer mill and sieved with 600 µm sieve size. The flour samples were sealed in polythene bag.

### 2.5. Preparation of blanched/soaked flour samples

The freshly harvested yam tubers were washed, drained and peeled. The peeled tubers were diced, blanched (60 °C) for 10 min in water bath, soaked for 12 h and dried in the oven (60 °C) for 48 h. The dried chips were milled into flour with hammer mill and sieved with 600 µm sieve size.

### 2.6. Preparation of pre-cooked samples

The freshly harvested yam tubers were washed, peeled, diced and pre-cooked for 10 min in water bath maintained at  $100 \pm 2$  °C. The samples were dried in an oven set at 60 °C for 48 h. The dried chips were milled into flour with hammer mill and sieved with 600 µm sieve size.

### 2.7. Tannin determination

Milled samples (200 g) was extracted in 10 ml 70% aqueous acetone for 2 h at 30 °C in a water bath. A Gallenkamp orbital shaker at 120 rpm was used. Diethyl ether containing 1% acetic acid was used to extract the fats and the pigments in the sample. Tannin was then determined as total phenols in 0.05 ml aliquot in test-tubes by the addition of distilled water to 1 ml mark and the addition of 0.5 ml Folin Ciocalteu reagent (Sigma) and 2.5 ml sodium carbonate. The absorbance of the solution was measured at 725 nm after 40 min using the method of Makkar & Goodchild (1996). The tannin equivalent in the form of phenol will be calculated from a standard curve.

### 2.8. Phytate determination

The phytin content was quantified by adding 8 g of the milled sample soaked into 200 ml of 2% HCl and allowed to stand for 3 h. The extract was filtered through a double layer filter paper and 50 ml of the duplicate samples of the filtrate was pipette into 400 ml beaker. Ammonium thiocyanate (0.3%w/v) (10 ml) was used as an indicator and distilled water (107 ml) was added to obtain a pH of 4.5. Ferrous chloride solution containing 0.00195 gm Fe/ml was titrated against the solution of the tests samples until a brownish yellow colouration persisted for 5 min. Phytin phosphorous was determined using the relationship that each milligram of iron is equivalent to 1.19 mg of phytin phosphorous. The phytin content was calculated by multiplying by a factor of 3.55 according to Dairo (2008).

### 2.9. Oxalate determination

Oxalate content was determined using the method of Nwin-nuka, Ibeh & Ekeke (2005). The flour sample (1.0 g) was extracted thrice by warming (40–50 °C) and stirring with a magnetic stirrer for 1 h with 20 ml of 0.3 N HCl. The combined extracts was diluted to 100 ml with water and used for the total oxalate estimation. About 5 ml of each extract was made alkaline with 1 ml of 5 N  $\text{NH}_4\text{OH}$ . This was then made acidic with glacial acetic acid and phenolphthalein indicator (2–3 drops) was added, excess decolourizes. Then, 1 ml of 5% calcium chloride was added and the mixture was allowed to stand for 3 h after which it was centrifuged (IEC Centra GP8) at 140.868 g for 15 min. The supernatant was discarded while the precipitate was washed thrice with hot water, with thorough mixing and centrifuging each time. Then, to each tube, 2 ml of 3 N  $\text{H}_2\text{SO}_4$  was added and the precipitate was dissolved by warming in a water bath at 70–80 °C for 30 min. The content of each tube was titrated with freshly prepared 0.01 N potassium permanganate solution. Titration was done at room temperature (29 °C) until the first pink colour appeared throughout the solution. The solution was allowed to stand until it was colourless. It was warmed to 70–80 °C and titration continued until a pink colour persisted for at least 30 s.

$$\% \text{ Oxalate} = \frac{W \times 100}{5}$$

W = Mass of oxalate in 100 ml of extract.

### 2.10. Determination of total phenol

The sample (5 g) was boiled with a 50 ml of ether for 14 min. 5 ml of the extract was pipette into a 50 ml flask, then 10 ml of distilled water was added. 2 ml of ammonium hydroxide solution and 5 ml of concentrated amyl-alcohol were also added. The sample was made up to mark and left to react for 30 min for colour

development. The absorbance was read using spectrophotometer at 505 nm wavelength (Obadoni & Ochuko, 2001)

### 2.11. Alkaloid profile determination

Alkaloid extraction was carried out using modified method of Wink (1993). 2 g flour material was homogenized in 30 ml 0.5 N HCl. After 30 min at room temperature, the homogenate was centrifuged for 10 min 5000g. For quantitative work, the residue was re-suspended in 0.5 N HCl and centrifuged again. Both supernatants were then pooled and adjusted to pH 10 with NH<sub>4</sub>OH (25%). Alkaloids were extracted by solid-phase extraction using Extrelut column (Merck, Darmstadt). Total alkaloids were eluted with CH<sub>2</sub>Cl<sub>2</sub> and the solvent evaporated *in vacuo*. The alkaloid extract was dissolved in CH<sub>2</sub>Cl<sub>2</sub> and applied into a GC apparatus (Hewlett Packard Model 6890 series). Experimental conditions for capillary GC analysis were developed under the following conditions. Injector temperature 250 °C, carrier gas helium (1 ml/min), split ratio 1/20, injection volume 0.2 µl. GC oven temperature was kept at 120 °C for 2 min, programmed to 300 °C at a rate of 6 °C/min, and kept constant at 300 °C for 10 min.

### 2.12. Statistical analysis

All procedures were carried out triplicates with replication. The mean and standard deviation of the data obtained were calculated. The data were evaluated for significant differences in their means with Analysis of Variance (ANOVA) ( $p \leq 0.05$ ). Differences between the means were separated using tukey test as packaged by SPSS (17.0) software.

## 3. Results and discussion

### 3.1. Effect of delayed harvesting and pre-treatment on phenol and tannin contents

The levels of phenols, phytates, oxalates and tannins in the two cultivars of trifoliate yam flours are given in Tables 1 and 2. The phenol and tannin contents of raw trifoliate yam flours were low with the white and yellow cultivars having values ranging from 0.02 to 0.32 mg/100 g and 0.04 to 0.53 mg/100 g respectively.

White trifoliate yam flour harvested at 9 months had the highest phenol value but the value was not significantly differently ( $p > 0.05$ ) from raw white flour harvested at 8 and 10 months. However, no significant difference ( $p > 0.05$ ) exit in phenol contents of raw yellow trifoliate flours harvested at 7–10 months. Blanching/soaking method had the least values in phenol contents but the values were not significantly different ( $p > 0.05$ ) from values observed for pre-cooked flour at various harvesting periods. Higher phenol values were reported for raw flours of *D. dumetorum* (1.83%), *Dioscorea alata* (0.49 mg/100 g), *Dioscorea bulbifera* (1.40 mg/100 g) and *Dioscorea esculenta* (0.56 mg/100 g) but there were reductions in their values after processing (Ezeocha et al., 2012; Shanthakumari et al., 2008). Ngoddy and Onuoha (1985) reported higher polyphenol in yellow yam (*Dioscorea cayenensis*) than other yam species. Polyphenolic compounds inhibit the activity of digestive as well as hydrolytic enzymes, such as amylase, trypsin, chymotrypsin and lipase (Salunkhe, Sathe, & Reddy, 1982; Shanthakumari et al. 2008).

Tannin contents of the trifoliate yam flour increased till 9 months in white cultivar and 10 months in yellow cultivar. There was reduction in the values at 11 months due to sprouting of the tuber according to Medoua et al. (2007). There was no significant ( $p > 0.05$ ) difference in the tannin contents of raw white flours at 8–10 months and yellow flour from 7 to 11 months. Tannin contents were greatly reduced by pre-cooking and blanching/soaking methods. Tannin concentrations in trifoliate yam flours were lower than reported values for other yam species: *D. cayenensis* and *D. alata* (4.40 mg/100 g and 13.20 mg/100 g respectively: Polycarp et al., 2012), *D. alata*, *D. bulbifera* and *D. esculenta* (0.51, 1.59 and 0.11 mg/100 g respectively: Ezeocha et al. 2012; Shanthakumari et al. 2008), under-utilized *Dioscorea* tubers (20–255 mg/100 g: Arinathan, Mohan, & Maruthupandian, 2009). Ahmed, Hamid, Ali, Hassan & Babiker (2006) and Essenwah and Ikenebomeh (2008) reported losses in tannins content in legumes during processing due to its solubility in water and its sensitivity to heat during boiling and toasting. The decrease in the contents of phenol and tannin during pre-processing may be due to leaching out of the phenolic substances in soaking and cooking medium. The phenolics compounds are water soluble compounds (Uzogara, Morton, & Daniel, 1990) and as such can be eliminated by soaking and cooking (Murugesan & Ananthalakshmi, 1991; Shanthakumari et al., 2008).

**Table 1**  
Antinutritional contents of white trifoliate yam flour.

| Cultivar | Harvesting periods (months) | Phenols (mg/100 g)         | Phytates (mg/100 g)          | Oxalate (mg/100 g)        | Tannin (mg/100 g)          |
|----------|-----------------------------|----------------------------|------------------------------|---------------------------|----------------------------|
| WRF      | 7                           | 0.23 ± 0.01 <sup>bc</sup>  | 0.88 ± 0.03 <sup>abcde</sup> | 2.64 ± 0.04 <sup>d</sup>  | 0.44 ± 0.05 <sup>b</sup>   |
|          | 8                           | 0.27 ± 0.01 <sup>ab</sup>  | 1.03 ± 0.09 <sup>ab</sup>    | 2.91 ± 0.04 <sup>c</sup>  | 0.48 ± 0.01 <sup>ab</sup>  |
|          | 9                           | 0.32 ± 0.01 <sup>a</sup>   | 0.91 ± 0.04 <sup>abcde</sup> | 4.01 ± 0.06 <sup>a</sup>  | 0.53 ± 0.03 <sup>a</sup>   |
|          | 10                          | 0.28 ± 0.01 <sup>a</sup>   | 1.05 ± 0.19 <sup>a</sup>     | 3.63 ± 0.04 <sup>b</sup>  | 0.48 ± 0.01 <sup>ab</sup>  |
|          | 11                          | 0.14 ± 0.01 <sup>de</sup>  | 0.86 ± 0.04 <sup>abcde</sup> | 3.74 ± 0.06 <sup>b</sup>  | 0.25 ± 0.02 <sup>fg</sup>  |
| WSF      | 7                           | 0.19 ± 0.01 <sup>cd</sup>  | 0.86 ± 0.06 <sup>abcde</sup> | 1.81 ± 0.01 <sup>g</sup>  | 0.27 ± 0.02 <sup>ef</sup>  |
|          | 8                           | 0.21 ± 0.01 <sup>c</sup>   | 0.99 ± 0.03 <sup>abcd</sup>  | 1.72 ± 0.06 <sup>g</sup>  | 0.33 ± 0.03 <sup>de</sup>  |
|          | 9                           | 0.28 ± 0.01 <sup>a</sup>   | 0.80 ± 0.04 <sup>bcdef</sup> | 2.22 ± 0.01 <sup>e</sup>  | 0.42 ± 0.01 <sup>bc</sup>  |
|          | 10                          | 0.20 ± 0.01 <sup>c</sup>   | 0.96 ± 0.06 <sup>abcde</sup> | 1.99 ± 0.02 <sup>f</sup>  | 0.36 ± 0.02 <sup>cd</sup>  |
|          | 11                          | 0.13 ± 0.02 <sup>ef</sup>  | 0.78 ± 0.03 <sup>cdef</sup>  | 2.13 ± 0.02 <sup>ef</sup> | 0.18 ± 0.02 <sup>gh</sup>  |
| WBF      | 7                           | 0.04 ± 0.01 <sup>h</sup>   | 0.24 ± 0.04 <sup>j</sup>     | 0.18 ± 0.01 <sup>h</sup>  | 0.08 ± 0.01 <sup>j</sup>   |
|          | 8                           | 0.03 ± 0.01 <sup>h</sup>   | 0.49 ± 0.01 <sup>ghi</sup>   | 0.17 ± 0.01 <sup>h</sup>  | 0.07 ± 0.01 <sup>j</sup>   |
|          | 9                           | 0.02 ± 0.01 <sup>h</sup>   | 0.34 ± 0.01 <sup>hij</sup>   | 0.16 ± 0.02 <sup>h</sup>  | 0.11 ± 0.01 <sup>hij</sup> |
|          | 10                          | 0.02 ± 0.01 <sup>h</sup>   | 0.39 ± 0.03 <sup>hij</sup>   | 0.17 ± 0.02 <sup>h</sup>  | 0.10 ± 0.01 <sup>ij</sup>  |
|          | 11                          | 0.02 ± 0.01 <sup>h</sup>   | 0.31 ± 0.01 <sup>ij</sup>    | 0.17 ± 0.03 <sup>h</sup>  | 0.04 ± 0.01 <sup>j</sup>   |
| WPF      | 7                           | 0.09 ± 0.01 <sup>fgh</sup> | 0.58 ± 0.04 <sup>fgh</sup>   | 0.24 ± 0.02 <sup>h</sup>  | 0.11 ± 0.01 <sup>hij</sup> |
|          | 8                           | 0.11 ± 0.01 <sup>efg</sup> | 0.73 ± 0.05 <sup>def</sup>   | 0.26 ± 0.02 <sup>h</sup>  | 0.10 ± 0.02 <sup>ij</sup>  |
|          | 9                           | 0.12 ± 0.01 <sup>ef</sup>  | 0.79 ± 0.05 <sup>cdef</sup>  | 0.28 ± 0.02 <sup>h</sup>  | 0.16 ± 0.02 <sup>hi</sup>  |
|          | 10                          | 0.06 ± 0.01 <sup>gh</sup>  | 0.69 ± 0.03 <sup>efg</sup>   | 0.21 ± 0.06 <sup>h</sup>  | 0.11 ± 0.01 <sup>hij</sup> |
|          | 11                          | 0.06 ± 0.01 <sup>gh</sup>  | 0.69 ± 0.01 <sup>efg</sup>   | 0.20 ± 0.06 <sup>h</sup>  | 0.06 ± 0.01 <sup>j</sup>   |

WRF – White raw flour; WSF – White soaked flour; WBF – White blanched flour; WPF – White pre-cooked flour.

<sup>a</sup> Values with the same letter down the column were not significant different ( $p \leq 0.05$ ).

<sup>b</sup> Values are means of three determinations ± S.D.

**Table 2**  
Antinutritional contents of white trifoliate yam flour.

| Cultivar | Harvesting periods (months) | Phenols (mg/100 g)          | Phytates (mg/100 g)        | Oxalate (mg/100 g)        | Tannin (mg/100 g)           |
|----------|-----------------------------|-----------------------------|----------------------------|---------------------------|-----------------------------|
| YRF      | 7                           | 0.21 ± 0.01 <sup>a</sup>    | 0.60 ± 0.03 <sup>de</sup>  | 3.03 ± 0.04 <sup>b</sup>  | 0.34 ± 0.03 <sup>abc</sup>  |
|          | 8                           | 0.21 ± 0.03 <sup>a</sup>    | 0.93 ± 0.01 <sup>ab</sup>  | 2.71 ± 0.01 <sup>bc</sup> | 0.31 ± 0.02 <sup>abc</sup>  |
|          | 9                           | 0.16 ± 0.03 <sup>abc</sup>  | 0.97 ± 0.01 <sup>a</sup>   | 4.32 ± 0.51 <sup>a</sup>  | 0.34 ± 0.023 <sup>abc</sup> |
|          | 10                          | 0.17 ± 0.01 <sup>ab</sup>   | 0.97 ± 0.04 <sup>a</sup>   | 2.40 ± 0.33 <sup>c</sup>  | 0.37 ± 0.02 <sup>a</sup>    |
|          | 11                          | 0.13 ± 0.01 <sup>bcd</sup>  | 0.80 ± 0.12 <sup>bc</sup>  | 1.29 ± 0.09 <sup>d</sup>  | 0.35 ± 0.02 <sup>ab</sup>   |
| YSF      | 7                           | 0.17 ± 0.02 <sup>ab</sup>   | 0.58 ± 0.04 <sup>de</sup>  | 2.78 ± 0.13 <sup>bc</sup> | 0.31 ± 0.01 <sup>abc</sup>  |
|          | 8                           | 0.12 ± 0.01 <sup>bcde</sup> | 0.85 ± 0.06 <sup>abc</sup> | 2.38 ± 0.11 <sup>c</sup>  | 0.28 ± 0.01 <sup>c</sup>    |
|          | 9                           | 0.11 ± 0.01 <sup>cde</sup>  | 0.89 ± 0.01 <sup>ab</sup>  | 3.84 ± 0.22 <sup>a</sup>  | 0.29 ± 0.01 <sup>bc</sup>   |
|          | 10                          | 0.10 ± 0.01 <sup>def</sup>  | 0.84 ± 0.08 <sup>abc</sup> | 2.20 ± 0.03 <sup>c</sup>  | 0.32 ± 0.01 <sup>abc</sup>  |
|          | 11                          | 0.10 ± 0.02 <sup>def</sup>  | 0.97 ± 0.01 <sup>a</sup>   | 1.17 ± 0.01 <sup>d</sup>  | 0.31 ± 0.01 <sup>abc</sup>  |
| YBF      | 7                           | 0.02 ± 0.01 <sup>g</sup>    | 0.22 ± 0.04 <sup>g</sup>   | 0.13 ± 0.03 <sup>e</sup>  | 0.02 ± 0.01 <sup>d</sup>    |
|          | 8                           | 0.03 ± 0.01 <sup>g</sup>    | 0.30 ± 0.01 <sup>fg</sup>  | 0.12 ± 0.21 <sup>e</sup>  | 0.04 ± 0.01 <sup>d</sup>    |
|          | 9                           | 0.02 ± 0.01 <sup>g</sup>    | 0.31 ± 0.01 <sup>fg</sup>  | 0.12 ± 0.04 <sup>e</sup>  | 0.04 ± 0.01 <sup>d</sup>    |
|          | 10                          | 0.02 ± 0.01 <sup>g</sup>    | 0.23 ± 0.04 <sup>g</sup>   | 0.11 ± 0.01 <sup>e</sup>  | 0.05 ± 0.01 <sup>d</sup>    |
|          | 11                          | 0.02 ± 0.01 <sup>g</sup>    | 0.20 ± 0.03 <sup>g</sup>   | 0.14 ± 0.02 <sup>e</sup>  | 0.05 ± 0.01 <sup>d</sup>    |
| YPF      | 7                           | 0.03 ± 0.02 <sup>g</sup>    | 0.46 ± 0.01 <sup>ef</sup>  | 0.25 ± 0.03 <sup>e</sup>  | 0.08 ± 0.01 <sup>d</sup>    |
|          | 8                           | 0.02 ± 0.01 <sup>g</sup>    | 0.71 ± 0.02 <sup>cd</sup>  | 0.26 ± 0.04 <sup>e</sup>  | 0.08 ± 0.02 <sup>d</sup>    |
|          | 9                           | 0.04 ± 0.01 <sup>fg</sup>   | 0.71 ± 0.01 <sup>cd</sup>  | 0.24 ± 0.03 <sup>e</sup>  | 0.05 ± 0.01 <sup>d</sup>    |
|          | 10                          | 0.04 ± 0.01 <sup>fg</sup>   | 0.71 ± 0.02 <sup>cd</sup>  | 0.22 ± 0.02 <sup>e</sup>  | 0.05 ± 0.01 <sup>d</sup>    |
|          | 11                          | 0.06 ± 0.01 <sup>efg</sup>  | 0.71 ± 0.01 <sup>cd</sup>  | 0.19 ± 0.02 <sup>e</sup>  | 0.07 ± 0.01 <sup>d</sup>    |

YRF – Yellow raw flour; YSF –Yellow soaked flour; YBF–Yellow blanched flour; YPF–Yellow pre-cooked flour

<sup>a</sup> Values with the same letter down the column were not significant different ( $p \leq 0.05$ ).<sup>b</sup> Values are means of three determinations  $\pm$  S.D.

### 3.2. Effect of delayed harvesting and pre-treatment on phytate contents

Phytate content was higher in white trifoliate yam flour harvested at 10 months (1.05 mg/100 g) but there was no significant difference ( $p > 0.05$ ) in the raw flours harvested at different periods. Likewise, there was no significant difference ( $p > 0.05$ ) in the raw yellow flours harvested at 8, 9 and 10 months. The phytate values of white trifoliate yam flour were higher than the yellow flours at different harvesting periods and there was slight decrease in the phytate level at 11 months. These corroborates the report of Medoua et al. (2007) that sprouting significantly reduced phytate contents of trifoliate yam flour. Among the pre-treatment methods used, blanching/soaking method was more effective in reducing phytate level by 51–72% in trifoliate yam flour. Higher phytate values were reported for white (2.50 mg/100 g) and yellow (2.10 mg/100 g) trifoliate yam flours but the values observed were comparable to the value for *D. alata* (0.89 mg/100 g) (Polycarp et al., 2012). Earlier studies on the phytate contents of yam flours showed higher results in other yam species (Bhandari & Kawabata, 2006; Wanasundera & Ravindran, 1992) indicating lower levels of phytate in trifoliate yam flour. Bhandari & Kawabata (2006) observed lower phytate level in processed yam flour and reduction of phytate is expected to enhance the bioavailability of proteins and dietary minerals of the yam tubers. Environmental factors, such as light, germination, stages of ripeness, variety, processing, storage and genetic factors can influence the levels of phytate contents in food (Khokhar & Apenten, 2003).

### 3.3. Effect of delayed harvesting and pre-treatment on oxalate contents

Oxalate contents of both cultivars increased till 9 months and there was gradual decrease in the value at 10 months. Raw yellow flour at 9 months had higher oxalate content which was significantly different ( $p > 0.05$ ) from other flours at other periods. The oxalate contents observed for raw trifoliate yam flours were higher than the pre-treated flours. There was 96% reduction in the oxalate content of blanched/soaked flour at 9 months. Although, blanching/soaking method was more effective in reducing oxalate but no significant difference ( $p > 0.05$ ) exist between blanched/soaked

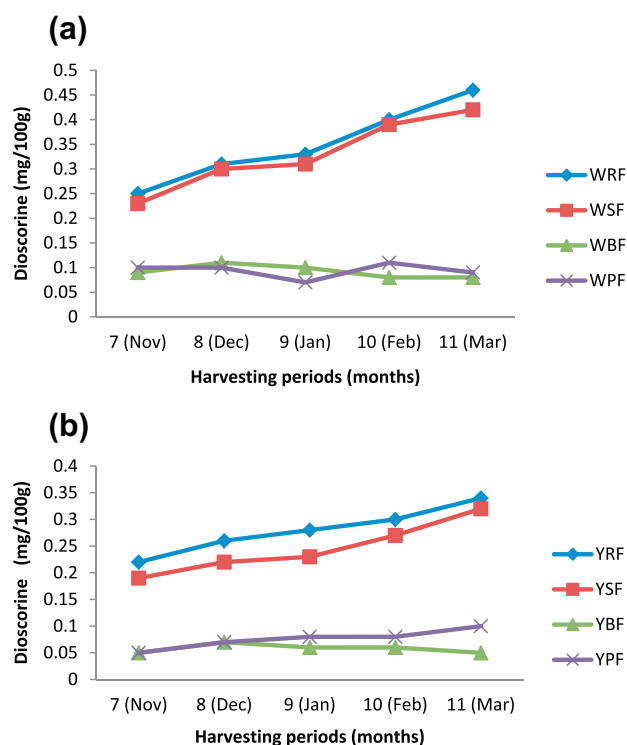


**Fig. 1.** Effect of delayed harvesting and pre-treatment methods on the dioscorine contents of A-white trifoliate yam flour, B-yellow trifoliate yam flour. WRF – White raw flour; WSF –White soaked flour; WBF – White blanched flour; WPF – White pre-cooked flour. YRF – Yellow raw flour; YSF –Yellow soaked flour; YBF – Yellow blanched/soaked flour; YPF – Yellow pre-cooked flour. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

flours and pre-cooked flours harvested at different periods for the two cultivars. Earlier reports showed lower oxalate contents 0.43 mg/100 g and 0.45 mg/100 g in raw white and yellow trifoliate yam flour respectively (Polycarp et al. 2012). However, Shanthakumari et al. (2008) and Shajeela, Mohan, Jesudas, and Soris (2011) observed lower oxalate contents ranging from 0.10

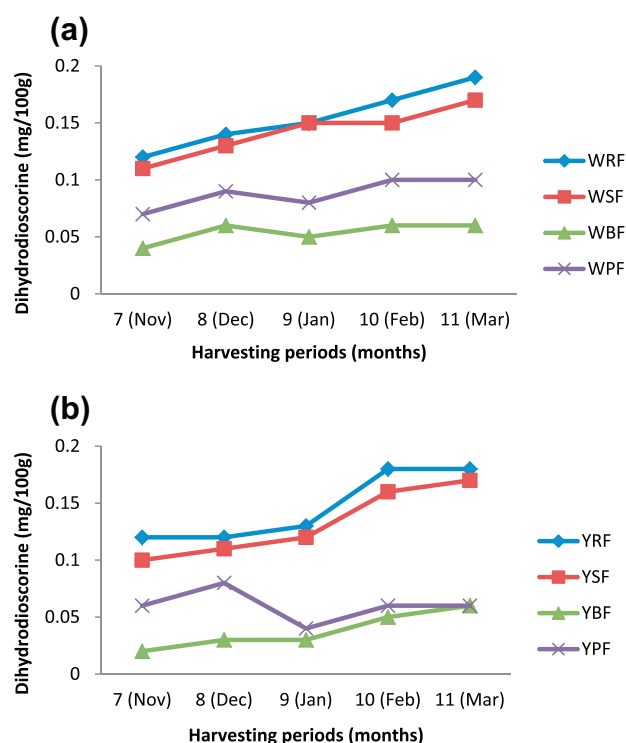


**Fig. 2.** Effect of delayed harvesting and pre-treatment methods on the dihydrodioscorine contents of A-white trifoliate yam flour, B-yellow trifoliate yam flour. WRF – White raw flour; WSF – White soaked flour; WBF – White blanched flour; WPF – White pre-cooked flour. YRF – Yellow raw flour; YSF – Yellow soaked flour; YBF – Yellow blanched flour; YPF – Yellow pre-cooked flour. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

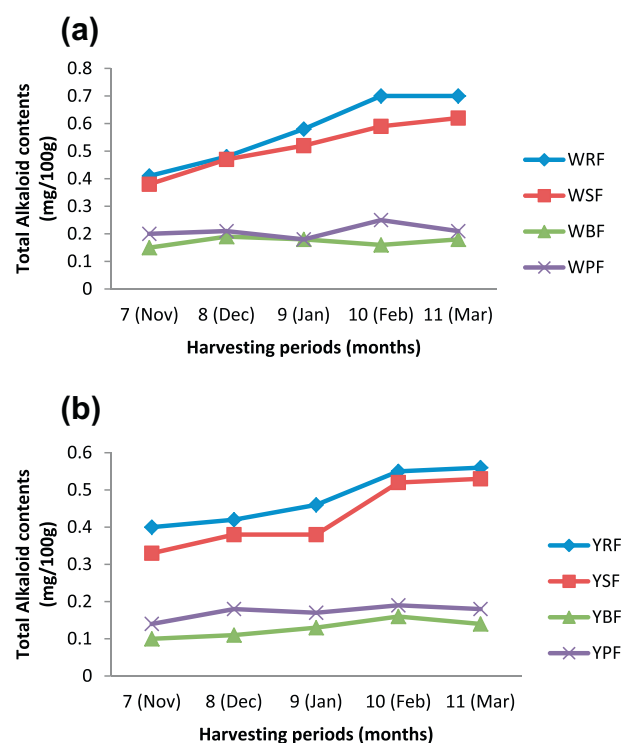


**Fig. 3.** Effect of delayed harvesting and pre-treatment methods on the total alkaloid contents of A-white trifoliate yam flour, B-yellow trifoliate yam flour. WRF – White raw flour; WSF – White soaked flour; WBF – White blanched flour; WPF – White pre-cooked flour. YRF – Yellow raw flour; YSF – Yellow soaked flour; YBF – Yellow blanched flour; YPF – Yellow pre-cooked flour. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

to 0.98 for different species of yam while Ogbuagu (2008) observed lower oxalate (2.32 mg/100 g) for raw trifoliate yam flour. Higher percentage of oxalate reduction during blanching/soaking and pre-cooking methods may be due to its solubility in boiling water. Boiling was reported to cause considerable skin rupture and facilitate the leakage of soluble oxalate into the cooking water (Albiñ & Salvage, 2001; Bhandari & Kawabata, 2006).

#### 3.4. Effect of delayed harvesting and pre-treatment on alkaloid contents

The dioscorine, dihydrodioscorine and total alkaloid contents of white and yellow trifoliate yam flours are presented in Figs. 1–3. The predominant alkaloids in trifoliate yam flours were dioscorine and dihydrodioscorine with values ranging from 0.05 to 0.46 mg/100 g and 0.04 to 0.19 mg/100 g respectively. Raw trifoliate yam flour at 11 months had higher values than the soaked, blanched/soaked and pre-cooked flours. Dioscorine and dihydrodioscorine levels in the yam increased with harvesting periods. The levels of alkaloid in white trifoliate yam flours were higher than the yellow trifoliate yam flours. Total alkaloid contents increased slightly with delayed harvesting periods while blanching/soaking drastically reduced the anti-nutrient contents compared to other methods. Higher alkaloid contents were reported in trifoliate yam flour ranging from 0.89% to 3.08% (Ezeocha et al., 2012; Ogbuagu, 2008; Okwu & Ndu, 2006). The longer the tubers are kept in the soil, the lower the moisture contents thereby increasing the alkaloid concentration of the tuber. Other alkaloid present in insignificant amount were chorine, trigonelline, angustifoline, sparteine, ellipicine, lupanine, 13-alphahydrochombifoline, 9-octadecenamide, dihydro-oxo-demethoxyhaemanthamine, augutamine, oxoasoanine, cincho-

nine, crinane-3 $\alpha$ -ol, buphanidine, cinchonidine, indicine-N-oxide, powelline, undulatin, ambelline, 6-hydroxybuphanidine, acronycine, monocrotaline, 6-hydropowelline, nitidine, crinamide, 1- $\beta$ , 2- $\beta$ -epoxyambelline, 6-hydroxyundulatin, epoxy-3,7-dimethoxycrinane-11-one, echitammidine, akuammidine, voacangine, mitraphylin camptothecin, echitamine, colchicine, tetrandrine, emetine, thalicarpin and paclitaxel. Alkaloids contributed to the bitter principles in trifoliate yam and higher levels in food are known to cause paralysis of the nervous system.

#### 4. Conclusion

Trifoliate yam had been considered as highly nutritious and the levels of anti-nutritional contents are very low when compared to other yam species. Raw white trifoliate yam flour had higher anti-nutrient contents than the yellow flours but irrespective of harvesting periods, blanching/soaking method was an effective method in reducing the anti-nutritional contents of trifoliate yam flours to minimal level. Therefore, reduction in these anti-nutrient contents provides a safe food for human consumption.

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