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## COUNCIL FOR SCIENTIFIC AND INDUSTRIAL RESEARCH



Studies on Traditional Processing and Quality of Fermented Fish.

Ghana/Netherlands Artisanal Fish Processing Project Report No. AFPP/AR/PH.2/94-001. FRI. Accra, Ghana.

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## GHANA/NETHERLANDS ARTISANAL FISH PROCESSING & APPLIED RESEARCH PROJECT

### RESEARCH PROJECT # AFPP/AR/Ph.2/94-001

STUDIES ON TRADITIONAL PROCESSING AND QUALITY OF FERMENTED FISH (MOMONE).

### FINAL REPORT

### By

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### ABSTRACT

Traditional momone fermentation sites were surveyed to examine the processing facilities and practices. Fermented fish samples from the sites were analysed for their chemical, microbial, sensory and physical quality. These were kingfish (<u>Scomberomorus tritor</u>) and jack mackerel (<u>Caranx hippos</u>).

Fifteen, 25 and 40% of salt to fish weight ratio were prepared with kingfish for fermentation trials to determine appropriate salt concentrations. Qualitative and quantitative microbial enumeration as well as chemical and sensory evaluation were carried out.

Storage studies on vacuum sealed momone from kingfish which had been given the following treatments :1% vinegar, 3% vinegar, 20% garlic extract dips, were carried out for a month. Untreated vacuum sealed momone served as the control.

Exudate from fermentation was concentrated to about 50% of its original volume and used as a condiment.

Poor hygienic practices and unclean equipment at the traditional processing sites were considered as potential sources of contamination, although fermented fish from these sites were found to be microbilogically safe. No organisms of public health significance were isolated.

A 25% salt to fish weight and over was considered appropriate for the production of momone. Micrococci and cocci were the predominant organisms isolated from the fermentation systems.

Vacuum packaged momone with either garlic or vinegar was not organoleptically affected and the growth of microorganisms also suppressed during storage.

Concentrated exudate could be used as a condiment during cooking.

### INTRODUCTION:

Fermentation has long been considered as one of the methods of preserving an otherwise highly perishable commodity. Although in fermentation, preservation remains the focus, the flavour enhancing effect of some fermented products are quite adequate to justify the process.

In Ghana and many other African countries, a number of dried products with mild to strong odour generally referred to as fermented commodity serve as protein sources. However, a typical Ghanaian fermented fish product <u>momone</u>, is widely used for its flavour enhancing properties. This intermediate moisture product is characteristically soft with a very strong flavour and it is sometimes considered offensive (Essuman 1992).

The processing of momone is largely artisanal and it is usually carried out to salvage large quantities of fish which would otherwise have been discarded due to poor quality. Other situations which could eventually lead to the processing of momone, are unmarketability or underutilization of the fish. Consequently, all kinds of raw fish irrespective of the quality are processed into fermented fish. Species used include catfish, barracuda, seabream, threadfin, croaker, grouper, bonito, mackerel, herrings, squid, octopus, bumper, snapper and ribbon fish.

Though the processes are simple, hygienic standards are generally poor and quality of products are varied.

Re-using of brine for subsequent fermentation could be a source of bacterial contamination. Partial drying of products on the ground could also be a source of bacterial, sand contamination and insect infestation. Products are also susceptible to larvae infestation, mould growth and bacterial spoilage. Microbial analysis of momone on sale showed a high microbial population of which 72% were gram positive micrococci and 17% for gram negative rods (Nerquaye-Tetteh <u>et al.</u> (1978). However no pathogenic organisms were isolated but, the poor quality of the raw fish before salting and insufficient acid production in low salt fermentation can cause the outbreak of botulism which though could be destroyed by cooking, is very stable in salty and acidic environments (Huss and Rye Pederson 1980). Other potential hazards associated with proteinaceous foods like fermented fish are presence of parasitic worms and production of physiologically active amines.

Yankah (1988), observed the growth of moulds on momone after 24 hours of fermentation and 24 hours of drying which were attributed to be contaminants from the atmosphere.

In addition, the wide variation in salt fish ratio may result in wastage of the salt. Since salt is the main preservative agent in this fermented product, a significant low amount would allow the growth of pathogenic or toxin producing microorganism which could pose a health hazard as mentioned earlier. However, heavy salting may lead to the loss of water soluble compounds including proteins (Gildbert 1991).

Despite the fact that there are no reported incidences of diseases associated with the consumption of momone, nevertheless, a convincing case could be made for the promotion of locally available foods and improvement on processing techniques. This is especially so in the artisanal fish industry where technology and standards are very low, coupled with fast depleting resources and limited income. Nerquaye-Tetteh <u>et al.</u> (1978) considered fermented fish as potential vehicles for transmission of food borne diseases.

Adoption of improved technology in the fermentation process and the use of good quality raw material would not only yield better products, but increased earnings would be expected by the processors. Subsequently, an improvement on their socioeconomic life could not be far fetched. This could be attained as consumers in general would have confidence in the product quality and patronize it.

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As could be inferred from the above, the objectives of this study are :

- i. To examine the traditional processes and determine their effect on the physical and microbial quality of the product.
- ii. To investigate the biochemical and microfloral changes in fermentation studies.

iii. To conduct storage studies on the product.

iv. To examine the possibility of using the exudate after fermentation as a fish sauce.

The study is to provide a basis for modification of the process leading to the design of an optimum fermentation conditions. Fermented fish is a value-added product and improvement in quality, packaging and presentation will achieve a long term objective of having the products on supermarket shelves in Ghana as in other parts of the world.

### 2. MATERIALS AND METHODS

### 2.1. Processing Sites.

A survey was undertaken at four processing sites to examine and follow the stages in the fermentation process. Sites visited were selected on recommendation from sellers as the current active processing areas as well as their proximity to the Institute.

Equipment, species, packaging, storage and any other materials used were noted.

Pictorial depictions of processing sites, equipment and products were also made. Informal discussions structured along the questionnaire in the Appendix, were held with processors for information on their activities.

Two fermented fish species were selected based on their popularity and the fact that they could be obtained almost round the year. These were jack mackerel (Caranx hippos) and Kingfish/Spanish mackerel (Scomberomorus tritor).

Their quality in terms of chemical, microbiological and physical characteristics were assessed.

2.2. Physical and Freshness Assessment

All samples subjected to physical assessment was by visual inspection. Sample colour and appearance were noted. International Standards tests of the Torry and EEC Schemes of freshness assessment were applied to the fresh fish before processing in order to ascertain the quality of the fish.

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### 2.3. Chemical Analysis

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where Aw = water activity
and ERH = equilibrium relative humidity (%).

### 2.3.2. Hydrogen Ion Concentration (pH)

The pH of the fish tissue was taken using a Metrohm 620 pH meter (Swiss-made). Approximately 10 g of fermented fish was weighed into 200 ml beakers and 90 ml of carbon dioxide-free distilled water was added and thoroughly macerated using a stomacher. The pH of the mixtures were then taken.

### 2.3.3. Total Volatile Base Nitrogen (TVBN)

Ten grams of fish flesh was macerated and added to 2g Magnesium oxide with 300 ml tap water in the distilling flask of macro-Kjeldahl distillation apparatus. The fermented fish samples' distillates collected in 2% Boric acid solution, were analysed for total Volatile Bases (TVBN) by the method outlined by Pearson (1970).

### 2.3.4. Crude Protein

Protein was determined by the method of Pearson (1970).

### 2.3.5. Total Chloride (Salt)

Salt was estimated by titrating against silver nitrate (AOAC 1990).

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### 2.3.6. Free Fatty Acids (FFA) AND TBA

Free fatty acids (FFA) were determined by the method of Pearson (1970). One gram of the crude fat extract from the fermented fish sample was mixed with a neutral solvent (25 ml diethylether added to 25 ml alcohol with 1 ml of 1% phenolphthalein solution neutralised with o.lN alkaline) and titrated with aqueous 0.lN NaOH. Thiobarbituric acid (TBA) value was measured according to Pearson 1970).

### 2.4. Microbiological examination

Microbiological analyses of fermented fish from the local processing sites as well as those at the Food Research Institute's Fish Laboratory were carried out for culture and quality evaluation.

### 2.4.1. Total Viable Count

The Pour Plate Technique was used in enumerating the total aerobic mesophilic flora per gram of sample of fish. The sterile polyethylene bags containing the samples of fish were opened over a bunsen flame. A 10 g portion of the fish was aseptically weighed and 90 ml Saline Peptone Solution (SPC) added and macerated using a stomacher. One ml was pipetted and used to prepare  $10^{-1}$  to  $10^{-6}$  serial dilutions. One ml of each dilution was then pippeted into sterile plates in duplicates. Each plate was overlaid with about 20 ml of Plate Count Agar (PCA) which had been cooled to  $45^{\circ}$ C. Thorough mixing was ensured by clockwise rotation of the plates. These were allowed to stand in order to solidify. The plates were then incubated at  $30^{\circ}$ C for 72 h (Anon. 1986).

### 2.4.2. Enterobacteriaceae (Coliforms)

By means of a sterile pipette, 1 ml of 10 and 10 dilutions of the fish suspensions were transferred into a sterile petri dish. Approximately 5 ml of melted Tryptone Soya Agar (TSA) cooled to 45<sup>°</sup>C was poured into the petri dish. The inoculum and the medium were thoroughly mixed by circular movements of the dish. After solidification of the medium, the plates were left at 25°C for 1 h. Following pre-incubation, 15 ml of melted Violet Red Bile Agar (VRBA) maintained at 45°C was poured on to the top. After solidification, the petri dishes were inverted and placed in an incubator at 30°C for 40 - 48 h. Characteristic colonies that are surrounded by a reddish zone and hence showing bile precipitation were verified by transferring a colony mass using a platinum wire into test tubes containing Brilliant Green Bile Broth. These were incubated at 30°C for 40 - 48 h. The presence of coliform bacteria was confirmed by the accumulation of gas in the Durham tubes (Anon. 1992a).

### 2.3.3. Yeast and Mould Counts

For the enumeration of yeast and mould, a low acid medium was used. This was prepared be sterilizing 250 ml of Potato Dextrose Agar (PDA) and adding 7.5 ml of sterilized acid (1.5 ml acid to 50 ml PDA). Employing the pour plate technique, 1.0 ml of the  $10^{-1}$  dilution of the fish suspension was pipetted into sterile petri dishes. This was overlaid with acidified PDA and thoroughly mixed. The plates were then incubated at  $25^{\circ}c$  for 5 - 7 days (Anon. 1987).

### 2.4.4. Pathogenic microorganisms

### 2.4.4.1. Staphylococcus sp.

A 5 g fish sample powder was aseptically weighed and placed in a cooked meat medium with 10% salt added. It was mixed thoroughly and incubated for 12 - 18 h at  $37^{\circ}$ C. Zero point one (0.1) of this

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was subcultured onto a Baird-Parker's medium and incubated for 24 and 48 h at 37 °C. Zero point one ml of this was subcultured onto Baird-Parker's medium and the inoculum distributed on the surface using a sterile angle bent rod and the plates incubated for 24 and 48 h at 37 °C for pure culture isolation and identification (Anon. 1992b).

### 2.4.4.2. Salmonella sp.

For pre-enrichment, 25 g sample fish was weighed and macerated in 225 ml of Buffered Peptone Water (BPW) using a stomacher. This was incubated at  $37^{\circ}$ C for 16 - 24 h. The contents were then mixed by shaking vigorously upon removal from the incubator, and 0.1 ml of the pre-enrichment broth was transferred to 10 ml Rappaport-Vassiliadis (RV) broth which has been prewarmed at  $42^{\circ}$ C. This was then incubated at  $42^{\circ}$ C for 24 h.

Using a loop, a sample from the enrichment broth was inoculated onto the surface of Xylose-lysine-desoxycholate agar (XLD) and Brilliant green-phenol red agar (BGA) so that well isolated colonies would develop. The plates were incubated in inverted position at 37<sup>o</sup>C for 18 - 24 h. Presumptive colonies isolated were then picked for biochemical verification. From each agar plate, at least two typical or presumptive colonies were picked and inoculated onto a suitable non-selective plates, so that well isolated colonies would develop. The plates were incubated at 37<sup>o</sup>C for 18 - 24 h. For the biochemical confirmation, tests carried out included urea, mannitol utilization, ornithine decarboxylase, lysine decarboxylase and Triple Sugar Iron (TSI) agar test (Anon. 1991).

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### 2.4.5. Gram Staining and Culture Identification

Smears of growth from the plates were made on clean slides with sterile wire loop. These were Gram stained using Crystal violet, Gram's iodine, Acid alcohol and Saffranin in sequence, The slide was viewed under the microscope to identify the morphology and Gram reaction.

### 2.5. Sensory Evaluation

Sensory quality of momone was evaluated by 15 untrained panelists from the Food Research Institute who were familiar with the product. The panel members were also very familiar with the aroma and taste in terms of desirability of momone in nkontomire (cocoyam leaves) stew and palm soup.

The panelists were asked to evaluate nkontomire stew and, or palm soup which were flavoured with momone. A 9 - point hedonic scale was used to evaluate the following sensory attributes: aroma, taste and overall acceptability

A preliminary study on the samples brought from the local processing sites was conducted to provide a guide as to the quantity of momone needed to impart the desired aroma and taste of dishes

In the study, a standardized formulation with varied levels of momone was used in dishes preparation.

### 2.5. Fermentation Studies

Fresh king fish (Scomberomorus tritor) which had not been subjected to any cooling preservation process were obtained. The fish were gutted, washed and cut into 3 pieces. These were left overnight at room temperature in a fly free enclosure. The seemingly spoiled fish were washed the next day and weighed into approximately 3 equal weights. A processor was asked to prepare the three lots of fish for fermentation as she would do normally. Amount of salt added were recorded and it was observed that at the end of the salting period, 3 varying concentrations of salt in the different lots were obtained. These were 15, 25 and 40% of the respective wet weights of the fish lots.

The salted fish were placed in a plastic bucket per each lot and covered. These were left undisturbed for three days. On the third day, the fish were removed and washed. This was followed by resalting but in this case the salt quantities were reduced to about threequarters of the initial weights.

The fish were placed back into their respective containers and allowed to ferment for a further 10 days. Fish samples were pulled from the initial raw material. Further samples were taken after the fish were left overnight, after first salting, after resalting and thereafter, at two days interval as the fermentation process progressed. These samples were subsequently subjected to the following physio-chemical analyses: pH, TVB-N, TBA, protein, total chloride (salt), FFA and Aw measurements.

Microbiological analyses were also carried out. These were total plate count, indicator organism (enterobacteriaceae), pathogenic organisms and culture identification.

Sensory evaluation were carried out on the final products of the three treatments. Parameters evaluated included, appearance, taste and overall acceptability.

### 2.6. Storage Trails on Fermented Fish

Following the previous studies and results obtained on the salting levels, a quantity of kingfish were processed in to momone. The processing and preparation were carried by the steps adopted from

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studies preceding this. These steps are outlined below;

- 1. Fresh fish was cleaned, gutted and washed.
- Drained and left overnight at room temperature in a fly-free enclosure.
- 3. Removed and washed clean.
- 4. First salting with salt to fish weight ratio of 1:4
- 5. Covered and left for 3 days.
- 6. Removed and washed lightly.
- 7. Second salting with 3/4 of the salt weight used in the first salting.
- 8. Allowed fermentation to proceed for at least one week.

In evaluating an effective condition of keeping and presenting fermented fish, four storage systems were prepared. These were:

- (a) One or two pieces of momone were vacuum packed as control (NT).
- (b) Pieces dipped for a minute or two in 1% vinegar and vacuum sealed (ACl).
- (c) Pieces dipped for a minute or two in 3% vinegar and vacuum sealed (AC3).
- (b) Pieces dipped for a minute or two in 20% raw garlic extract and vacuum sealed (GS).

These conditions were necessary if one considers the following:

- That the product has a strong pungent smell which easily attracts flies and could be offensive.
- The product contains active bacteria and enzymes whose actions still persist.
- Excessive exposure of the product may lead to severe dehydration which could affect its properties.

The sealed products were left at ambient temperature and the following physio-chemical properties were monitored: pH, TVB-N, protein, total chloride (salt), FFA and TBA.

Initial microbiological analyses were also carried out. These were total plate count, indicator organism (enterobacteriaceae), pathogenic organisms and culture identification.

Sensory evaluation was carried out on the products being stored under the four conditions. Parameters evaluated include, appearance, aroma, taste and overall acceptability.

### 2.7. Sauce Preparation

The exudate from the fermenting fish were collected and concentrated to half the original volume by boiling. Sodium chloride (salt), protein, pH and microbial analysis were carried out before and after the boiling. Sensory evaluation was only carried out on the concentrated sauce. The parameter evaluated was the effectiveness of the flavour enhancing property of the sauce.

### 3. RESULTS AND DISCUSSIONS

### 3.1 Field Survey

Females were the sole processors involved in the industry. They were assisted by relations who did not receive income formally.

The processing sites (Table 3.1) were located close to the beach and activities of the processors were carried out rather late in the evenings or very early in the mornings. This was considered very desirable since the processors were harassed during daytime, as a result of the offensive smell coming from their activities.

It must be pointed out that all the processes at the sites were in the open and hence unprotected from houseflies which were found everywhere. The flies generally presented a great problem to the processors leading them to adopt various means to ward off the flies.

Table	3.1.	Processing	Sites	and	Processing	Characteristics

Site	Type of fish and state	Duration of fermentation
Tema New Town	) Refer	3 to 10 days depending on
Kpone	) below *	specie (Ave of 4 days)
Kokrobite	)	1
Nyanyaanor	)	

\* Types of specie used include: Surgeon-fish, cassava fish, Spanish mackerel, sea bream, tuna, parrot wrasse, squid, brotula blue shark etc.

Usual raw materials used are fresh fish which are left overnight for spoilage to set in. However, fish which has become stale as result of poor handling, storage etc. are ready raw materials for processing. Sharks and squids were usually not left overnight.

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Salting and fermentation were carried out immediately these species were obtained. Prior to salting the fish was eviscerated and left overnight, at which time a rather quite offensive and foul was produced. This step, was considered extremely important for the production of good, soft textured and well flavoured momone. This offensive smell necessitated that the processing be carried at the odd hours of the day.

Fermentation periods were varied and these depended on the processor and marketing conditions. Processors may sell their products as and when there was a market for the products, otherwise products were kept in fermentation vats.

A pictorial depiction of the typical scenes and facilities at the processing sites are shown in Figs. 1 - 6.

It was observed that the processing, required simple pieces of equipment throughout the processing stages as indicated in Table 3.2. below.

Processing stage	Equipment
Washing	Bowl
Gutting	Knife
Salting	Bowl
Packing	Concrete Vats
Fermentation	
Washing off salt Packaging for sale	Bowl Baskets

Table 3.2. Equipment Used

Fermentation tanks were observed to have been moulded out of cement and have rough surfaces (Figs 1-6) These surfaces when not cleaned properly of disinfected after each fermentation process may serve as sources of contamination to the next batch of fish since the

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Figure 1. Evisceration of Fish before being left overnight



Figure 2. Dry salting of fish prior to fermentation

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Figure 3. Concrete fermentation vat in basket mould



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Figure 4. Fermenting fish in the open

![](_page_25_Picture_0.jpeg)

Figure 5. Packaging fermented fish for the market

![](_page_25_Picture_2.jpeg)

Figure 6. A typical displaying scene for momone marketing

crevices in the surface may harbour microorganisms or even larvae.

### 3.1.1. Protection from insect infestation

Alum was liberally added to salt in resalting to kill maggots of the blowflies and for long term storage. Lime juice extracts were also applied. Processors claimed the lime improved the flavour of the product. Though the application of these substances may achieve the end results, their use could present health hazards since it is known that some processors illegally apply household insecticides to the products.

### 3.1.2. Salt and salt ratio

Rock salt was predominantly used and may be reused in other fermentation processes. The salt was normally stored in sacks and placed on the bare floor or the ground at the processing sites, hence exposed to all the elements of environmental hazards. The salt could therefore be a potential source of contamination of various microorganisms and especially halophilic bacteria.

Used salt were usually tainted yellowish probably from materials leached out from the fish. For long term storage some processors used less coarse salt but could not assign any reason for that.

Exudate after fermentation may be discarded but some processors reused it in further fermentation processes. This latter practice could lead to cross contamination in subsequent fermentation processes. Added to this, processors used the same sea water contained in a basin to repeatedly wash large quantities of gutted fish for fermentation.

Ratio of salt to fish varied and could not be quantified. It is possible that the cost of salt may not be of any economic significance yet, salt as the main preservative in this instance plays another important role in the safety of the product. Low salt fermentation could cause the outbreak of botulism which though could be destroyed by cooking, is very stable in salty and acidic environments (Huss and Rye Pederson 1980).

Processors traditionally, packaged fermented fish in baskets lined with brown paper, cement bags paper or old sacks when transporting the products to the markets. Apart from their very unhygienic nature, these materials could serve as potential sources of bacterial and other contaminants.

3.2. Chemical, Microbial and Sensory Evaluation of Samples

The chemical analysis of the two fermented fish samples bought at the processing sites are shown in Tables 3.3 and 3.4 below

Table 3.3. Chemical Analyses \* of Jack Mackerel (Caranx hippos)

3.2.1. Chemical analyses

Sample No.	Aw	рН	TVB-N (g/100g)	TBA (mg/kg)	FFA (g/100g	Protein ) (%)	Total Chloride (%)
1	0.73	7.6	305.9	7.08	26.9	31.4	4.1
2	0.65	7.5	326.9	9.06	19.7	26.3	5.7
3	0.71	7.4	318.1	8.10	18.6	25.4	4.3
4	0.59	7.4	.341.2	8.12	16.1	29.4	5.1
5	0.85	7.5	295.4	9.09	14.7	22.9	4.2

\* Values are means of duplicates for five sample lots

# Table 3.4. Chemical Analyses of Kingfish/Spanish mackerel (Scomberomorus tritor).

Sample N	0.	Aw	Η	TVB-N (g/100g)	TBA (mg/kg)	FFA (g/100g)	Protein ) (%)	Total Chloride (%)
1		0.76	7.7	357.0	9.12	20.9	23.9	4.6
2		0.77	7.8	387.8	6.09	31.6	21.7	4.4
3		0.59	7.9	385.0	8.17	27.2	29.2	6.1
4		0.87	7.8	394.8	9.11	39.9	21.2	3.9
5		0.60	7.7	277.5	10.13	21.7	27.9	6.3

\* Values are means of duplicates for two sample lots

### 3.2.1.1. Water Activity

The water activity of the fish species, kingfish (0.59-0.87) and jack mackerel (0.59-0.87) varied significantly within each sample pool as recorded in Tables 3.3 and 3.4. However these values though may be safe for the lower values, higher values of 0.87 and above are considered not safe since aerobic and other food poisoning bacteria action may not be hampered at such values (Owens and Mendoza 1985).

### 3.2.1.2. pH

Second Second

-10-14

Overall, the pH values as listed on the above Tables 3.3 and 3.4, for the two species (above pH of 7) were not significantly different from each other and may well be taken as the usual pH for fermented fish. Yankah (1988) observed pHs of above 7 at the end of 2 days of fermentation and inferred that since the fish must be in the deteriorated form before fermentation, the high pHs obtained were expected with such products. Van Veen (1965) considered a pH of 6.5 or higher as indicative of poor quality of a Thai fermented fish 'Pedah siam'. Comparative higher values of above 7 were also reported on fermented fish in Ghana by Nerquaye- Tetteh <u>et</u> al (1978).

### 3.2.1.3. TVB-N

Within the species the high values of the TVB's are expected for such products as well as the variations within sample lots. Nerquaye-Tetteh <u>et al</u> (1978), reported values of TVBN of 366 and above as compared to values shown on Tables 3.3 (295) and 3.4. (277.5). TVB measurements indicate the extent of the break-down of proteins leading to amines production and thus a low nutritional value of the product (Mackie et al. 1971).

### 3.2.1.4. Free Fatty Acids and TBA

Free fatty acids were recorded as very high for both species (Tables 3.3 and 3.4.) and were characteristic of products that had undergone some form of spoilage (Lassen et al. 1951).

TBA values of all samples showed high values as a reflection of spoilage activity with its attendent rancidity.

### 3.2.1.5. Crude Protein

Protein values (22-31%) recorded in the two Tables 3.3 and 3.4. are significantly low for a fish. Accordingly these values could be compared favourably as not different from values obtained by other workers (Nerguaye-Tetteh <u>et al.</u> 1978). However Yankah (1988) recorded a higher value of 57.1%. This could be attributed to probably the variations in fish species used and the short period of her fermentation process in which much proteolytic activity might not have occurred.

### 3.2.1.6. Total Chloride (Salt)

The apparent low salt values for the species and within the sample lots (4-6%) as compared to results of other workers 10-15% (Nerquaye-Tetteh <u>et al.</u> 1978), may not be ideal for storage purposes. This could well be described as under salting. However, the low water activity in the products could provide a safe barrier to the growth of some food poisoning bacteria (Owens and Mendoza 1985). Yet problems may arise as a result of some toxins being stable in low salt conditions when already present before the salting (Huss and Rye Pederson 1980).

### 3.2.2. Microbial enumeration of samples

Table 3.5. Microbiological <sup>\*</sup> Quality of Fermented Fish Species

Microbiological	Fish Spe	cie
Determination	Kingfish	Jack Mackerel
Total viable count (Aerobic)	5	ρ
PCA at 30 °C	3.3x10 <sup>5</sup>	2.1x10 <sup>0</sup>
PCA+15% NaCl at 30 <sup>O</sup> C	3.8×10 <sup>5</sup>	2.1x10 <sup>5</sup>
Culture	Micrococci	Micrococci Bacillus spp.
Coliforms (0.1 g)	Absent	Absent
E. coli Pathogens:	Absent	Absent
S. aureus (0.1 g)	Absent	Absent
V. parahaemolyticus in 25 g sample	Absent	Absent
Salmonella in 25 g sample	Absent	Absent
Sampre	Absent	Absent

\* Counts are means of five sample lots for each specie

Total counts on all samples were not high and within acceptable limits (Table 3.5). The predominant and only organisms identified were Micrococci and Bacillus sp. No <u>S. aureus</u>, Vibro organisms or Salmonella were isolated. No coliforms or <u>E. coli</u> was also isolated. This indicates that no faecal material has contaminated the fish during handling and processing. In a study on momone fermentation, Yankah (1988) observed that the predominant organisms were Bacilli and Micrococci. Overall, the two fish species, though traditionally prepared with their observed potential problems, posed no public health hazard.

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### 3.2.3 Sensory Evaluation of Samples

This was used as a preliminary assessment of the taste, aroma and overall acceptabilty of the samples in stews. The study showed that 30 g to 50 g of fermented fish was enough to influence the palatability of nkontomire stew.

### 3.3. Fermentation Studies

### 3.3.1. Physical Characteristics of Products

The following physical changes were observed on the fish lots from day one when the fish were left overnight. These changes are noted in Table 3.6.

Days	Salt	Concentrations-	-
	15% Salt	25% Salt	40% Salt
	-		
Ţ	Shiny colour Rotten smell	Shiny colour Rotten smell	Shiny colour Rotten smell
	Slimy	Slimy	Slimy
3	Dull colour	Less shiny	Less shiny
	Pungent smell	Less pungent	Less pungent
5	Brownnish	Shiny tinge	Shiny tinge
	Less pungent	Slight aroma	Slight aroma
	Watery	Slightly dry	Slightly dry
7	Musty Brown	Shiny tinge	Shiny tinge
	Slight aroma/	Strong aroma	Strong aroma
	Slightly wet	Soft texture	Soft texture
9	Intensely musty	Shine tinge	Shiny tinge
	brown.	Strong aroma	Strong aroma
	Aroma perceived but pungent	Very strong aroma	Very strong aroma.
	Slightly wet	Partially dry	Partially dry

### Table 3.6. Physical Changes in fermenting fish lots

\* % salt is (% w/w) of unfermented fish

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The physical changes in the three preparations were quite significant especially towards the end of the fermentation period. Fish with salt concentrations above 15% appeared to maintain their original state in terms of colour and were found to be comparatively dry. The colour changes were quite obvious for the 15% salt concentrations and these could have resulted from the growth of a bacteria of the Halobacterium group (Murray 1974), which is able to tolerate the 15% salt concentration but might not be able to do so in the higher concentrations.

The partial dryness of the fish from the higher salt concentrations of 25 and 40% could mean that more water was lost in these products as a result of the greater osmotic pressure the salt concentrations exerted than the 15% salt concentration.

### 3.3.2. Chemical characteristics of Samples

Changes in chemical characteristics of the fermenting fish lots were measured as depicted in the Tables below (Tables 3.7, 3.8 and 3.9).

	Day.	Aw	рН	TVB-N (g/100g)	TBA (mg/kg)	FFA (g/100g	Protein ) (%)	Total Chloride
	0		6.2	27.0	0 9	2 0	6 8 0	( % )
٦	(ouernight)	_	0.3	325 1	1.9	10.7	62 3	_
T	2	0 81	7.8	464 1	2 8	14.6	50 4	5 3
	5	0.01	7.8	422.2	2.8	16.1	42.4	5.1
	7	0.67	7.5	436.4	4.9	14.7	40.9	5.2
	9	0.62	7.3	438.2	6.7	16.5	30.8	5.8

# Table 3.7. Changes in chemical properties during fermentation of Kingfish at 15% salt concentration.

\* Values are means of duplicates for samples.

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						×		
	Day.	Aw	рН	TVB-N (g/100g)	TBA (mg/kg)	FFA (g/l00g)	Protein (%)	Total Chloride (%)
	0	-	6.3	27.9	0.8	3.9	69.9	
1	(overnight)	) —	7.8	325.1	1.9	10.7	62.3	
-	3	0.74	7.7	366.8	2.8	16.6	60.8.	5.3
	5	0.71	7.9	413.0	4.2	18.3	55.4	5.8
	7	0.67	7.6	404.6	8.6	17.9	53.9	9.2
	9	0.59	7.2	408.7	7.9	15.8	45.7	10.6

Table 3.8. Changes in chemical properties during fermentation of Kingfish at 25% salt concentration.

\* Values are means of duplicates for samples.

# Table 3.9. Changes in chemical properties during fermentation of Kingfish at 40% salt concentration.

	Day.	Aw	рН	TVB-N (g/100g)	TBA (mg/kg)	FFA (g/100g)	Protein (%)	Total Chloride (%)
	0	_	6.3	27.9	0.8	3.9	69.9	
1	(overnight)	-	7.8	325.1	1.9	10.7	62.3	
	3	0.72	7.8	330.6	5.1	18.6	61.8.	5.8
	5	0.67	7.6	381.0	5.4	18.5	57.4	7.9
	7	0.58	7.4	324.6	14.6	21.9	54.9	9.9
	9	0.56	7.4	343.7	10.2	15.8	43.6	15.7

\* Values are means of duplicates for samples.

Water activity of the fish samples were reduced as the fermentation progressed with the final level of 0.56 recorded for the 40% salt concentration (Table 3.9) and 0.59 for the 25% salt concentration (Table 3.8). However the final level for the 15% salt concentration

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was 0.62 (Table 3.7). Though this level was considered sufficiently low to check the growth of many microorganisms, the low salt level may not be enough to guarantee a long term storage (Huss and Rye Pederson 1980).

The changes in pH across the Tables 3.7, 3.8 and 3.9 were not significantly different from their initial readings. This was so because the samples were already in their advanced stage of spoilage before the commencement of the fermentation and hence pHs were already high. The activities of the organisms involved in the fermentation process appeared not have had much influence on the pH levels.

In comparing the changing pattern of the TVB of the salt concentrations, high values as expected, were observed throughout the fermentation period in all three preparations (Tables 3.7, 3.8 and 3.9).

The changing pattern of the FFA and TBA contents across the three preparations did not vary significantly within each treatment and almost constant values were attained after day one (Tables 3.7, 3.8 and 3.9).

Gradual decreases in protein content were recorded for all the three preparations as shown in Tables 3.7, 3.8 and 3.9. These decreases were considered significant from their initial values. The 15% salt concentration recorded the lowest amount of 30.8% from the initial value of 68.9% protein content. However the final protein content of the 25 and 40% salt concentrations were much higher than that of the 15% salt concentration. These values of 45.7% (25% salt concentration) and 43.6% (40% salt concentration) could mean that, protein degradation in these preparations was not so rapid as compared to that of the 15% salt concentration. The higher salt concentrations maintained in these two preparations may have had an effect on proteolytic activities within the 25 and 40% salt concentrations. Overall, these values were much higher

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than those determined for the samples bought from the processing sites and other values reported by Nerquaye-Tetteh <u>et al.</u> (1978). However they appeared to be consistent with the values found by Yankah (1988).

Final salt contents were significantly higher in the 25 and 40% salt concentrations and were consistent with the amount of salt added. Gradual increases in salt content were observed as the general trend for the 25 and 40% salt concentrations (Tables 3.7, 3.8 and 3.9). Whilst the 15% salt concentration achieved an almost constant amount of 5%, levels reached for 25 and 40% salt concentrations were 10.6 and 15.7% respectively.

### 3.3.3. Microbial enumeration of samples

It was generally observed that there were increased total counts on all samples after they were left overnight (Tables 3.10-3.12). However, whilst these counts appeared to decrease with the treatments having salt concentrations of 25 and 40%, the lot with the 15% salt concentration increased steadily. Counts of about 8.8  $\times 10^7$ organisms were observed in certain cases. The dominant microorganisms observed at the onset of the fermentation process were Gram positive halophilic aerobic micrococci and <u>Bacillus sp.</u>. Beddows (1985) also observed similar microbial culture. In their studies, Nerquaye-Teth <u>et al.</u> (1978) and Yankah (1988), observed a greater percentage of the microflora on momone to be micrococci. It was observed that the microbial degradation of the fish during the fermentation, aided in the formation of the flavour characteristics. This development coincided with increased number of Bacillus sp..

The breakdown of lipids in the fish by microorganisms during fermentation to yield fatty acids may also be responsible for flavour and aroma compounds imparted to the products, and these may serve as precursors for the slight browning reactions that occurred on the fermenting fish. Pathogenic microorganisms such as staphylococcus, salmonella and vibro were however absent from all

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the samples. The absence of coliforms or faecal coli was indicative of the fact that good handling and processing procedures were applied to eliminate contamination of the fish and hence the production of wholesome end products.

No moulds were also isolated especially since the intermediate moisture state of the fish would not support mould growth. Manifestation of mould growth is shown by discolouration on the fish and may result in the accumulation of harmful mycotoxins (Christensen and Kaufmann 1974).

Table 3.10. Changes in microbial properties during fermentation of Kingfish at 15% salt concentration.

Day	у ТV РСА а 30°С	at PCA +1 at 30 <sup>0</sup>	Coli- 5% forms C	E. coli	Staph in 0.lg	Sal- monella in 25g	Culture	Vibro
0	2.1x10 <sup>6</sup>	4.1x10 <sup>3</sup>	Found in 0.01g	Found in 0.lg	Absent	Absent	Gram+ve Cocci & Micro- cocci	Absent
la	8.8x10 <sup>7</sup>	1.5×10 <sup>4</sup>	Absent in 0.lg	Absent in o.lg	11	"	п.,	Absent
3	5.6x10 <sup>5</sup>	1.6x10 <sup>2</sup>	н		"	п		
5	1,5x10 <sup>5</sup>	5.2x10 <sup>3</sup>		н.,	п	п		н
7	2.0x10 <sup>7</sup>	3.0x10 <sup>6</sup>				п	с <b>П</b>	
9	1.1x10 <sup>7</sup>	8.9x10 <sup>6</sup>	п	н	н	п	н	н

\* Values are means of duplicates for samples.

l Total Viable Count

a Samples left overnight.

Day	Y TV PCA a 30 <sup>°</sup> C	c <sup>1</sup> t PCA +1 at 30 <sup>°</sup>	Coli- 5% forms C	E. coli	Staph in 0.lg	Sal- monella in 25g	Culture	Vibro
0	6.1x10 <sup>5</sup>	5.2x10 <sup>3</sup>	Absent in 0.01g	Absent in 0.lg	Absent	Absent	Gram+ve Cocci & Micro- cocci	Absent
l <sup>a</sup> .	6.1x10 <sup>5</sup>	6.0x10 <sup>1</sup>	Absent in 0.lg	Absent in o.lg	п			Absent
3	3.0x10 <sup>6</sup>	2.4x10 <sup>2</sup>	ц		п		п	u
5	6.0x10 <sup>3</sup>	7.0x10 <sup>4</sup>	н	п	ч н.		п	п
7	2.6x10 <sup>4</sup>	4.0x10 <sup>1</sup>	п	п		п	п	п
9	2.0x10 <sup>3</sup>	1.0x10 <sup>3</sup>			п			
Tał	* Value l Total ble 3.12.	s are mea Viable C Changes of King	ns of dup ount. a - * in micro fish at 40	licates for - Samples Dbial prop D% salt con	r sampl left ov erties ncentra	es. ernight during tion	fermenta	tion
Tab	* Value l Total ble 3.12. y TV PCA a 30°C	s are mea Viable C Changes of King C <sup>1</sup> t PCA +1 at 30 <sup>0</sup>	ns of dup ount. a fish at 40 Coli- 5% forms C	licates for - Samples for Obial prope O% salt con E. coli	r sampl left ov erties ncentra Staph in 0.lg	es. ernight during tion Sal- monella in 25g	fermentat Culture	vibro
Tab Day	* Value l Total ble 3.12. y TV PCA a 30 <sup>o</sup> C 1.2x10 <sup>5</sup>	s are mea Viable C Changes of King C <sup>1</sup> t PCA +1 at 30 <sup>0</sup> 4.5x10 <sup>3</sup>	ns of dup ount. a fish at 40 Coli- 5% forms C Absent in 0.01g	licates for - Samples for obial prope 0% salt con E. coli E. coli Absent in 0.1g	r sampl left ov erties ncentra Staph in 0.lg Absent	es. ernight during tion Sal- monella in 25g Absent	Gram+ve Cocci & Micro- cocci	Vibro Absen
Day 0	<pre>* Value 1 Total ole 3.12. y TV PCA a 30°C 1.2x10<sup>5</sup> 7.7x10<sup>5</sup></pre>	s are mea Viable C Changes of King c <sup>1</sup> t PCA +1 at 30 <sup>0</sup> 4.5x10 <sup>3</sup> 2.7x10 <sup>2</sup>	ns of dup ount. a fish micro fish at 40 Coli- 5% forms C Absent in 0.01g Absent in 0.1g	Licates for - Samples for obial prope 0% salt con E. coli Absent in 0.1g Absent in o.1g	r sampl left ov erties ncentra Staph in 0.lg Absent	es. ernight during tion Sal- monella in 25g Absent	Gram+ve Cocci & Micro- cocci	Vibro Absen Absen
Tak Day 0	<pre>* Value 1 Total ple 3.12. y TV PCA a 30°C 1.2x10<sup>5</sup> 7.7x10<sup>5</sup> 1.4x10<sup>5</sup></pre>	s are mea Viable C Changes of King c <sup>1</sup> t PCA +1 at 30 <sup>0</sup> 4.5x10 <sup>3</sup> 2.7x10 <sup>2</sup> 1.0x10 <sup>1</sup>	ns of dup ount. a fin micro fish at 40 Coli- 5% forms C Absent in 0.01g Absent in 0.1g	Licates for - Samples - - Samples - - Samples - 	r sampl left ov erties ncentra Staph in 0.lg Absent	es. ernight during tion Sal- monella in 25g Absent "	Gram+ve Cocci & Micro- cocci	Vibro Absen "
Tab Day 0	<pre>* Value 1 Total ole 3.12.  y TV PCA a 30°C 1.2x10<sup>5</sup> 1.2x10<sup>5</sup> 1.4x10<sup>5</sup> 1.5x10<sup>3</sup></pre>	s are mea Viable C Changes of King c <sup>1</sup> t PCA +1 at 30 <sup>0</sup> 4.5x10 <sup>3</sup> 4.5x10 <sup>3</sup> 2.7x10 <sup>2</sup> 1.0x10 <sup>1</sup> 1.0x10 <sup>2</sup>	ns of dup ount. a * in micro fish at 40 Coli- 5% forms C Absent in 0.01g Absent in 0.1g	Licates for - Samples - Samples	r sampl left ov erties ncentra Staph in 0.lg Absent " "	es. ernight during tion Sal- monella in 25g Absent "	fermentat Culture Gram+ve Cocci & Micro- cocci " "	Vibro Absen Absen
Tak Day 0 1 <sup>a</sup> 3 5 7	<pre>* Value 1 Total ple 3.12.  y TV PCA a 30°C 1.2x10<sup>5</sup> 1.2x10<sup>5</sup> 1.4x10<sup>5</sup> 1.5x10<sup>3</sup> 3.8x10<sup>4</sup></pre>	s are mea Viable C Changes of King c <sup>1</sup> t PCA +1 at 30 <sup>0</sup> 4.5x10 <sup>3</sup> 4.5x10 <sup>2</sup> 1.0x10 <sup>1</sup> 1.0x10 <sup>2</sup> 2.5x10 <sup>2</sup>	ns of dup ount. a - * in micro fish at 40 Coli- 5% forms C Absent in 0.01g Absent in 0.1g	Licates for - Samples - Samples	r sampl left ov erties ncentra Staph in 0.lg Absent " "	es. ernight during tion Sal- monella in 25g Absent " "	Culture Gram+ve Cocci & Micro- cocci " "	Vibro Absen "

Changes in 3 11 perties during fermentation Table h i

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### 3.3.4. Sensory Evaluation of Samples

The taste, aroma and overall acceptability of the samples in stews were evaluated. There were no significant differences between the three treatments as Tables 3.13 and 3.14 below show.

Table 3.13. Mean sensory score for momone prepared at varying levels of salt concentrations and used in Nkontomire stew

Treatments % salt added	Appearance	Smell	Taste	Overall Acceptability
15	8.13+0.57	7.66+0.61	7.26+0.79	7.46+0.63
25	8.06+0.59	7.13 <u>+</u> 1.18	6.93 <u>+</u> 1.09	7.26+1.16
40	8.00+0.53	7.20+0.77	7.13+0.83	7.13 <u>+</u> 0.63
Means for s	ensory scores	are for 15	panelists.	

Table 3.14. Mean sensory score for momone prepared at varying

levels of salt concentrations and used in Palm soup

				¥
Treatments % salt added	Appearance	Smell	Taste	Overall Acceptability
15	8.18+0.54	7.62+1.08	6.93+1.94	7.25+1.69
25	8.12 <u>+</u> 0.61	7.56+0.81	6.93 <u>+</u> 1.12	7.12+1.02
40	8.00+0.73	7.81+0.91	6.93 <u>+</u> 1.23	7.18+1.42
Moons for s	ancory scores	are for 15	nanolists	
means LUL S	ensury scores	are ror ro	punctioco.	

### 3.4. Storage studies

### 3.4.1. Physical characteristics of Products

There were no significant physical changes on the products at the start of the storage trial. However at the end of a month's study, all the products developed a slight brownish colour.

### 3.4.2. Chemical characteristics of Samples

Changes in chemical characteristics of the stored products as determined after a month's study are presented in Tables 3.15, 3.16, 3.17 and 3.18).

Water activity values across the Tables did not vary significantly and were to a large extent constant. The same could be said of the pH, TVB, FFA, Total Chloride and the TBA (Tables 3.16, 3.17 and 3.18). However decreases were noted for all the protein contents within the treatments. The greatest decrease was observed for the untreated vacuum sealed product (control), from 40.9 to 29.8% (Table 3.16) within the storage period. Though there were decreases in the other treatments, these were much lower than that of the untreated sample. The 20% garlic extract treatment, showed a decrease of 10%, from that of 52% to 42% (Table 3.18). This was a little more than those of the vinegar treated products, with the 3% vinegar treatment showing the least, from 54 to 49% (Table 3.17).

Though the storage period was limited, the vacuum packaging and treatments could have had some effects on the values of the properties determined, especially for the protein contents.

Vacuum packaging was chosen to minimise oxidative reactions as the product contained highly oxidisable compounds from its lipids (Ames and Poulter 1987). Though the vacuum was also intended to suppress aerobic microbiological activity, the problem of other bacteria namely, histamine formers, <u>V. parahaemolyticus</u> and <u>C.</u>

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botulinun type E, could not be overlooked. These could occur naturally on fresh seafoods and pose a health hazard during storage (Taylor 1985). The packaging could be an attractive tool to get the product onto market shelves without any fuss.

The use of the vinegar was to create an acidic condition which would lower the pH of the product. Low pHs are known to prevent the growth of pathogenic bacteria, especially <u>Clostridium botulinum</u> (Huss and Rye Pedersen 1980).

Garlic has a strong antimicrobial activity. and this property could possibly inhibit pathogenic and active microorganisms during storage of the product (Kato 1973).

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Day.	Aw	рН	TVB-N (g/100g)	TBA (mg/kg)	FFA (g/100g)	Protein (%)	Total Chloride (%)
1	0.63	7.5	436.4	8.9	14.7	40.9	9.2
28	0.58	7.3	451.2	12.9	16.1	29.8	8.9

Table 3.15. Changes tin chemical properties of vacuum sealed momone (control) during storage.

\* Values are means of duplicates for samples.

Table 3.16. Changes in chemical properties of vacuum sealed momone treated with 1% vinegar during storage.

Day	Aw	рН	TVB-N (g/100g)	TBA (mg/kg)	FFA (g/100g)	Protein (%)	Total Chloride (%)
1	0.66	6.2	404.6	8.6	17.9	48.9	9.2
28	0.65	6.9	412.3	9.3	18.4	40.6	8.9

\* Values are means of duplicates for samples.

Table 3.17. Changes \* in chemical properties of momone treated with 3% vinegar during storage.

						9	
Day.	Aw	рН	TVB-N (g/100g)	TBA (mg∕kg)	FFA (g/100g	Protein ) (%)	Total Chloride (%)
l	0.54	6.0	324.6	10.6	21.9	54.9	9.9
28	0.57	6.9	331.2	11,1	19.9	47.3	9.7

\* Values are means of duplicates for samples.

Day.	Aw	рН	TVB-N	TBA	FFA	Protein	Total
			(g/100g)	( mg / kg )	(9/100	g) (8)	(%)
1	0.63	7.2	344.6	11.6	26.2	52.3	8.4
28	0.65	7.0	349.3	12.7	24.8	42.9	8.5

Table 3.18. Changes in chemical properties of momone treated with 20% garlic extract during storage.

\* Values are means of duplicates for samples.

### 3.4.3. Microbial enumeration of samples

The effect of the treatments on the culture and microbial quality of the products are presented in Tables 3.19-3.22

Table 3.19. Changes in microbial properties during storage of untreated vacuum sealed momone (control)

Day	PCA a 30°C	c <sup>1</sup> t PCA +1 at 30 <sup>°</sup>	Coli- 5% forms C	E. coli	Staph in 0.lg	Sal- monella in 25g	Culture	Vibro
1.	4.5×10 <sup>4</sup>	1.3×10 <sup>3</sup>	Absent in 0.01g	Absent in 0.lg	Absent	Absent	Gram+ve Cocci & Micro- cocci	Absent
28	4.8x10 <sup>5</sup>	1.6x10 <sup>4</sup>	Absent in 0.lg	Absent in o.lg		п	н	Absent

\* Values are means of duplicates for samples. 1 Total Viable Count.

Table 3.20. Changes tin microbial properties during storage of vacuum sealed momone treated with 1% vinegar .

No. of Street, Street,

<pre>1. 4.0x10<sup>2</sup> 1.0x10<sup>2</sup> Absent Absent in Absent Absent Gram+ve Absent in 0.01g 0.1g Cocci &amp; Micro- cocci 28 3.2x10<sup>2</sup> 9.5x10<sup>1</sup> Absent Absent " " Absent in 0.1g in o.1g * Values are means of duplicates for samples. 1 Total Viable Count. Table 3.21. Changes* in microbial properties during storage of vacuum sealed momone treated with 3% vinegar . Day TVC<sup>1</sup> Coli- E. coli Staph Sal- Culture Vibro in monella 30<sup>o</sup>C at 30<sup>o</sup>C 0.1g in 25g 1. 3.6x10<sup>2</sup> 7.1x10<sup>1</sup> Absent Absent in Absent Absent Gram+ve Absent in 0.01g 0.1g Cocci &amp; Micro- cocci 28 2.9x10<sup>2</sup> 6.7x10<sup>1</sup> Absent Absent " " Absent in 0.1g in 0.1g in 0.1g</pre>	Day	TVC PCA at 30 <sup>°</sup> C	c <sup>1</sup> t PCA +1 at 30°	Coli- 5% forms C	E. coli	Staph in 0.lg	Sål- monella in 25g	Culture	Vibro
<pre>28 3.2x10<sup>2</sup> 9.5x10<sup>1</sup> Absent Absent " " Absent in 0.1g in o.1g * Values are means of duplicates for samples. 1 Total Viable Count. Table 3.21. Changes<sup>*</sup> in microbial properties during storage of vacuum sealed momone treated with 3% vinegar. Day TVC<sup>1</sup> Coli- E. coli Staph Sal- Culture Vibro pcA at PCA +15% forms in monella 30°C at 30°C 0.1g in 25g 1. 3.6x10<sup>2</sup> 7.1x10<sup>1</sup> Absent Absent in Absent Absent Gram+ve Absent in 0.01g 0.1g Cocci &amp; Micro- cocci</pre>	1.4.	0x10 <sup>2</sup>	1.0x10 <sup>2</sup>	Absent in 0.01g	Absent in 0.lg	Absent	Absent	Gram+ve Cocci & Micro- cocci	Absent
<pre>* Values are means of duplicates for samples. 1 Total Viable Count. Table 3.21. Changes in microbial properties during storage of vacuum sealed momone treated with 3% vinegar . Day TVC<sup>1</sup> Coli- E. coli Staph Sal- Culture Vibro PCA at PCA +15% forms in monella 30°C at 30°C 0.1g in 25g 1. 3.6x10<sup>2</sup> 7.1x10<sup>1</sup> Absent Absent in Absent Absent Gram+ve Absent in 0.01g 0.1g Cocci &amp; Micro- cocci 28 2.9x10<sup>2</sup> 6.7x10<sup>1</sup> Absent Absent " " Absent in 0.1g in 0.1g</pre>	28 3.	2x10 <sup>2</sup>	9.5x10 <sup>1</sup>	Absent in 0.lg	Absent in o.lg		"	"	Absent
Day $TVC^{\perp}$ Coli- E. coli Staph Sal- Culture Vibri PCA at PCA +15% forms in monella $30^{\circ}C$ at $30^{\circ}C$ 0.1g in 25g 1. $3.6 \times 10^{2}$ 7.1 $\times 10^{1}$ Absent Absent in Absent in Absent Gram+ve Absent in 0.01g 0.1g Cocci & Micro- cocci 28 2.9 $\times 10^{2}$ 6.7 $\times 10^{1}$ Absent Absent The sent " " Absent " " Absent In Absent " " Absent In Absent " " Absent In A	Table	e 3.21.	Changes vacuum	* in micro sealed mor	obial prop none treat	erties ed with	during n 3% vin	storage ( egar .	of
PCA at PCA +15% forms $30^{\circ}C$ at $30^{\circ}C$ 1. $3.6 \times 10^{2}$ 7.1 $\times 10^{1}$ Absent Absent in Absent in Absent Gram+ve Absent in 0.01g 0.1g 28 2.9 $\times 10^{2}$ 6.7 $\times 10^{1}$ Absent Absent The order of the order	Day			Coli-	E coli	Staph	Sal-	Culture	Vibro
1. $3.6 \times 10^2$ 7.1 $\times 10^1$ Absent Absent in Absent Absent Gram+ve Absent in 0.01g 0.1g Cocci & Micro- cocci 28 2.9 $\times 10^2$ 6.7 $\times 10^1$ Absent Absent " " Absent " Absent		PCA at 30°C	t PCA +1 at 30	5% forms C		in 0.lg	monella in 25g	I	
$28 2.9 \times 10^2 6.7 \times 10^1$ Absent Absent " " Absent " Absent	1.3.	.6x10 <sup>2</sup>	7.1x10 <sup>1</sup>	Absent in 0.01g	Absent in 0.lg	Absent	t Absent	Gram+ve Cocci & Micro- cocci	Absent
1	28 2.	9x10 <sup>2</sup>	6.7x10 <sup>1</sup>	Absent in 0.1g	Absent in o.lg		п		Absent

Table 3.22. Changes in microbial properties during storage of vacuum sealed momone treated with 20% garlic extract

Day	TVC PCA at 30°C	1 PCA +15 at 30°C	Coli- 5% forms C	E. coli	Staph in 0.lg	Sal- monella in 25g	Culture	Vibro
1.4	.6x10 <sup>2</sup>	9.3x10 <sup>2</sup>	Absent in 0.01g	Absent in 0.1g	Absent	Absent	Gram+ve Cocci & Micro- cocci	Absent
28 2	.4x10 <sup>1</sup>	4.1x10 <sup>1</sup>	Absent in 0.lg	Absent in o.lg				Absent

\* Values are means of duplicates for samples. 1 Total Viable Count.

Products of momone that were only vacuum sealed were found to be comparatively higher in microbial load than the treated products from either the vinegar or the garlic dips. However these counts (4.5x10<sup>4</sup> in PCA and 1.34x10<sup>3</sup> in PCA + 15% NaCl) were within microbiologically acceptable limits for the fermented fish. Counts for the garlic treated products (4.6x10<sup>2</sup> in PCA and 9.3x10<sup>2</sup> in PCA + 15% NaCl) were higher than those of the vinegar treated products which had the lowest. The counts for the products of the 1% vinegar solution were 4x10<sup>2</sup> in PCA and 1x10<sup>2</sup> in PCA + 15% NaCl and those for the 3% Vinegar solution were 3.6x10<sup>2</sup> in PCA and 7x10<sup>1</sup> in PCA + 15% NaCl. The predominant organisms were micrococci and <u>Bacillus</u> \_SP.

Microbial count of fermented fish stored for 1 month showed a slight increase for the untreated product (control) (Table 3.19). The slight decrease in bacterial count for the products treated with the garlic extract may be due to the antimicrobial properties of the garlic. Antimicrobial effect of garlic on some putrefactive microorganisms such as <u>Bacillus</u> in lactic acid fermented fish fish products had been observed by Soane <u>et al.</u> (1987). Dominant microorganisms observed at the onset of the fermentation process were gram positive halophilic aerobic Micrococci and <u>Bacillus sp.</u> Beddows (1985) also observed similar microbial counts. <u>Bacillus</u> sp. were however observed in smaller numbers especially in the

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vinegar-treated products. In the 1% (Table 3.21) and 3% (Table 3.22) vinegar treated samples, it was observed that there was no significant difference in counts between the two vacuum packed products. There was also no significant reduction in the numbers and types of microorganisms observed after 1 month's storage. In addition, no food poisoning organisms that may pose any public health hazards were isolated in any of the products.

### 3.4.4 Sensory Evaluation of Samples

The taste, aroma and overall acceptabilty of the samples in stews were evaluated after the treatments and after 1 month storge.

Table	3.23.	Mean	sensory	score	for	vacuum	sealed	momone
		used	in stew o	luring	stor	cage.		

Treatment	s <sup>#</sup> <u>Tast</u> 1 day	<u>e</u> 28 days	Aroma l day	28 days
NT	7.43	8.00	7.14	8.00
ACl	7.14	7.40	7.35	7.40
AC3	7.30	7.30	7.29	6.60
GS	6.64	6.80	6.50	7.30

\* Means for sensory scores are for 15 panelists. # NT = Untreated product (control). ACl = Product dipped in 1% vinegar solution. AC3 = Product dipped in 3% vinegar solution. GS = Product dipped in 20% garlic extract.

After one month of storage, the taste and aroma of the products had not changed significantly as shown in Table 3.23. The four products were observed to have significant influence on the palatability of the stews prepared as Table 3.23 shows.

### 3.5 Fish sauce from liquid after fermention

### 3.5.1 Chemical Analysis of Exudate

The results of the chemical analysis carried out on the exudate are presented in Table 3.24.

Exudate	рН	Protein (%)	Total Chloride (%)
Untreated	6.65	9.87	9.01
50% Con- centrated	6.06	12.76	23.50

## Table 3.24. Chemical properties of exudate after fermentation of momone

\* Values are means of duplicates for samples.

The pH of the exudate did not vary significantly after the heat concentration. At a pH of 6.06 (Table 3.24) the concentrated exudate could be said to compare favourably with other sauces produced in South East Asia which should have a maximum allowable pH of 7.5 (Tyn 1989). Though there are no limits for protein, the protein content of 12.76% of the concentrated exudate (Table 3.24) was quite adequate nutritionally, since it would be used mainly as a condiment. The salt content of 23.50% was rather below the recommended level of 25% (Tyn 1989) but this could be augmented by adding more salt to ensure safety of the product during storage.

### 3.5.2. Microbial properties of Exudate

The microbial properties of the exudate are presented in Table 3.25.

		~				
Table 3.25.	. Microbial	properties	of	exudate	after	fermentation

Exudate	PCA <sup>1</sup>	Coli- form in 0.lg	Staph in 5g	Sal- monella in 25g	Culture
Untreated	2.70x10 <sup>3</sup>	Absent	Absent	Absent	Gram+ve Cocci & Micro- cocci
50% Con- centrated	1.4×10 <sup>2</sup>	Absent	Absent	н	п

\* Values are means of duplicates for samples.

l Plate Count Agar.

The total microbial count of the exudate after concentration, of  $1.4 \times 10^2$  was much lower than the minimum of one million per gram of sample, suggested by Tynn (1989) for some Asian sauces. The exudate contained no food spoilage bacteria or food intoxication microorganisms as no coliforms, staphlococci nor <u>Salmonella</u> organisms were isolated (Table 3.25).

### 3.5.3. Sensory evaluations

The concentrated exudate was added to stews in place of momone to determine its flavouring efficacy. The preliminary results indicated that the flavour of the stews was enhanced to the same extent as when momone was added. However, the resultant stews were a little bit salty in taste. This could be overcome by using just enough of the exudate and avoid adding any salt until the exudate had been added to the stew. The strong flavour of the exudate made it unsuitable to be used at the dining table like other sauces, hence it is recommended that the exudate should be used only during cooking.

### 4. CONCLUSIONS AND RECOMMENDATIONS

The traditional processing of momone was found to be rather simple in nature. However, the practices and the state of the equipment employed in the process were sources of potential problems which could lead to serious health hazards. The use of concrete vats with crevices on the inner surfaces and the unhygienic handling of salt, water and packaging materials were considered as potential sources of contamination which could have effect on the safety of the fermented products inspite of the fact that sample products analysed were found to be microbiologically safe. The application of unapproved chemicals such as household insecticides, to check the menace of blowflies could be dangerous.

Fermentation trials with the inclusion of a second salting step, yielded products which were better in terms of protein content and microbial quality. Protein contents of 30 - 43% were obtained for the trial products as compared to 20 -25% of the products from the traditional processing site. Twenty-five percent and over, of salt to fish weight ratio appeared to offer the best conditions for the studies in terms of product quality characteristics and economy of salt usage as compared to the other two salt to fish weight ratios.

Vacuum sealing of momone treated with antimicrobial agents such as garlic and vinegar reduced the microbial load of stored momone and with a comparatively less losses of protein content than a vacuum sealed momone without any treatment during one month storage period. These treated products were not affected organoleptically.

The exudate after fermentation contained protein content of about 10% and when concentrated by boiling, this could serve as a condiment in food preparation.

Subsequent to the studies, it is suggested that processors need to be educated on hygienic procedures, and if possible processing

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sites should be enclosed or screened against flies. In salting, adequate amounts of salt should be applied. The salt used should amount to 25% or over, of the weight of fish to be fermented. Products could be displayed under mosquito netting screens during marketing so as to prevent the menace of flies. The use of chemicals should be discouraged and it is further recommended that approved insectides should be identified and their efficacy, practical application and affordability be evaluated before being adopted by processors.

The prospects of using biological means such as spices and food acids together with vacuum packaging appear quite attractive and need more extensive investigation. This could add a lot of prestige and value to momone.

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### 6. APPENDIX

### QUESTIONNAIRE ON FERMENTED FISH PROCESSING

1. Do you normally ferment fish?

- 2. Do you have other people to assist you?
- 3. Do you give them anything for their help?
- 4. Is there any particular reason why you ferment fish?
- 5. What types of fish do you normally ferment?
- 6. Why would you not ferment other species?
- 7. How do you start the fermentation?
- 8. Do you have to prepare the fish in a special way?
- 9. Why would you use a particular equipment?
- 10. Do you have any idea as to the quantity of salt used?
- 11. Why would you use a particular type of salt?
- 12. How long does the fermentation process last?
- 13. Does it vary with the type of fish used, state of fish, amount of salt and particular period of the year?
- 14. What signifies the end of the fermentation?
- 15. What significant characteristics are associated with the product?
- 16. What happens when the usual procedure is altered?
- 17. Do you use or throw away the liquid that remains after the product is removed?
- 18. During processing, how do try to keep flies, dogs etc. from attacking the fish.
- 19. How do you prevent moulds and insects from attacking the stored product.
- 20. Do you retail or sell the product in bulk?
- 21. Do you have to transport it the market?
- 22. How much fish do you normally ferment?
- 23. What happens to unsold products?
- 24. How do you notice that the product could no longer be offered for sale?
- 25. Do you know whether people show preference for products from a particular kind of fish?
- 26. Do you know of any reason why some people may not use your product?

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27. Do you face any problem in your operations?

- 28. Do you think there is the need for any improvement in processing techniques, equipment and product quality?
- 29. What are some of the improvements you envisage in these operations?
- 30. Would you like the product to be sold in supermarkets or exported?

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