

CONSTRUCTION AND PERFORMANCE EVALUATION OF AN ON-FARM SCALE SOLID STATE FERMENTER

Ezekiel, O. O. and Aworh, O.C.

Department of Food Technology, University of Ibadan, Ibadan, Nigeria

ABSTRACT

The protein enrichment of cassava peel using solid state fermentation (SSF) was studied on both laboratory and on-farm scales using Trichoderma viride as a starter culture. The fermentation of cassava peel was carried out using cassava peel with particle size 4.00 > p > 3.35 mm, initial moisture content of 60% at pH 6.0, 30°C incubation temperature with ammonium sulphate (10g N / kg substrate) as additional nitrogen source for 8 days at the laboratory scale and 28-30°C at the on-farm scale. A high relative humidity of 90-95% was maintained throughout the fermentation period. The fermented peel was oven-dried at 60°C, ground and analysed for crude protein, true protein, crude fat, crude fibre, ash, carbohydrate, starch and cyanide using standard methods. Comparable values were obtained for laboratory and on-farm scale fermentation. However, the on-farm technique yielded higher protein enrichment compared with laboratory experiments. Cassava peel fermented on-farm yielded 10.93% protein while laboratory scale yielded 10.43%

Key words: cassava peel, laboratory scale, on-farm scale, protein enrichment, solid-state fermentation.

INTRODUCTION

Cassava (*Manihot esculenta* Crantz syn. *Manihot utilissima* Pohl) a staple food of the majority of people in tropical Africa, Central and South America (Nestel, 1973; Subrahmanyam, 1990), is often subjected to series of fermentations in the different countries to produce similar or different products (Akinrele, 1967). In Brazil, Costa Rica and Bolivia, farina is often the end product while in tropical Africa gari, fufu, lafun, chiwangue and myondo are produced from cassava (Subrahmanyam, 1990, Giraud *et al.*, 1991). The various production processes are usually accompanied with some waste products that act as environmental pollutants (Onilude, 1996).

Cassava peels, leaves and starch residues constitute 25% of the cassava plant (Iyayi and Losel, 2001). These are usually discarded as wastes after harvesting and processing, with limited utilization due to low protein, high crude fibre and cyanide contents (Iyayi and Tewe, 1988). The peel is about 10-20% of the tuber and is available all-year-round in Nigeria

with an annual yield of approximately 4 million metric tonnes from the processing of cassava tuber (Hahn and Keyser, 1985). Little attention has been paid to the handling of the large quantity of cassava wastes that generated. Solid state fermentation (SSF) is a veritable way of converting such wastes to wealth. Micro-organisms have the ability to upgrade low protein plant material to high protein feed (Rajoka, 2005). Large-scale fermentation of methanol, starch and molasses-based media has proved economically viable for the production of animal feed and human food (Rosenberg 1993; Hongpattarakere and H-kittim, 1995; Paul *et al.* 2002).

Processing by SSF is a relevant, initial approach to lignocellulose bioconversion appropriate for developing countries. It offers the possibility of using by-products and wastes from food and agricultural industries (Gomez *et al.*, 2005) for food, feed, chemicals and fuel. The greatest socio-economical potential of SSF is the raising of living standards through the production of protein rich foods for human consumption (Raimbault, 1998). This can be achieved

by exploring two alternatives: production of protein-enriched fermented foods for direct human consumption and production of fermented materials for animal feeding.

Starch substrate protein-enriched by SSF could be fed to monogastric animals or poultry.

Bacteria, yeasts and fungi can grow on solid substrate and find application in SSF processes. Filamentous fungi are the most important group of microorganisms used in solid-state process owing to their physiological, enzymological and biochemical properties (Raimbault, 1998). *Trichoderma viride*, a filamentous fungus, has been previously used in the fermentation of several food processing wastes De Gregorio *et al.* (2002) fermented lemon pulp with *T. viride* and *A. niger* by slurry-state fermentation and reported higher protein content in the residue with *T. viride* than with *A. niger*. Palm-tree leaflets and midribs, both of very low protein content, when chemically hydrolysed and used for the growth of *T. viride* were enriched with protein (Abou-Zeid, 1991).

Many high value products such as enzymes, primary and secondary metabolites, could be produced in SSF. However, improvements in engineering and socio-economic aspects are required because processes must use cheap substrates available locally, low technology adaptable to rural areas, and simplified processes (Raimbault, 1998).

The objective of this work was to evaluate the performance of a constructed on-farm scale fermenter, for the fermentation of cassava peel by comparing it with laboratory scale using *Trichoderme viride* as a starter culture.

MATERIALS AND METHODS

Materials of Construction

The materials used in the construction of the solid state fermenter were:

- Aluminium 2-inch square pipes
- Galvanized steel sieve
- Pipex glass
- Aluminium long-span sheet
- Hinges
- Rivet pins
- ¾ inch diameter plastic pipes
- Formica sheet
- 0.033hp fan

The fermenter was constructed based on the procedure described by Noorhom *et al.* (1992) for fermenting cassava pulp using *Aspergillus niger* as a starter culture. The size of the perforations on the tray was determined based on preliminary findings from the laboratory experiments for the appropriate sample particle size which was found to be between 3.35 and 4.00 mm.

The following points were considered in the design of the fermenter:

- capacity of the fan to give proper aeration
- constant supply of water to be supplied to give relative humidity of 90-95%

Performance Evaluation

The peel from fresh cassava tubers, variety TME I, obtained from a farm at Ajibade village in Akinyele Local Government Area, Ibadan was used for this study.

Micro-organism

The filamentous fungi *T. viride* ATCC 36316 was obtained from American Type Culture Collection.

Maintenance Medium

T. viride ATCC 36316 was maintained on malt extract agar slants and stored at 4°C. The organism was sub-cultured once every 3 months.

Inoculum Preparation

The fungus was sub-cultured on malt extract agar in petri dishes for 5 days. Spore suspension containing approximately 3×10^6 spores per ml was prepared in Ringer solution.

Solid State Fermentation of Cassava peel: Laboratory Scale

Solid state fermentation of cassava peel on laboratory scale was carried out using cassava peel with particle size $4.00 > p > 3.35$ mm, initial moisture content of 60% at pH 6.0, 30°C incubation temperature with ammonium sulphate (10g N / kg substrate) as additional nitrogen source. These conditions were chosen based on the results of previous work carried out on optimisation of processing conditions for protein enrichment of cassava peel (Awoyele, 2010). Fermentation was allowed for 8 days. At the end of fermentation, samples were dried at 60°C for subsequent analysis. Figure 1 shows the flowchart for the fermentation procedure.

Solid State Fermentation of Cassava Peel: On-Farm Scale

Perforated mesh trays (Figures 2) with an area of 3920 cm² were stacked leaving a gap of 15.5mm between trays. The stack of trays loaded with uniformly spread inoculated cassava peel was covered with sacks and water, supplied through perforated pipes, was made to drip onto the sacks. The cassava peel was prepared for fermentation as in the laboratory scale experiment.

Compositional Analysis of Cassava Peel

Moisture content of the cassava peel was determined by drying at 105°C to constant weight (AOAC, 1995). The crude protein was by Kjeldahl method (total nitrogen x 6.25), crude fibre, fat, ash, carbohydrates (estimated by difference), total dietary fibre and gross metabolizable energy were quantified as described by AOAC methods (AOAC, 1995). True protein content was determined by the method of Lowry (1951). Total cyanide of dried samples was determined by phosphoric acid extraction, hydrolysis of cyanogenic glucosides with linamarase from cassava, followed by colorimetric determination of cyanide (Cooke, 1978).

RESULTS AND DISCUSSION

Features of the Fermenter

The fermenter has the shape of a cabinet with fixed racks and capacity for five perforated trays made with 0.2mm gauge stainless steel pipes. (Figure 2) Each tray can hold up to 400 g of substrate per fermentation batch.

The trays are 70 cm by 56 cm with a thickness of 5.5 cm each while each perforation has a diameter of 2 mm. Perforated pipes were installed at the top of the fermenter to supply water in order to create and maintain an environment with a high relative humidity during the fermentation process. At the bottom of all the trays was placed a fixed aluminium tray with a small outlet for removing overflow water being supplied. The fermenter was set on stands for stability. A portable electric fan with a capacity of 1/30hp was placed at the bottom of the bottom of the stack of trays in order to maintain proper aeration. The fermenter achieved temperature in the range of 28-30°C.

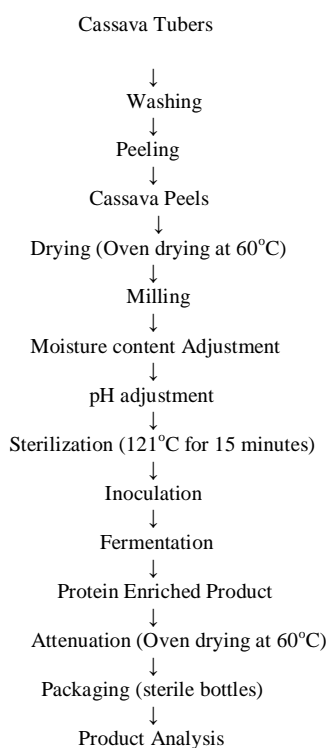


Figure 1: Flow Chart for Protein Enrichment of Cassava Peels

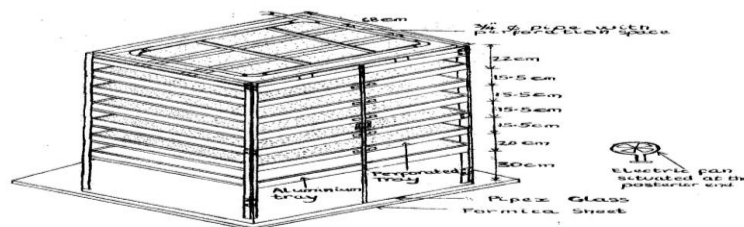


Figure 2: Schematic Isometric Projection of On-Farm Scale Fermenter

Table 1: Chemical Composition of *Trichoderma viride* Enriched Cassava Peel during Solid-State Fermentation at Laboratory and On-Farm Scales.

Composition (%)	Fermented Cassava Peel Samples		
	Unfermented cassava peel	Laboratory Scale	On-farm Scale
Crude Protein	4.21 ^b	10.43 ^a	10.93 ^a
True Protein	1.36 ^b	7.90 ^a	8.60 ^a
Moisture	8.73 ^a	7.69 ^a	7.72 ^a
Crude Fat	1.37 ^a	1.20 ^b	1.22 ^b
Crude Fibre	8.46 ^a	6.37 ^b	6.15 ^b
Ash	3.27 ^b	7.82 ^a	7.83 ^a
Carbohydrate by Difference.	91.15 ^a	80.55 ^b	80.02 ^b
HCN(mg/100g)	0.72 ^a	0.21 ^b	0.23 ^b
Starch	51.93 ^a	34.59 ^b	32.42 ^c
Gross Calories (kcal/100g)	393.79 ^a	374.72 ^b	374.78 ^b
Total Dietary Fibre	24.96 ^a	13.90 ^b	11.92 ^c

Results are expressed on a dry matter basis. Each value is a mean of three independent experiments.

Means followed by the same superscript in the same row are not significantly different ($p \leq 0.05$)

Compositional Analysis of Fermented Cassava Peel at Laboratory and On-Farm Scale

At the laboratory scale, crude protein, true protein and ash increased from 4.21 to 10.43%, 1.36 to 7.90%, 3.27 to 7.82%, respectively while crude fat, crude fibre, carbohydrate, cyanide, starch, gross calorie and total dietary fibre reduced from 1.37 to 1.20, 8.46 to 6.37%, 91.15 to 80.55 % 0.72 to 0.21 mg/100g, 51.93 to 34.59%, 374.72 kcal/100g and 24.96 to 13.90 % respectively. At the on-farm scale crude protein, true protein, and ash increased to 10.93%, 8.60% and 7.83% respectively while crude fat, crude fibre, carbohydrate, cyanide, starch and total dietary fibre reduced to 1.22%, 6.15%, 80.02%, 0.23 mg/100g, 32.42%, 374.78 kcal/100g and 11.92% respectively. Higher crude protein, true protein and ash content were observed in the on-farm scale when compared with the laboratory scale. However, there was no significant ($p \leq 0.05$) difference in the measured parameters between laboratory and on farm scale fermented cassava peel except in the starch and total dietary fibre.

This is in agreement with the results of Noorhom *et al* (1992) during the solid state fermentation of cassava with *Aspergillus niger* at laboratory and on-farm scale. The higher protein content may be due to better aeration provided by the fan as well as maintenance of high relative humidity in the fermenter (Noorhom *et al.*, 1992). The increase in the protein content could also be attributed to the possible secretion of some extracellular enzymes (proteins) such as amylases, linamarase and cellulose (Obloh *et al.*, 2003) into the cassava mash by the fermenting organisms (Raimbault, 1998), as well as increase in the growth and proliferation of the fungi complex in the form of single cell proteins (Antai and Mbongo, 1994; Obloh *et al.*, 2000). However, there were significance differences ($p \leq 0.05$) in the chemical composition of unfermented and fermented cassava peel.

CONCLUSION

The on-farm scale fermenter achieved fermentation of cassava peel comparable to laboratory scale but with an advantage of larger capacity in each fermentation batch. Consequently, an on-farm scale fermenter can be used in rural areas to handle relatively large amount of cassava peel in processing sites and rural farm settlements. The fermenter can also be adapted as a solid-state fermentation frame for many other indigenous substrates.

Further studies on improvement of the constructed on-farm scale fermenter in terms of sterility, process control and size should be carried out.

ACKNOWLEDGEMENT

The authors acknowledge the funding support of the University of Ibadan Senate Research Grant given to O.C Aworh to carry out this project.

REFERENCES

- Abou-Zeid A. A. (1991). Increasing the protein content of palm by-products. *Bioresource Technology* 37: 239-242.
- Akinrele, I.A.(1967). Further studies on the fermentation of cassava. *Research. Report No.20 of the Federal Institute of Industrial Research, Oshodi, Nigeria*. Antai, S.P. and Mbongo, P.M. (1994). Utilization of cassava peels as substrate for crude protein formation. *Plant Foods Human Nutrition* 46(4): 345-351.
- A.O.A.C. (1990). *Official methods of analysis*. 13th edition. Association of Official Analytical Chemists. Washington, DC. Awoyele, O.O. (2010). Fermentation of cassava peel with *Trichoderma viride* and *Candida utilis* for protein enrichment. *Ph.D Thesis Department of Food Technology, University of Ibadan, Ibadan, Nigeria*.
- Cooke R.D. (1978). An enzymatic assay for total cyanide content of cassava (*manihot esculanta* Crantz). *Journal of Science Food and Agriculture* 29:345-352.
- De Gregorio, A., Mandalari, G., Arena N, Nucita, F., Tripodo, M.M. and Lo Curto, R.B. (2002). Single cell protein and crude pectinase production by slurry-state fermentation of lemon pulps. *Bioresource Technology* 83(2):89-94.
- Dubois, M., Gilles, K. A., Hamilton, J. K., Rebers, P. A. and Smith, F. (1956). Colorimetric method for determination of sugars and related substances. *Analytical Chemistry* 28.3: 250-356.
- Giraud, E., Brauman, A., Keleke, S., Lelong, B. and Raimbult, M. (1991). Isolation and physiological study of an amyolytic strain of *Lactobacillus Plantarum*. *Applied Microbiology and Biotechnology* 36: 379-383.
- Gomez, J., Pazos, M. Couto, S.R. and Sanroman, M.A. (2005). Chestnut shell and barley bran as potential substrates for laccase production by *Coriolopsis rigida* under solid-state conditions. *Journal of Food Engineering* 68: 315-319.
- Hahn, S. K. and Keyser, J. (1985). Cassava: A basic food in Africa. *Outlook on Agriculture*. 4: 95-100.
- Hongpattarakere, T. and H-Kittikun, A. (1995). Optimization of single cell-protein production from cassava starch using *Schwanniomyces castelli*. *World Journal of Microbiology and Biotechnology* 11, 607-609.
- Iyayi, E.A. and Losel D.M. (2001). Protein Enrichment of cassava By-Products through Solid State Fermentation by Fungi. *The Journal of Food Technology in Africa* 6.4: 116-118.
- Iyayi, E.A. and Tewe, O.O. (1988). Effect of protein deficiency in utilization of cassava peel by growing pigs. *Cassava as Livestock feed in Africa*. Eds.S.K. Haha, L. Reynolds and G.N. Egbunike. Proceedings of the IITA/ILCA/University of Ibadan workshop. 14-18 November. 54-59.
- Lowry, O. H., Rosebrough, N. J., Farr, A. L., Randall, R. J. (1951). Protein Measurement with Folin-phenol reagent. *Journal of Biological Chemistry* 193: 265-275.
- Nestel, B. 1973. Current utilization and future potential for cassava. *Chronic Cassava Toxicity*. Eds. Nestel, B., Macintyre, R. Ottawa: International Development Centre. 11-26.
- Noomhorm, A., Ilangantileke, S. and Bautista, M.B. (1992). Factors in the Protein Enrichment of Cassava by Solid State Fermentation. *Journal of Food and Agriculture* 52: 117-123.
- Oboh, G., Akindahunsi, A.A. and Oshodi, A.A. (2000). Nutrient and anti-nutrient content of *Aspergillus niger* fermented cassava products (flour and garri). *Journal of Food Composition and Analysis* 15(5): 617-622.
- Oboh, G. and Akindahunsi, A. A. (2003). Biochemical changes in cassava products (flour and garri) subjected to *Saccharomyces cerevisiae* solid media fermentation. *Food Chemistry* 82(4):559-602.
- Onilude, A.A., (1994). Production, characterisation and utilization of some dietary fiber degrading enzymes as additives in Broiler diets. *Ph.D*.

- Thesis, Department of Botany and Microbiology University of Ibadan, Nigeria.*
- Paul, D., Mukhopadhyay, R., Chatterjee, B. P and Guha, A. K. (2002). Nutritional profile of food yeast *Kluyveromyces fragilis* biomass grown on whey. *Applied Biochemistry and Biotechnology* 97: 209-218
- Raimbault, M. (1998). General and Microbiological aspects of Solid Substrate Fermentation. *Electronic Journal of Biotechnology: Process Biotechnology* 1: 3. Accessed 23/03/2006
- Rajoka, M. I. (2005). Production of single cell protein through fermentation of a perennial grass grown on saline lands with *Cellulomonas biazotea*. *World Journal of Microbiology & Biotechnology* 21:207-211
- Rosenberg, E. (1993). Exploiting microbial growth on hydrocarbons: new markets. *Trends in Biotechnology* 11: 419-424.
- Subrahmanyam, D. (1990). Processing: Fermented Foods of Cassava. *Food Laboratory News* No.21, 6 (3): 9-12.