

PRELIMINARY STUDIES IN THE PRODUCTION OF BUNYI-YOURI
(A PUTRIFIED SUNDRIED FISH PRODUCT)

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ABSTRACT

Fresh fish samples of Lates niloticus species obtained from Gwange fresh fish market, Maiduguri were used for laboratory and field studies. Fresh fish was collected aseptically, gutted, washed and allowed to ferment at ambient temperature of 43-49°C for 6-8 days. Total mesophilic plate count, moisture content, trimethylamine (TMA) values, and pH were determined. The micro-organisms isolated were identified using standard methods. The proximate compositions of the fresh and fermented products were also evaluated.

Results indicated two main stages of processing: the spoilage or putrifactive stage which was characterized by high microbial count and TMA values and the fermentative stage which was characterized by decrease in pH, moisture content, microbial count and TMA values. After 8 days of processing, there was a significant decrease ($P < 0.05$) in moisture content followed by corresponding increase in the amounts of fat, protein and ash when compared to the fresh unprocessed samples. Of the microbial isolates 80% were of micrococci and 20% staphylococci. All the staphylococcal isolates were coagulase negative.

Key Words: Preliminary studies, Bunyi-youri, Putrified Sundried Fish.

INTRODUCTION

Fish is a rich source of lysine and other amino acids and is therefore suitable for complementing high carbohydrate diets, especially in developing countries (Kent, 1984).

Consumption of fermented food has been an age old practice in many parts of the world (Van Veen, 1962). This is either used as condiment or as a main meal (Wood and Min, 1975; Vishwanath and Sarojnalini, 1989). In Nigeria this practice is common among

the Kanuris of Northern Nigeria, where Bunyi-youri (a putrified sun-dried fish) prepared from Lates niloticus or Clarias species is used as a condiment in the flavouring of soups. The initial period of processing could attract a lot of flies as a result of the unpleasant odour emitted from the fish. This could render the process unhygienic. It is possible that the final product may be contaminated or microbiologically unwholesome.

The skin of the freshly caught fish is known to be coated with slime and it is associated with various organisms. Liston (1979) reported the presence of Moraxella, Achromobacter, Aeromonas, Enterobacter and Bacillus on the skin of fish, none of which are pathogenic to man. However, other pathogenic and toxigenic organisms which could be introduced to man as a result of poor handling of fish include Bacillus cereus, Staphylococcus aureus, Clostridium botulinum, Clostridium perfringens, Salmonella, Shigella and Vibrio parahaemolyticus (Johnson, 1987).

Although the processing of "Bunyi-youri" had been practised for years among the Kanuris of Northern Nigeria, no attempt has been made to investigate the nature of micro-organism involved in the processing of "Bunyi-youri" or the proximate composition of the product.

This study was therefore designed to investigate the nature of micro-organisms involved in the processing of "Bunyi-youri" and the proximate composition of the product.

MATERIALS AND METHODS

Collection of Samples:

Fresh fish samples mainly Lates niloticus species were obtained from fresh fish market along Gwange Road, Maiduguri. Samples were collected using ice chest containing crushed ice to minimize deterioration of the fish during transportation to the laboratory. This served as the laboratory samples. Similarly samples were taken to a local processor in Monday market, this served as the field study.

Samples Preparation:

The fish sample was cut open longitudinally from one side through the ventral surface. It was gutted and thoroughly washed with potable water before being allowed to putrify at ambient temperature of 43-49°C and relatively humidity of 16-18% for 6-10 days. The experiments were carried out under both field and laboratory conditions and subjected to chemical and microbiological analyses.

Microbiological Analysis:

Twenty grams of each ground fish samples was weighed out and thoroughly mixed with 180ml nutrient broth and serially diluted up to 10^6 . Duplicate plates were plated out from each dilution by the pour plate technique using nutrient agar, blood agar and mannitol salt agar media, with incubation at 32°C for 24 to 48 hour to determine the total aerobic plate count as described by Collins and Lyne (1970); Harrigan and McCance (1976). The samples were analyzed on Day one and every two days for a period of 8-10 days.

The colonies were isolated, purified and subjected to microbiological and biochemical tests as shown in Table 3. Identification of the isolates were carried out by comparison with standard diagnostic tables (Cowan and Steel 1961; Collins and Lyne 1970).

Chemical Analysis:

The crude protein, ash and fat contents of the samples were determined as described by AOAC (1984) and the moisture content by the oven method (Pearson, 1981). The trimethylamine (TMA) value was carried out as described by Licke and Geidel (1955).

To determined the pH of the ground samples, five grams was thoroughly mixed with 20ml of distilled water. The pH of the resulting slurry was determined using a pH meter (Kent E.C 7045/46).

Statistical Analysis:

Data was subjected to analysis of

variance (Steel and Torrie, 1980). Duncan's multiple range test and multiple F test (Duncan, 1955) was used to separate the differences among the means.

RESULTS AND DISCUSSION

The dimensions of the fish used in this study are shown in Table 1. The processing appears to be divided into two phases. The first phase was the 'spoilage' or putrefactive stage followed by the fermentative phase. The spoilage phase was characterized by high trimethylamine (TMA) value and high total aerobic plate count on days 2 and 4 (Table 2). The trimethylamine (TMA) values from the field studies were generally higher than those from the laboratory studies. This is probably due to the high initial microbial load in the field samples and the higher ambient temperature of 42°C (Table 5) when compared to 32°C in the laboratory.

Fish spoilage started immediately it was removed from water. Enzymatic and microbial activities played a major role in this decomposition process (James, 1970). According to Shewan (1961) slime from the skin is composed of mucopolysaccharide compounds, free amino acids, trimethylamine oxide, pepridine derivative and other related compounds. The spoilage organisms first utilized the simpler compounds and in the process released various volatile off-odour components. These off-odour compounds are probably responsible for attracting flies to the processing site as was observed during the field processing.

The second stage or fermentation phase was characterized by decrease in moisture content, TMA values, total aerobic plate count and pH (Table 2, Fig. 2). The decrease in moisture content during processing was a result of the relatively low humidity and high ambient temperature of the environment (Table 5). This decrease in moisture content led to a decrease in water activity of the samples to a level at which most micro-organisms would not proliferate

favourably in the product (Asdel *et al.*, 1973). Troller (1979) has shown that the lower limits of water activity for growth of some bacterial including micrococcus is about 0.96 to 0.97. The progressive decrease in microbial count which was higher for field samples when compared to the laboratory samples may be related to the higher moisture loss from the field samples (Table 2).

Microbial Isolates:

There was little or no difference in the number or nature of isolates between those of the field and laboratory samples. So samples from both locations were pooled together. There were six isolates labelled G, P, LY, W, M and LG (Table 3). In both studies all isolates were gram positive cocci. Isolates G and M increased ($P < 0.05$) after 8 days of processing, while little or no increase was observed in the other isolates (Figure 1). From this result it seemed isolates M and G were the organisms which played significant roles in the processing of "bunyi-youri".

Table 3 shows the biochemical characteristics of the isolates involved during the processing of "Bunyi-youri". From the biochemical studies 80% and 20% of the isolates were micrococci and staphylococci species respectively. The staphylococcal isolates were coagulase negative suggesting they may not be pathogenic. These organisms might be responsible for producing the unique flavour and colour of "bunyi-youri". According to Coretti (1977), starter cultures such as micrococci and harmless staphylococci are important in meat fermentations because of their catalase activities and the production of unique flavour and colour development. Although the functions of isolates LY, LG, W, and F are not clear, it is possible they help to retard the proliferation of undesirable micro-organisms. Baccus and Brown (1981) reported that the presence of high number of harmless bacteria seems to retard the development of pathogens through the generation of antibiotics, bacterocins and

peroxides. It is possible that using a similar mechanism, the proliferation of pathogenic organisms may have been inhibited in "bunyi youri", although the initial stages of processing attracts flies and looks unhygienic.

Proximate Composition:

The proximate compositions of both fresh and fermented fish are shown in Table 4. After fermentation, there was a significant increase ($P < 0.05$) in the amount of protein, fat and ash in the processed "bunyi-youri" but a decrease in the amount of moisture content of the fermented product when compared with the fresh unfermented product. This decrease in moisture content could have led to a concentration of nutrients in the finished product (bunyi-youri). Carbohydrate analysis of the processed fish revealed little or no carbohydrates and this finding agrees with Sharp (1934) who demonstrated that fish muscle is very poor in glycogen content following post mortem. The poor glycogen content was probably responsible for relatively small decreases in pH from 6.9 and 6.8 in both field and laboratory samples respectively to 6.1 after 6 and 8 days of fish processing both in the laboratory and in the field (Figure 2).

CONCLUSION

The method of processing "Bunyi-youri" is effective in enhancing the microbial quality of the product as it leads to a concentration of nutrient by decreasing the moisture content of the final products with a better shelf-life. The presence of staphylococci and micrococci species suggest that they are the primary organisms responsible for producing the unique flavour associated with "Bunyi-youri".

REFERENCES

- AOAC (1984). Official Methods of Analysis. Assoc. Official Analytical Chem. 12th Ed. Washington D.C.
- Asdel, W.G.; Gopley, M.T. and Morgan, A.I. (1973). Food Dehydration, Vol. 1 2nd Edition, Avi Pub. Co. Westport Con. pp.246-250.
- Baccus, J.M. and Brown, W.L. (1981). Use of Microbial cultures in meat products. Food Technol. 35(1): 74-76.
- Collins, C.H. and Lyne, P.H. (1970). Microbiological Methods 3rd Ed. Butterworth and Co. (Pub) Ltd. London Univ. Park Press Baltimore Great Britain pp. 51-207.
- Coretti, K.U. (1977). Starter culture in the meat industry, Die Fleischirrschaft. 3: 386-391.
- Cowan, S.T. and Steel, K.J. (1961). Diagnostic tables for the common medical bacteria. J. Hyg. Cambridge, 59 : 357.
- Duncan, D.B. (1955). Multiple range test and multiple F test. Diometrics II : 1-42.
- Harrigan, W.F. and McCance, M.E. (1976). Laboratory Methods in Food and Dairy Microbiology. Academic Press, London.
- James, M.J. (1970). Modern Food Microbiology. McGraw Hill, New York. pp. 20-40.
- Johnson, A.M. (1987). Microbiological studies of the Nigerian marine fish species. A preliminary investigation into the microflora of the skin. J. Food Agric. 1: 47-49.
- Kent, G. (1984). National fishery policies and alleviation of malnutrition in the Philippines and Thailand. F.A.O. Fisheries Circular No. 777.
- Liston, J. (1979). Flavour of fermented fish sauce. Techn. J. Agric. Food Chem. 31(2) : 93-94.
- Pearson, J.D. (1981). Chemical Analysis of Food 8th Ed. Churchill Livingstone, Edinburgh, London and New York.
- Sharp, J.D. (1934). Postmortem breakdown of glycogen and accumulation of lactic acid. Proc. Royal Soc. London B. 114. pp. 506-512.
- Shewan, J.M. (1961). The Microbiology of Sea water fish. Fish as food 1: 487-560. Acad. Press London.
- Steel, R.G.D. and Torrie, J.H. (1980). Principles and Procedures of Statistics, McGraw-Hill Book Co. Inc., New York.
- Troller, J.A. (1979). Food Spoilage micro-organism tolerating low water activity environments. Food Technol. 15 : 72-75.

- Van Veen A.G. (1962). Sea food products in South East Asia. In "Fish as Food" Vol. 2 Borgstron G. (Ed.) Acad. Press London.
- Vishwanath, W. and Sarojnalini C.H. (1989). Changes in the total oxalate content in the fermentation of fish paste Hentak. J. Food Sci. 54: 754-755.
- Wood, B.J.B., and Min, Y.F. (1975). Oriental foodfermentations. In "Filamentous Fungi" Vol. 1 Smith, J.E. and Berry, D.R. (Ed) Edward Arnold, London.

Table 1: Dimensions of the fish (Lates niloticus) used during fermentation of "Bunyi-youri"¹

Parameter	Field Study	Laboratory Study
Average length (cm)	42.00 ± 0.20	39.25 ± 1.01
Average width (cm)	12.00 ± 0.15	10.75 ± 5.05
Average weight (kg)	1.20 ± 0.25	1.00 ± 0.47

¹ Each value is a mean and standard deviation of five determinations.

Table 2: Bacterial count, trimethylamine value and moisture content during the processing of Bunyi-yourri^{1,2}

DAYS	Log Bacterial Count		TMA values (ml.eq/100g)		Moisture content(%)	
	Field study	Lab. study	Field study	Lab. study	Field study	Lab study
0	4.68±0.01a	3.82±0.12b	87.5±3.5a	69.24±1.49b	70.12±1.20a	68.82±21a
2	6.66±0.50b	7.61±0.12a	136.5±10.5a	97.65±0.50b	58.32±0.15a	59.12±1.31a
4	7.61±0.70a	6.59±1.20b	65.1±2.2b	98.60±1.98a	20.47±0.78b	42.61±2.14a
6	4.89±1.21a	4.67±2.71a	60.92±2.7	66.5±0.99a	18.50±1.21b	25.67±1.50a
8	N.D	3.28±1.31	N.D.	31.5±4.95	N.D.	24.80±2.55

¹Each value is a mean of five determinations.

²Means within a row with unlike letters differ significantly (P<0.05)

N.D., not determined.

Table 3: Summary of some physical and biochemical characteristics of microbial isolates of Bunyi-yourri during fermentation

Iso-lates	Gram stain and Morphology	Pigmentation	Motility in 0.5% Agar	High and Leifson Test	Oxi-dase	Coagu-lase	Cata-lase	Possible genus
G	Gram positive cocci	Gold	-	Ferm	-	-	+	<u>Staphylococcus</u>
LY	Gram positive cocci	Light	-	Oxi	-	-	+	<u>Micrococcus</u>
M	"	Yellow/White	-	Oxi	-	-	+	<u>Micrococcus</u>
LG	"	Cream	-	Oxi	-	-	+	<u>Micrococcus</u>
W	"	Light	-	Oxi	-	-	+	<u>Micrococcus</u>
P	"	Cream Pink	-	Oxi	-	-	+	<u>Micrococcus</u>

Oxi - Oxidative

Ferm - Fermentative

- - Negative reactions

+ - Positive reactions

Table 4: Proximate compositions of fresh and fermented Lates niloticus^{1,2}

Parameter Determined	Fresh Fish (%)	Fermented Fish (%)
Protein	18.36 ± 0.53a	32.62 ± 1.05b
Fat	11.84 ± 0.55a	40.59 ± 0.12b
Ash	0.85 ± 0.34a	1.88 ± 0.32b
Moisture content	68.82 ± 0.30a	24.80 ± 2.55b

¹Each value is a mean of 4 determinations

²Means within a row with unlike letters differ significantly (P < 0.05)

Table 5: Environmental conditions during the fermentation of Lates niloticus

Days	Relative Humidity (%)	Temperature °C		Wind Speed (MS ⁻¹)
	Field	Field	Lab.	Field
0	18	41	32	7.97
2	16	44	33	7.98
4	17	43.0	34	7.99
6	18	40	33	7.97
8	N.D	N.D	35	N.D

N.D. = not determined

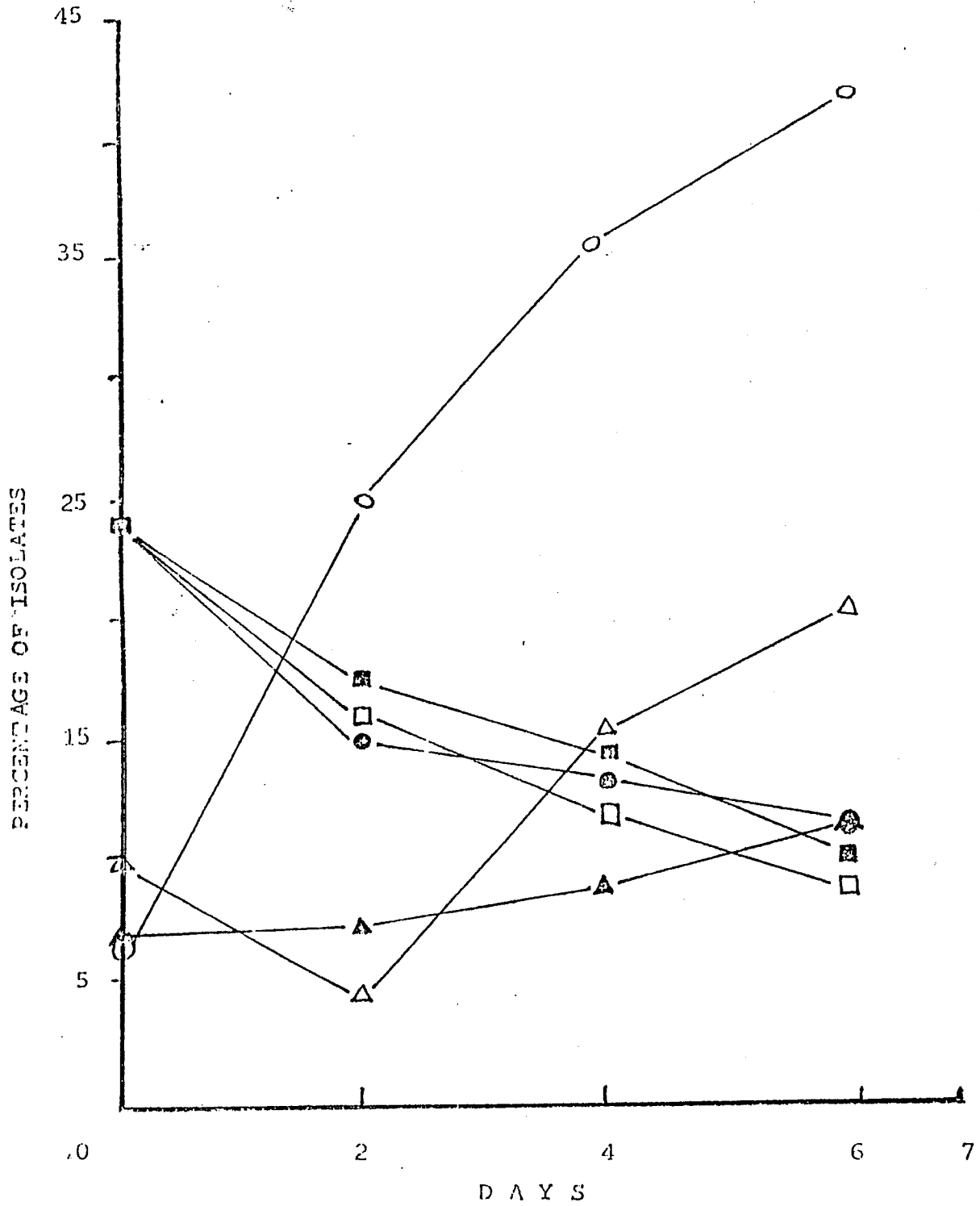


Figure 1. Percentage representation of isolates during processing of Bunyl youri.

△, G; ▲, LY; ○, M; ●, LG; □, W; ■, P.

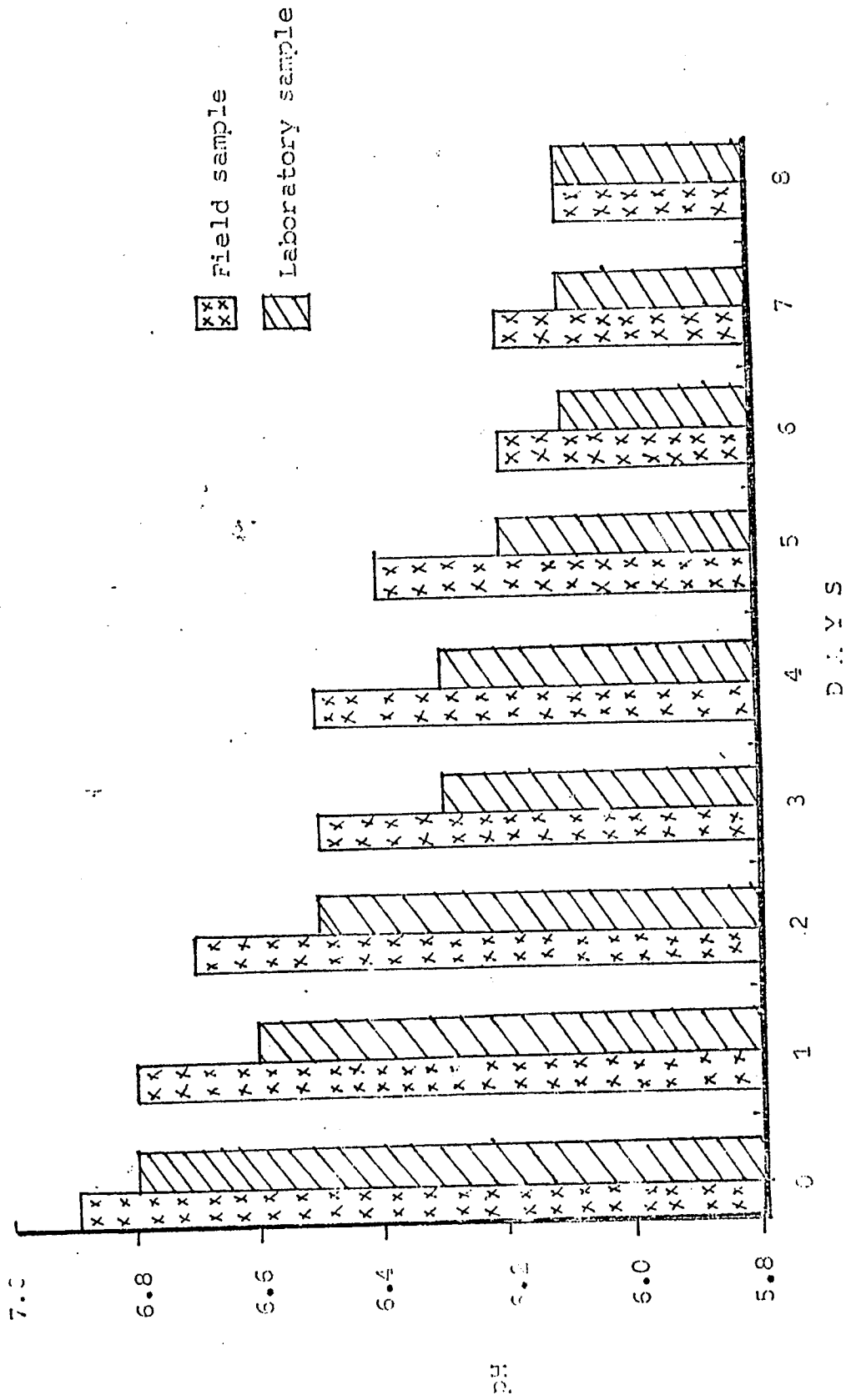


Figure 2. pH changes during the processing of Sunvi youni.