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Fermentation Studies on Roselle (*Hibiscus Sabdariffa*) Calyces Neutralised with Trona

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ABSTRACT

The effect of trona on the fermentation of roselle calyces was evaluated. The addition of trona to the calyces raised the initial pH from 3.3 to 5.3. The important microorganisms of roselle calyces fermentation were enumerated, isolated and identified. The fungi isolated consist of one yeast identified as *Saccharomyces cerevisiae* and two moulds, *Apergillus niger* and *Aspergillus flavus*. Of the aerobic bacteria, only *Escherichia coli*, *Bacillus subtilis* and *Klebsiella* sp. were isolated. The total titratable acid (TTA) value increased throughout the fermentation (0.027-0.043). The nutritional evaluation showed that there was an increase in the protein (6.56%), lipid (4.25%) and carbohydrate contents (74.33%) of the fermented sample with a higher ash content (4.54%) in the unfermented sample. The mineral contents (PPM); Zn (31.86), Fe (12.35), Mg (132.00), Ca (294.31), Na (214.62) and K (382.13) of the fermented sample were generally low. This study reveals that trona can be used to neutralize the acid in roselle calyx and that fermentation can greatly influence the nutritional composition positively.

Key words; Roselle, calyces, Trona, nutritional evaluation, fermentation

INTRODUCTION

Fermentation processes are those in which microorganisms contribute to the production of substances or materials which have well defined or potential commercial value (Brock *et al.*, 1994). Pederson (1971) described fermentation as a complex chemical transformation of organic substances brought about by the catalytic action of enzymes either native or elaborated by the microorganisms fermenting the raw materials (fruits, vegetables, plant materials etc.)

Roselle (*Hibiscus Sabdariffa*) is an herbaceous upright plant specie of malvaceae family growing up to 2m. The vegetable is widely grown in the North- Eastern and Middle belt regions of Nigeria (Akanya *et al.*, 1997). Two botanical varieties are recognized and have been consumed over a long period of time; these are red calyx varieties used for juice and green calyx used to cook soup, stew and sauces. The fresh roselle (green) calyxes is very rich in carbohydrate with relatively high fibre content and contains enough moisture to permit action by their own enzyme and microorganisms. Babalola (2000) reported that the calyx is very high in vitamin C and riboflavin with some major minerals present. Roselle finds many uses in traditional medicine as a digestive and purgative agent and a folk remedy for abscesses, billows, cancer, hypertension etc. (Duke, 1985). Currently in Western Nigeria Roselle calyx used in cooking vegetable soups is usually prepared by steeping it with wood ash over night or parboiled with wood ash to neutralize the acid is it.

Trona is the second most used salt in Nigerian homes. It is a hydrated sesquicarbonate of sodium and it is highly basic (Makanjuola and Beetlestone, 1975). It is used as a tenderizing agent of food materials such as cereals, pulses, meat and vegetables and usually added to these foods during cooking (Ankrah and Dovlo, 1978). This study is aimed at evaluating the possibility of neutralizing roselle calyx with trona and to study the effect of fermentation on the nutrients and some nutritionally valuable materials.

MATERIALS AND METHODS

Sample source and preparation The green roselle calyx which was dry and trona were obtained from Oja Oba market, Akure. The roselle herbs (300g) were soaked with trona (3g) in warm water for few minutes and thereafter washed with several changes of water until a clean calyx free from the neutralizing agent was obtained. The sample was divided into two (2) sub groups A and B. Group A was allowed to ferment for 3 days at room temperature while group B was not fermented.

Microbial analysis

The method according to Akinrele (1990) was used based on the plate dilution technique. Oxoid nutrient agar and malt extract agar used for the isolation of bacteria and fungi were prepared according to the manufacturer instruction (Oxoid). Aliquots of the samples and nutrient were mixed and poured aseptically into sterile petridishes for incubation at 30°C for bacteria and at 25° for Fungi. The number of colonies growing in each plate was counted after 48h of incubation and classified by microscopic examination of colony morphology, after separation and growth on slants.

Physicochemical changes

The pH of each sample was measured every day with a Cambridge direct reading pH meter. Total titratable acidity (TTA) was determined on 5 ml aliquot of the sample against 0.01N. NaOH, using phenol red as indicator.

CHEMICAL ANALYSIS

Proximate analysis

Proximate analysis of the samples were performed according to AOAC (1990) procedures for ash, crude fibre, fat, moisture and protein using a nitrogen to protein conversion factor of 6.25. Carbohydrate was determined by difference.

Mineral analysis

The nutritionally essential elements (Na, Ca, Mg, K, Fe and Zn) were determined using Atomic Absorption Spectrophotometer (ASS).

RESULTS AND DISCUSSION

The results given in **Table1** shows that a wide variety of microorganisms were isolated from the fermenting roselle. The bacteria isolates recognized were *Escherichia coli*, *Bacillus subtilis* and *Kiebsiella* sp., while the fungal isolates were *Aspergillus niger*, *Aspergillus flavus* and *Saccharomyces cerevisiae*. *Klebsiella* sp. and *E. coli*

were eliminated early during the fermentation as their growth was probably inhibited by unfavourable low pH. Okafor (1977) in his study on the fermentation of palm wine also reported that *Klebsiella* sp. and *E. coli* disappeared early in fermentation, as they are sensitive to acid and alcohol.

The occurrence of *B. subtilis* throughout the fermentation period confirms that it grows in close association with food substrate and elaborate extracellular enzymes during fermentation (Aderibigbe, 1997). The roles of the fungi; *A. niger* and *A. flavus* isolated throughout the fermentation seem to be associated with their amylolytic activity (Balagopal and Maini, 1976). *S. cerevisiae* was present throughout the fermentation period being a fermenting organism capable of growing and surviving in acidic and alcohol medium (Ayanru *et al.* 1985).

Table 2 shows the physiochemical changes that occurred during the fermentation of the rosette calyx. An initial pH value of 3.320 recorded for the fresh roselle calyx increased to 5.310 due to the neutralizing effect of the trona. The pH dropped from 5.310 at zero day to 4.280 by the third day and TTA increased throughout with time. The decrease in pH with increase in TTA is due to the increased activities of the microorganisms resulting in production of organic acids from available nutrients (Okafor, 1978).

The proximate composition of fermented and unfermented roselle in **Table 3** reveals that the protein and lipid content of the fermented sample was higher (6.56% and 4.25%) compared to the unfermented (5.80%) and (4.09%). The increase in protein and lipid contents may be due to the fact that the microorganisms which fermented the roselle, may have secreted extracellular enzymes which consequently increases the protein and lipid contents (Kao and Robinson 1978). The great decrease in crude fibre content of the fermented roselle may be due to the activities of the saprophytic mould which synthesize cellulose enzyme that breaks down the cellulose in the vegetables leading to an increase in the carbohydrate value of the fermented sample.

As shown in **Table 4**, there was a decrease in the mineral content for the fermented sample due to utilization by the fermenting organisms. The study therefore reveals that trona can be used to neutralize the acid in the roselle and that fermentation greatly influences the chemical composition positively by increasing the protein, carbohydrate and lipid content.

References

- **Aderibigbe, E.Y. (1997).** Characteristics of extra cellular proteinases from strains of *Bacillus subtilis* Group. Nig. J. Microbial 11: 93-97.

- **Akanya, H.O., Oyeleke, S.B. Jigam, A.A. and Lawal, F.F. (1997)** Analysis of sorrel drink. Nig. J. Biochem. 12: 77-79.
- **Akinrele, L.A. (1990).** Fermentation studies on maize during the preparation of a traditional African starch-cake food. J Sci. Agric. 21, 619-624.
- **Ankrah, E. E. and Dovlo, G. F.E. (1978).** The properties of trona and its effect on the cooking time of cow pea. J. Sci. Ed. Agric. 20, 950-952.
- **AOAC, (1990).** Official Methods of Analysis (15th Ed.) Association of Analytical Chemists. Washington D.C. USA.
- **Ayanru, D.K.G., Sharma, V. C. and Ogbeide, O. N. (1985).** Effects of the quality and type of microorganisms on ethanol production from papaw. Energy. 10: 1000-1016.
- **Babalola S.O. (2000).** Chemical analysis of roselle leaf (*Hibiscus Sabdariffa*)¹, in proceedings of 24th annual conference of NIFST, pp.228-229.
- **Balagopal, C. and Maini, S.B. (1976).** Evaluation of certain species of fungi for single cell protein and amylase in cassava liquid medium. J Root Crops. 2,49-51.
- **Brock T.D., Madigan M. T., Martinko J. M. and Parker, J. (1994).** Biology of microorganisms. 7th Ed. Prentice Hall Int., USA, pp 282.
- **Duke. Y.A. (1985).** Handbook of medicinal herbs. 13th Ed. Livingstone Group Ltd., Edinburgh, pp 228-229.
- **Kao. C. and Robinson, R. J. (1978).** Nutritional aspect of fermented food from chickpea and soybeans¹. Cereal chem. 55; 512.
- **Makanjuola A. A. and Beetlestone, J. O. (1975).** Some chemical and mineralogical notes on kaun (trona). J. Min. and Geol. 10: 31- 41.
- **Okafor N. (1977).** The microbiological basis of a method for palm wine preservation J Appl. Bact. 43:159-161.
- **Okafor N. (1978).** Microbiology and biochemistry of oil palm. Ad. Appl. Microbiol. 24: 237-256.
- **Pederson, C. S. (1971).** Microbiology of food fermentations. 2nd Ed. AVI Pub Co. Inc., Westport, Conn. 537pp.

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Table 1: Occurrence of microbial isolates during fermentation

Microorganism	Hours of fermentation			
	0	24	48	72
<i>Bacillus subtilis</i>	+	+	+	+
<i>Escherichia coli</i>	+	+	-	-
<i>Klebsiella</i> sp.	+	+	-	-
<i>Saccharomyces cerevisiae</i>	+	+	+	+
<i>Aspergillus niger</i>	+	+	+	+
<i>Aspergillus flavus</i>	+	+	+	+

Table 2: Physio-chemical changes during fermentation of roselle

Hours of fermentation	TTA	pH
0	-	5.310
24	0.027	4.600
48	0.036	4.420
72	0.043	4.280

Table 3: Promate composition (%) of fermented and unfermented roselle

Properties	Fermented	Unfermented
Moisture	9.52	9.87
Fat	4.25	4.09
Protein	6.56	5.80
Crude fibre	1.24	9.50
Ash	4.10	4.54
Carbohydrate	74.33	66.20

Table 4: Mineral content (PPM) of fermented and unfermented roselle

Properties	Fermented	Unfermented
Mg	132.00	336.74
Zn	31.86	107.00
Fe	12.35	52.48
Na	214.62	414.54
K	382.13	441.60
Ca	294.31	936.00