Advanced Journal of Microbiology Research Vol. 2012
Available online at http://internationalscholarsjournals.org/journal/ajmr


*Full Length Research Paper*

# Quantitative survey and anti-microbial effect of *Piliostigma thonnigii* and *Khaya ivorensis* leaves on traditional dry-yam

**Babajide, J. M * and Atanda, O.O**

Department of Food Science and Technology, University of Agriculture, P. O. Box 52, Alabata-UNAAB, Abeokuta, Nigeria.

Accepted 20 March, 2012

**A survey was conducted to determine the quantity of local preservatives ('Abafe' *Piliostigma thonnigii* and 'Agehu' *Khaya ivorensis* leaves) used for parboiling yam and the anti-microbial effect of the leaves on dry- yam 'Gbodo'. Although, the study revealed that 23 g of abafe, 13 g of agehu and a combination of 12.8 g of abafe and 12.3 g of agehu will be required for 1 kg of yam, there were wide ranges between the quantities of yam as well as the quantities of local preservatives used by the processors. Futhermore, there were reduction in the microbial loads of samples parboiled with the leaves (singly or combined) respectively, immediately after processing and after 3 months of storage compared with the control. Thus, *P. thonnigii* leaves had bactericidal while *K. ivorensis* leaves had fungicidal effect on the microorganisms encountered during processing and storage of Gbodo.**

**Key words:** Quantitative survey, anti-microbial, *Piliostigma thonningii*, Khaya ivorensis, dry-yam.

## INTRODUCTION

Yam, Dioscorea (*spp.*) is an important source of carbo-hydrate for many people of the sub-Saharan region, especially in the yam zones of West Africa (Akissoe et al., 2003). Several species of yams are grown in the tropics and temperate zones of the world (Kordylas, 1990). It is the second most important root/tuber crop in Africa, after cassava, with production reaching just under one third the level of cassava (FAO, 1997). Some are grown only for medicinal purposes and others for edible purposes. Of the edible species, *Dioscorea alata* L., known as the greater yam, *Dioscorea cayenensis* Lam., the yellow yam, and *Dioscorea rotundata* Poir. and *Dioscorea esculenta* the white yam, are the most common (O'Hair, 1990). In order to minimize losses, considerable quantities of roots and tubers are trans-formed into more durable products by drying, fermen-tation and comminuted in different combinations and sequences producing a variety of materials each with distinctive characteristics. This involves processing the yams into dry-yam tubers/slices and flour (Bricas et al., 1997). In some West African countries such as Nigeria, Benin and Ghana, the age old traditional method is still being used for processing of Gbodo. The dry yam tubers/slices are processed by peeling, slicing, parboiling in hot water (40 to 60°C for 1 to 3 h), steeping (24 h) and sun-drying, into a product called 'Gbodo' by the Yorubas of south-west Nigeria (Onayemi and Potter, 1974).

Preliminary survey carried out on 263 processors of the yams in southwest Nigeria revealed that the local con-sumers have preference for the dry yams made by the Baruba/Baruten people of Kwara state who incidentally are the major producers of the traditional dry-yams (Babajide, 2005, 2007). This could be because they add 'Abafe' *Piliostigma thonnigii* and 'Agehu' *Khaya ivorensis* leaves and their combinations as local preservatives during parboiling of the yams (Babajide, 2005), but the quality of the dry- yams vary from processor to processor and from location to location as there is wide variation in the parboiling operation. Furthermore, the quantity of leaves per kilogram of yam could not be easily ascer-tained during the investgation as the process is crude and unstandardised. *P. thonningii* (Schum.) Milne-Rech is a member of the family Caesalpinioideae and is locally known as *Olofoo* (Hamer-Bena), *Kalkalla* (Wolayetna), *Yekallo wanza* (Amargna), Camel's foot tree and Monkey Bread (English) (Jimoh and Oladimeji, 2005). The plant is a small, rounded deciduous tree, 3 - 5m in height and grows on sandy soils in the bush (Bartha, 1970). It is a

*Corresponding author. Email: wodubajng@yahoo.com.

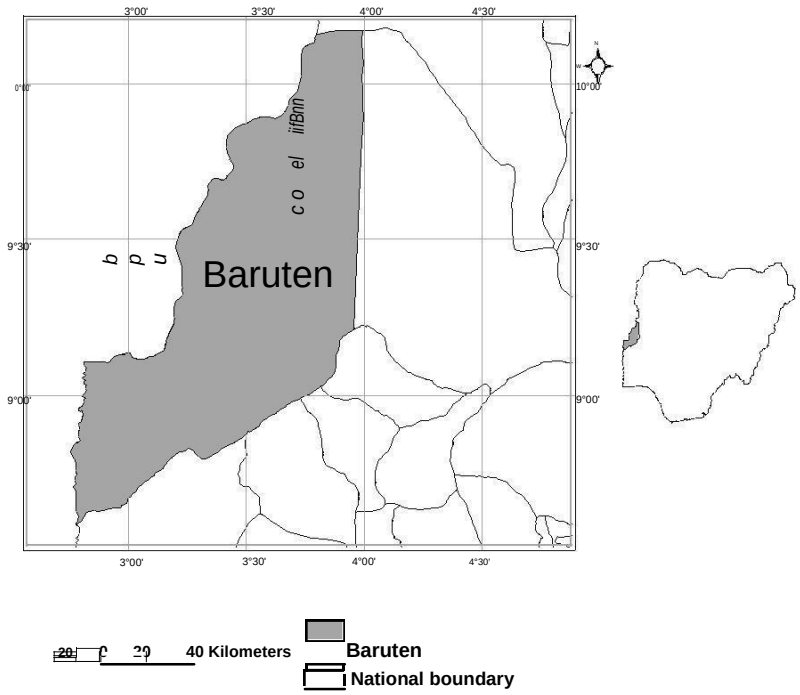


**Figure 1.** Map of Baruten - major processing area of Gbodo in Nigeria.

small tree with large two-lobed simple leaves and without thorns or spines Akinpelu and Obuotor, (2000) found that 60% methanolic extract of *P. thonningii* stem bark exhibited bactericidal activity against six out of eight isolates at a concentration of 20 mg/ml. Similarly, *Khaya ivorensis* commonly referred to as African mahogany is one of the plant species cultivated in the Centre for Scientific Research (CSRPM) arboretum at Ayikuma in the GaDangbe District of Ghana for medical purposes (Laira, 2000; Ameyaw and Ampaw, 2004). The stem barks of *K. ivorensis* A. Juss are commonly used by CSRPM, traditional medical practitioners (because of resistance to fungal decay) and other alcoholic beverage brewers in Ghana in preparing tonics for anaemia and appetizers (Samir et al, 2005).

Thus, this study reports the result of a survey conducted on the quantity of local preservatives ('Abafe' *Piliostigma thonnigii* and 'Agehu' *K. ivorensis* leaves) used for parboiling yam and the anti-microbial effect of the leaves on microbial loads of traditional dry-yam 'Gbodo'.

## MATERIALS AND METHODS

### Study location

A survey of traditional processing methods of Gbodo was carried out in the major processing area (Baruba/ Baruten) of Kwara state, Nigeria (Figure 1).

### Study method

A Rapid Rural Appraisal (RRA) process was followed in the survey of Gbodo following the method of Scrimshaw and Hurtado (1987). Direct interviews and observations and structured questionnaires were employed in collecting information on the average quantity of Gbodo and local preservatives used (singly or in combination) by each processor. Four lots consisting of twenty processors/lot each using Abafe, Agehu, combination of both and a control without the addition of local preservatives were interviewed. A total of 80 respondents minus the control were used in the quantitative survey which were analysed using descriptive statistical analysis.

### Microbiological analyses of Gbodo

Samples from each lot were thoroughly mixed together and 100 g of each lot was collected in sterile polythene bags. The samples were subsequently milled in a plate mill and stored at 4°C prior to microbiological analyses. The microbial loads of the samples were determined at monthly intervals of 3 months of storage**.**

### Enumeration of microorganisms

The aerobic plate count was determined by plating 0.1 ml of the aliquots separately on triplicate plates of Nutrient agar (Oxoid) and incubated in Gallenkamp incubator at 30°C for 48 h. The samples were diluted decimally and 0.1 ml surface spread on duplicate plates using sterile glass spreader. Pure cultures of isolates were stored on Nutrient agar slants at 4°C for further confirmatory tests which included carbohydrate utilization, and reaction on TSI. Large, flat, irregular, wrinkled or smooth, ground-glass colonies, 4 to 6 mm in diameter were counted as *Bacillus*. Confirmation was as described by Yusuf et al. (1992). The Fungi count was determined by plating 0.1ml aliquots of the samples on potato dextrose agar (Oxoid) to which 0.01% chloramphenicol has been added to inhibit bacterial growth and incubating the plates at room temperature (28°C) for 72 h. Observed colonies were sub cultured to obtain pure

**Table 1.** Descriptive statistics of local preservatives per quantity of yam.

| Quantity (kg) | Minimum | Maximum | Mean | Std. Deviation |
|---|---|---|---|---|
| **Abafe (n = 20)** | | | | |
| Yam | 40.00 | 135.00 | 85.25 | 26.77 |
| Abafe | 0.36 | 2.70 | 1.31 | 0.76 |
| 1 kg of yam will require 23 g of Abafe | | | | |
| **Agehu (n = 20)** | | | | |
| Yam | 40.00 | 125.00 | 88.75 | 26.50 |
| Agehu | 0.31 | 2.70 | 1.19 | 0.63 |
| 1 kg of yam will require 13 g of Agehu | | | | |
| **Combination of abafe and agehu (n = 20)** | | | | |
| Yam | 45.00 | 135.00 | 90.75 | 26.67 |
| Abafe | 0.53 | 2.79 | 0.16 | 0.64 |
| Agehu | 0.48 | 2.21 | 1.11 | 0.49 |
| 1 kg of yam will require 12.8 g of abafe and 12.3 g of agehu | | | | |

cultures which were incubated up to 5 - 7 days and subsequently isolated and identified using standard methods (International Commission on Microbiological Specification for Food ICMSF, 1996).

Presumptive staphylococcal count was determined by inoculating staphylococcal medium 110 (Oxoid) with the specimens and incubating at 32°C for 72 h. Pigmented colonies surrounded by bright yellow zones (halo) resulting from mannitol fermentation were counted. Confirmation of *S. aureus* was by positive coagulase test.

## RESULTS AND DISCUSSION

Table 1 shows the quantities of local preservatives used per quantity of yam by each processor. On the average, 1.31 kg abafe was added to 85.25 kg yam; 1.19 kg agehu was added to 88.75 kg yam; combination of 0.16 kg abafe and 1.11 kg agehu was added to 90.75 kg yam during parboiling. The study revealed that 23 g of abafe, 13 g of agehu and a combination of 12.8 g of abafe and 12.3 g of agehu will be required for 1 kg of yam. However, there exist wide ranges in the quantities of yam per quantities of local preservatives; for instance, there were 40 kg yam; 0.36 kg abafe minimum and 135 kg yam; 2.70 kg abafe maximum. Thus, the need to further study the effect of varied quantities of the local preservatives on the microbial load of gbodo.

Table 2 showed that Gbodo parboiled with abafe leaves had lower total plate counts and stapylococci counts than the control samples, as there were further reduction after 3 months storage period ($3.0x10^3$ cfu/g and $3.1 \times 10^2$ cfu/g respectively). Yams parboiled with agehu had lower fungi count ($2.1 \times 10^3$ cfu/g) compared with yams without preservatives ($5.4 \times 10^6$ cfu/g) whose count further reduced to $2.4 \times 10^2$ cfu/g after 3 months of storage. The combination of abafe and agehu leaves as local preservatives had lower plate, fungi and staphy-lococci counts after processing and at 3 months period of storage, compared with the control. The anti-microbial effect of abafe leaves on Gbodo confirmed the findings of Ibewuike et al. (1997) that abafe has bactericidal effect on *Staphylococus aureus* while Agehu was also found to have fungicidal effect on fungi species (Botanic Gardens Conservation International BGCI, 2002; Samir et al., 2005). There was a reduction in the microbial load of the samples parboiled with mixtures of the two types of leaves after three months storage period suggesting that the leaves are preservative in nature. This probably explains the reason why the local dry-yam processors are in the habit of adding the leaves during parboiling.

The microorganisms identified from the samples namely *S. aureus, Bacillus spp, Proteus spp, Pseudomonas spp. Aspergillus niger and A. flavus* were identified from both treated and untreated dry-yams. The occurrence of these organisms in treated samples could be due to recontamination through handling by the processors, nevertheless, the occurrence were lower compared to the sample with no local preservative.

## Conclusion

This study revealed that 23 g of abafe, 13 g of agehu and a combination of 12.8 g of abafe and 12.3 g of agehu will be required for 1 kg of yam. Futhermore, the *P. thonnigii* Leaves had bactericidal, *K. ivorensis* leaves had fungi-cidal effect on the microorganisms encountered during the processing. Further study is on going to standardize the application of local preservatives by varying the quantities of these local preservatives to fixed quantities of yam also *in-vitro* evaluation of anti-microbial activities of leave extracts against isolated micro-organisms is ongoing.

## ACKNOWLEDGEMENT

The authors wish to acknowledge the Nutricia Research Foundation, Netherlands, for sponsoring this research

**Table 2.** Microbial load of processed and stored dry-yam (gbodo).

| Dry-yams (gbodo) parboiled with Local preservatives | Microbial load of processed gbodo | | | Microbial load of gbodo at one month of storage | | | Microbial load of gbodo at two months of storage | | | Microbial load of gbodo at three months of storage | | | Micro organisms isolated from samples |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Total plate count (10 cfu/g) | Fungal count (10 cfu/g) | Staphylococcal count ($10^3$ cfu/g) | Total plate count ($10^3$ cfu/g) | Fungal count (10 cfu/g) | Staphylococcal count ($10^3$ cfu/g) | Total plate count ($10^3$ cfu/g) | Fungal count (10 cfu/g) | Staphylococcal count ($10^3$ cfu/g) | Total plate count ($10^3$ cfu/g) | Fungal count (10 cfu/g) | Staphylococcal count ($10^3$ cfu/g) | |
| Abafe | 4.1 | 560 | 0.83 | 4 | 220 | 0.71 | 3.4 | 93 | 0.52 | 3 | 52 | 0.31 | *Staphylococcus aureus, Bacillus spp, Proteus spp, Pseudomonas spp. Aspergillus niger and A. flavus* |
| Agehu | 660 | 2.1 | 32 | 120 | 0.94 | 30 | 48 | 0.39 | 30 | 0.9 | 0.24 | 28 | |
| Abafe + Agehu (Control) | 3.5 | 8.1 | 0.68 | 2.2 | 5.4 | 0.5 | 1.3 | 3.2 | 0.28 | 0.27 | 1.4 | 0.13 | |
| No preservative | 7200 | 5400 | 9100 | 6300 | 6200 | 9500 | 6800 | 6900 | 9300 | 5700 | 7300 | 9200 | |

and other ongoing work in this study.

**REFERENCES**


Ameyaw Y, Ampaw I (2004). Healing or recovery ability of Harvested Stem Bark of a Medicinal Plant, *Khaya Senegelensis* (Desr.) A. Juss., In A Cultivated Arboretum In The Greater-Accra Region Of Ghana. Centre For Scientific Research Into Plant Medicine, Mampong-Akuapem, Eastern Region, Ghana. 1:216.

Akinpelu DA, Obuotor EM (2000). Antibacterial activity of Piliostigma thonningii stem bark. *Fitoterapia* Elsevier publishers. 71(4): 442-3.

Akissoe NH, Hounhouigan JD, Mestres C, Nago M (2003). How blanching and drying affect the colour and functional characteristics of yam (*Dioscorea cayenensis-rotundata*) flours. Food Chem. 82: 257-264.

Babajide JM (2005). Processing and Quality Improvement of Traditional Dry-Yam Tubers/Slices (Gbodo) and Yam Flour (Elubo). Ph D. Thesis. 1:182.

Babajide JM, Babajide SO, Oyewole OB (2007). Survey of Traditional Dry-yam Slices (Gbodo) Processing Operations in Southwest, Nigeria. America-Eurasian J. Sustainable Agric. 1(1): 45-49.

Bartha R (1970). Fodder plants in the Sahel zone of Africa, München, Weltforum Verlag. 1:245.

Botanic Gardens Conservation International (BGCI) (2002). Plants as medicine. Padua Botanic Garden, Foxgloves.pp. 121- 129.

Bricas N, Vernier P, Ategbo E, Hounhouigan J, Mitchikpe E, N'kpenu KE, Orkwor G (1997). The Development of Pounded Yam Machine in West Afr. J. Res. Dev.. 44(2):100-114.

Food and Agricultural Organisation FAO (1997). Food Production Yearbook, 50:160-168. Ibewuike JC, Ogungbamila FO, Ogundaini AO, Okeke IN,

Bohlin L (1997). Anti flammatory and antibacterial activities of C-methyl flavonoids from Pilistigma thonningii. *Society of* Chemical Industry. John Wiley and Sons, Ltd. 61(2): 186-190. International Commission on Microbiological Specification for

Food (ICMSF) (1996). Sampling for Microbiological analysis. Principles and Specific Application. University of Toronto. pp.1-18.

Jimoh FO, Oladiji AT (2005). Preliminary Studies on *Piliostigma thonningii* seeds: Proximate analysis, mineral composition and phytochemical screening. Afr. J. Biotechnol. 4(12):1439-1442.

Kordylas, JM (1990). Processing and Preservation of Tropical and Subtropical Foods. Macmillan Publishers Ltd. London and Basingstoke. pp.49-71.

Laira SA (2000). The management of forests for timber and non-wood forest products in central Africa. FAO Corporate Document Repository .pp. 1-150

O'Hair SK (1990). Tropical Root and Tuber Crops. In: Janick J and Simon JE (eds.), Advances in new crops. Timber Press, Portland. pp.424-428.

Samir AM, Abdelgaleil FH, Munehiro N (2005). Antifungal activity of limonoids from *Khaya ivorensis.* Pest Mgt. Sci. 61(2): 186-190.

Scimshaw SCM, Hurtado E (1987). Rural Rapid Assessment (RRA) procedures for nutrition and primary health care; Anthopological approaches to improving programmes effectiveness. Los Angeles, California, USA, Latin American Centre. 1: 377.

Yusuf IZ, Umoh VJ, Ahmad AA (1992). Occurrence and survival of enterotoxigenic *Bacillus cereus* in some Nigerian flour-based foods. Food Control . 3:149-152.