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RESEARCH CORPORATION OF THAILAND

COOPERATIVE RESEARCH PROGRAMME NO. 31

IMPROVEMENTS IN THE PRODUCTION OF FISH SAUCE (NAMPLA)

RESEARCH PROJECT NO. 31/4

ACCELERATION OF NAMPLA FERMENTATION BY A BIOLOGICAL QUICK PROCESS

REPORT NO. 2

EFFECT OF SALT CONCENTRATION AND SIZE OF FISH ON COURSE OF FERMENTATION AND PRODUCT QUALITY OF NAMPLA

BY

SMAN VARDHANABHUTI

JIRAPORN CHOUVALIT

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ASRCT, BANGKOK 1970

not for publication

DEPARTMENT OF SCIENCE, MINISTRY OF INDUSTRY
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CONTENTS

F O R E W O R D

Because of new developments, the Cooperative Research Programme No. 31 (Improvements in the production of fish sauce (nampla)) undertaken jointly by the Department of Science, Ministry of Industry, the Department of Fisheries, Ministry of Agriculture, and the Applied Scientific Research Corporation of Thailand, has been revised and re-issued. Under the revised programme, the name of Research Project 31/4 was changed to "Acceleration of nampla fermentation by a biological quick process". The present report is the continuation of the work accomplished by ASRCT and covers attempt to define more precisely the working conditions necessary for scale-up operation and later industrial application. It appears that the new biological quick process can be applied to various fish species, both small and large and both fresh and iced. Methods have been worked out to deal appropriately with each particular fish species. Scale-up work is now to be undertaken to find out whether the process will work as well on a large scale and to evaluate the economics of industrial production.

EFFECT OF SALT CONCENTRATION AND SIZE OF FISH ON COURSE OF FERMENTATION AND PRODUCT QUALITY OF NAMPLA

By Sman Vardhanabhuti,* Jiraporn Chouvalit,*
Yuk-Hang Sombatpanit,+ and Pravat Lauhasiri+

SUMMARY

Salt concentration was found to affect the course of fermentation and the aroma of the primary extract of nampla. Concentration lower than 20 per cent by weight of the fish caused more rapid disintegration and breakdown of nitrogenous materials, but it also allowed too much putrefaction with consequent higher ammonia nitrogen content and bad aroma of the product. Good quality primary extracts were obtained with 20, 25, and 30 per cent salt, but the higher the salt concentration, the less the breakdown of nitrogenous materials, and the better the aroma. These were true for both large and small fishes.

Adding more salt to the mash initially fermented with 15 per cent salt for one month in case of pla thu did not improve the aroma of the product and resulted in reduction of nitrogenous compounds in the primary extract. For the latter effect, raw salt was worse than sodium chloride.

Size of fish also affected the course of fermentation. For pla kratak it was found sufficient to carry out the fermentation at 37°C for one month followed by sun-exposure for another month. Accelerating the initial disintegration of fish by conducting the fermentation at 49°C for two days first, and then continuing the rest of the fermentation at 37°C for two months was not significantly more advantageous.

For the larger-size fishes (pla thu and pla sai daeng), it was found necessary to conduct the initial fermentation at 49°C until the fish was fairly well broken up, and then

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continue the rest of the fermentation at 37°C for at least two months (or three months in the case of pla thu). Fine chopping or machine grinding could substitute for the initial incubation at 49°C when the fermentation was carried out with 25 per cent salt. In the case of pla thu, better products were obtained with the latter treatment.

For the economics of commercial production, fermentation with 30 per cent salt is recommended since one more acceptable leaching (third) could be made.

INTRODUCTION

It was found in earlier work* that nampla fermentation could be accelerated by maintaining the fermentation at elevated temperatures ranging from 37°C to 49°C, provided the fermenting material contained sufficiently high salt concentration. With this new method good quality primary extract could be obtained with pla kratak (Stolephorus sp.) in about two months. There was good nitrogen retention and very little ammonia in the product. It appeared that good aroma was temperature dependent and different kinds of good aroma could be produced by changing the fermentation temperature. In this preliminary work only one species of fish and two salt concentrations (15 and 30 per cent by weight of fish) were tried. Good quality primary extracts were obtained only from the products made with 30 per cent salt, fermented either at 37°C for one month, or at 49°C for two nights and then at 37°C for two more weeks, or at 49°C for two nights and then at room temperature (27-35°C) for two more weeks. Each of these preparations was then exposed to the sun for another month. The yields of the primary extracts were approximately the same, i.e. about 600 millilitres for 1.5 kilogrammes of fish used.

In the various works being reported in this paper, we intended to study more closely the effect of salt concentration and size of fish on

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* "Acceleration of nampla fermentation by a biological quick process" by Sman Vardhanabhuti, Jiraporn Chouvalit, Yuk-Hang Sombatpanit, and Pravat Lauhasiri. Report No. 1 on Research Project 31/4 (Acceleration of nampla fermentation by artificial means). ASRCT unpublished report.

the course of fermentation and the product quality. The fermentation at 37°C was increased to two months in the case of pla kratak and sun exposure was omitted to see whether we could achieve a better yield or improve further the quality of the primary extract. Leaching was also tried to determine how this could be best accomplished, since the main profit of conventional nampla production comes from the leachings rather than from the primary extract.

MATERIALS AND METHODS

Pla kratak was obtained from Khlong Dan as previously. During the transport from the sea, sometimes the fish was iced and sometimes not.

Pla thu (Rastrelliger neglectus) and pla sai daeng (Nemipterus hexodon) were obtained from the Fish Marketing Organization of the Department of Fisheries, Ministry of Agriculture, at the landing place on New Road. They were already partially iced during the transport from the sea. No addition of ice was made during the transport to the laboratory.

The fishes were processed as soon as they arrived at the laboratory.

Pla kratak was processed with 4 different raw sea salt concentrations, namely 15, 20, 25, and 30 per cent by weight of the fish. Four kilogrammes of fish were used in each experiment. The fish with salt, after mixing by hand, was left to deslime at room temperature (30-35°C) overnight. The fluid from desliming was collected, boiled for five minutes, and then, together with the salt and fish mixture, was placed in a 5-litre glass beaker to be fermented in an incubator at 37°C for one month. After this the beaker, well-covered to keep out rain and flies, was left in the sun for another month.

To ascertain whether optimal enzymatic breakdown at 49°C (Croston 1960) and longer incubation at 37°C would further increase the yield or improve the quality of the product, pla kratak preparations containing 20, 25, and 30 per cent salt were also incubated at 49°C for two days, and then kept at 37°C for two months.

Because of the larger size of pla thu, two preparations, each with four kilogrammes of fish containing 15 and 30 per cent salt respectively, were left to deslime overnight, and then coarsely chopped into pieces about 1 to $1\frac{1}{2}$ inches square. The chopped fish with salt and the boiled desliming fluid were placed in a 5-litre glass beaker and incubated at 37°C for three months. It was found that pla thu did not disintegrate well at this temperature. There was only 60 per cent disintegration with the preparation containing 15 per cent salt and only 15 per cent disintegration with the one containing 30 per cent salt at the end of the period.

With this experience, pla thu preparations containing 15, 20, 25, and 30 per cent salt were similarly processed but incubated at 49°C until the fish was well disintegrated (one-two days). They were then kept at 37°C for two months.

Pla thu processed with 25 per cent salt, but more finely chopped after desliming (into pieces 0.5-1 cm square) was kept at 37°C for two months.

Another pla thu preparation, also with 25 per cent salt, after desliming was ground in an electric coconut grating machine, made locally. The ground material, together with the boiled desliming fluid, was kept at 37°C for two months.

The last lot of pla thu (4 kg) processed with 15 per cent salt, coarsely chopped after desliming, and with the boiled desliming fluid added back, was kept at 49°C overnight. It was then kept at 37°C for one month. At the end of the period, an aliquot was removed for routine analyses and the rest divided into five equal portions (about 750 ml each), each put into a 1-litre glass beaker, and subjected to the following treatments:

(1) Exposed to the sun for another month.

(2) Pure sodium chloride added to make 15 per cent (wt./vol.) and exposed to the sun for one more month.

(3) Same as (2), but kept at 37°C for another month instead of sun exposure.

(4) Raw sea salt added to make 15 per cent (wt./vol.) more of the

salt and exposed to the sun for one more month.

(5) Same as (4), but kept at 37°C for another month instead of sun exposure.

For pla sai daeng, only preparations with 20 and 25 per cent salt were studied, each with four kilogrammes of fish.

In one preparation, whole fish was used with 25 per cent salt. The fish was similarly treated as pla thu initially. The fish and salt mixture with the boiled desliming liquid added was incubated at 49°C until the fish was well disintegrated. This took four days. The preparation was then kept at 37°C for two months.

Four experiments were made with pla sai daeng finely chopped after desliming. Two of these were made with 25 per cent salt. One was subjected to 49°C incubation for four days and then kept at 37°C for two months. The other was kept at 37°C for two months. The other two experiments were carried out with 20 per cent salt and subjected to the same methods of incubation as the ones with 25 per cent salt.

In all the experiments with the various species of fish, the fermenting mash in the incubator was stirred vigorously once daily except during week-ends and holidays. During the 49°C incubation, the fish was broken up as much as possible with the use of glass stirring rod, while during the 37°C incubation, stirring helped to break up the layer of oil which formed on the surface and to drive out the evolving gas. Tap water was added whenever necessary to replace the amount lost through evaporation.

At the end of the first and second months of fermentation, a small aliquot was taken out to be examined for the degree of liquefaction, and for the pH and specific gravity of the filtrate. The filtrate was also analysed for the contents of total solid, sodium chloride, total nitrogen, ammonia nitrogen, organic nitrogen, formaldehyde nitrogen, and amino acid nitrogen. These procedures were described in the earlier report. The colour and aroma of the primary extract were also recorded.

At the end of the fermentation period, each preparation was filtered, the primary extract collected, and the residue leached.

Leaching studies were first carried out with residues from pla

kratak fermentations, originally fermented with 30 per cent salt and kept either at 49°C for two days, then at 37°C for two weeks followed by sun exposure for one month; or at 49°C for two days, then at room temperature for two weeks followed by sun exposure for one month; or at 49°C for two weeks followed by sun exposure for one month. In each case the leaching was made with 30 per cent (wt./vol.) salt solution in tap water, up to the original volume. After thorough mixing, it was left in the sun for two months. At the end of the first and second months an aliquot was filtered and the filtrate analysed. The colour and aroma of the leachings were also noted.

It was found that leaching in the sun longer than one month caused significant drops of total nitrogen, organic nitrogen and formaldehyde nitrogen in the leaching filtrates of most cases. Amino nitrogen was not much affected, however. Later leaching studies were therefore carried out with salt solutions of similar concentrations to those used in the original formentations, but leaching time was limited to one month only for each leaching. These included leachings of residues from pla kratak, pla thu, and pla sai daeng fermentations. Three successive leachings were made from each residue. Pla kratak leachings were kept in the sun, while pla thu and pla sai daeng leachings were kept in the incubator at 37°C.

Four samples of residues from pla thu fermentations after the third leachings were analysed for per cent of sodium chloride and per cent of total nitrogen. They were residues from preparations made with 20, 25, and 30 per cent salt originally, coarsely chopped, kept at 49°C for two days and 37°C for two months, as well as one preparation made with 30 per cent salt, kept at 49°C for two days and 37°C for three months.

Five hundred grammes each of fresh pla kratak, pla thu, and pla sai daeng were homogenized in a Waring blender, and aliquots of the homogenized materials were analysed for moisture, total solids, total nitrogen, and fat by the methods described in the AOAC manual (Horwitz 1965).

RESULTS

The results of pla kratak fermentations using different salt concentrations incubated at 37°C for one month and then exposed to the sun for another month are given in Table 1.

Table 2 records the results of pla kratak fermentations initially incubated at 49°C for two days and then kept at 37°C for two months.

The results of pla thu fermentations with different salt concentrations but similarly incubated at 49°C for one to two days, then kept at 37°C for two months are given in Table 3, which also includes the results of nampla fermentations using finely chopped and machine-ground pla thu with 25 per cent salt and incubated at 37°C for two months.

Table 4 records the results of pla sai daeng fermentations using whole fish with 25 per cent salt incubated at 49°C for four days and then kept at 37°C for two months, as well as the results of the fermentations using finely chopped fish with 20 and 25 per cent salt similarly incubated and also incubated at 37°C only.

The results of chemical analysis of fresh raw fishes are given in Table 5.

The results of pla thu fermentations using 15 per cent salt initially but more salt added at the end of the first month are given in Table 5.

Tables 7 and 8 record the results of leaching studies.

The results of chemical analysis of fish residues from pla thu fermentations are given in Table 9.

TABLE 1

RESULTS OF PLA KRATAK FERMENTATIONS WITH DIFFERENT SALT CONCENTRATIONS BUT SIMILARLY INCUBATED AT 37°C FOR ONE MONTH FOLLOWED BY SUN EXPOSURE FOR ANOTHER MONTH

Salt concentration	pH	Specific gravity	Total solids (g-%)	Sodium chloride (g-%)	Total N (g-%)	Organic N (g-%)	Formal-dehyde N (g-%)	Amino acid N (g-%)	Ammonia N (g-%)	Liq. portion from 150 ml (ml)	Aroma	Colour
15% salt												
After 1 month	6.6	1.1456	27.15	14.05	2.08	1.65	1.30	0.87	0.43	70	Fair	Yellowish brown
After 2 months	7.3	1.1451	27.66	14.08	2.18	1.72	1.35	0.89	0.46	75	Fair	Dark brown
20% salt												
After 1 month	6.3	1.1699	29.67	17.00	2.00	1.66	1.19	0.85	0.34	70	Good	Brown
After 2 months	6.6	1.1706	30.61	17.38	2.07	1.62	1.18	0.83	0.35	70	Good	Reddish brown
25% salt												
After 1 month	6.5	1.1709	28.61	20.00	1.65	1.36	0.93	0.64	0.29	65	Good	Brown
After 2 months	6.1	1.1964	32.17	20.63	1.76	1.48	0.94	0.66	0.28	70	Good	Dark brown
30% salt												
After 1 month	6.1	1.2162	33.27	24.19	1.33	1.18	0.75	0.60	0.15	65	Good	Yellowish brown
After 2 months	6.0	1.2162	34.25	24.19	1.57	1.38	0.81	0.62	0.19	70	Good	Brown

TABLE 2

RESULTS OF PLA KRATAK FERMENTATIONS WITH DIFFERENT SALT CONCENTRATIONS BUT SIMILARLY INCUBATED AT 49°C FOR TWO DAYS THEN KEPT AT 37°C FOR TWO MONTHS

Salt concentration	pH	Specific gravity	Total solids (g-%)	Sodium chloride (g-%)	Total N (g-%)	Organic N (g-%)	Formal-dehyde N (g-%)	Amino acid N (g-%)	Ammonia N (g-%)	Liq. portion from 150 ml (ml)	Aroma	Colour
20 % salt												
After 1 month	5.5	1.1876	31.85	18.98	2.08	1.87	0.84	0.63	0.21	40	Fair	Yellow
After 2 months	6.0	1.1880	33.91	19.11	1.99	1.79	1.21	1.01	0.20	75	Good	Brown
25% salt												
After 1 month	5.5	1.2077	33.27	21.74	1.92	1.70	0.84	0.62	0.22	40	Good	Yellow
After 2 months	6.2	1.1876	34.50	21.57	1.91	1.72	0.97	0.78	0.19	70	Good	Yellowish brown
30% salt												
After 1 month	5.8	1.2146	32.43	24.21	1.28	1.15	0.60	0.47	0.13	60	Good	Yellow
After 2 months	5.5	1.2093	32.39	23.43	1.33	1.19	0.65	0.51	0.14	75	Good	Brown

TABLE 3

RESULTS OF PLA THU FERMENTATIONS WITH DIFFERENT SALT CONCENTRATIONS BUT SIMILARLY INCUBATED AT 49°C FOR ONE-TWO DAYS THEN KEPT AT 37°C FOR TWO MONTHS

Salt concentration	pH	Specific gravity	Total solids (g-%)	Sodium chloride (g-%)	Total N (g-%)	Organic N (g-%)	Formal-dehyde N (g-%)	Amino acid N (g-%)	Ammonia N (g-%)	Liq. portion from 150 ml (ml)	Aroma	Colour
						COARSELY CHOPPED FISH						
15 % salt												
After 1 month	6.7	1.1500	27.62	14.27	2.28	1.75	1.24	0.71	0.53	75	Fair, sour	Reddish brown
After 2 months	6.5	1.1495	27.59	14.05	2.34	1.81	1.33	0.80	0.53	80	Fair, sour	Reddish brown
20 % salt												
After 1 month	5.7	1.1703	36.52	16.95	1.91	1.71	0.79	0.59	0.20	75	Fair	Brown
After 2 months	5.4	1.1797	31.64	17.65	2.17	1.92	0.96	0.71	0.25	75	Rather good	Brown
25 % salt												
After 1 month	5.7	1.2024	33.36	21.06	1.64	1.42	0.65	0.53	0.12	70	Fair	Brown
After 2 months	5.4	1.2058	33.44	21.47	1.92	1.76	0.78	0.62	0.16	75	Rather good	Brown
30 % salt												
After 2 months	5.6	1.2243	35.80	24.22	1.67	1.44	0.66	0.53	0.13	60	Fair	Brown
After 3 months	5.1	1.2260	35.46	24.22	1.70	1.55	0.74	0.59	0.15	65	Good	Brown
						FINELY CHOPPED FISH*						
25 % salt												
After 1 month	5.8	1.2045	31.46	21.21	1.82	1.67	0.83	0.68	0.15	80	Good	Brown
After 2 months	6.1	1.1921	32.30	19.75	1.86	1.66	0.91	0.71	0.20	80	Good	Brown
						MACHINE GROUND FISH*						
25 % salt												
After 1 month	5.7	1.1989	32.09	20.77	1.73	1.55	0.81	0.63	0.18	65	Good	Yellowish brown
After 2 months	6.0	1.2043	32.44	21.62	1.75	1.57	0.80	0.62	0.18	65	Good	Yellowish brown

*Incubation at 37°C for two months only.

TABLE 4
RESULTS OF FERMENTATIONS OF WHOLE AND FINELY CHOPPED PLA SAI DAENG

Salt concentration	pH	Specific gravity	Total solids (g-%)	Sodium chloride (g-%)	Total N (g-%)	Organic N (g-%)	Formal-dehyde N (g-%)	Amino acid N (g-%)	Ammonia N (g-%)	Liq. portion from 150 ml (ml)	Aroma	Colour
WHOLE FISH, INCUBATED AT 49°C FOR 4 DAYS, 37°C FOR 2 MONTHS												
25% salt												
After 1 month	6.0	1.1997	32.78	21.68	1.78	1.56	0.86	0.64	0.22	70	Rather good	Dark brown
After 2 months	5.9	1.2005	35.92	23.69	1.90	1.62	0.95	0.67	0.28	70	Good	Dark brown
FINELY CHOPPED FISH, INCUBATED AT 49°C FOR 4 DAYS, 37°C FOR 2 MONTHS												
25% salt												
After 1 month	6.1	1.1966	31.97	21.09	1.87	1.60	0.83	0.56	0.27	75	Fair	Dark brown
After 2 months	6.2	1.2069	33.40	21.69	1.95	1.66	0.93	0.64	0.29	80	Good	Dark brown
20% salt												
After 1 month	6.2	1.1822	31.18	18.05	2.19	1.87	1.07	0.75	0.32	75	Fair	Dark brown
After 2 months	6.5	1.1805	31.09	18.05	2.20	1.79	1.18	0.77	0.41	75	Good	Dark brown
FINELY CHOPPED FISH, INCUBATED AT 37°C FOR 2 MONTHS												
25% salt												
After 1 month	6.8	1.1928	32.08	20.70	1.83	1.47	1.04	0.68	0.36	30	Rather good	Yellow
After 2 months	6.5	1.1970	33.37	21.03	1.93	1.56	0.99	0.62	0.37	55	Rather good	Yellowish brown
20% salt												
After 1 month	7.6	1.1715	31.57	18.27	1.99	1.32	1.21	0.64	0.67	60	Bad	Yellowish brown
After 2 months	7.4	1.1705	31.61	19.15	2.21	1.46	1.27	0.52	0.75	65	Fair	Yellowish brown

TABLE 5
RESULTS OF CHEMICAL ANALYSIS OF FRESH RAW FISHES

Kind of fish	Moisture (g-%)	Total solids (g-%)	Total N (g-%)	Crude fat (ether extract) (g-%)
Pla kratak	70.57	29.43	3.21	4.83
Pla thu	74.18	25.82	3.31	3.30
Pla sai daeng	78.51	21.49	2.71	1.45

TABLE 6
RESULTS OF FERMENTATIONS OF PLA THU (INITIALLY PROCESSED WITH 15 PER CENT SALT, KEPT AT 49°C OVERNIGHT, AND AT 37°C FOR ONE MONTH) UNDER VARIOUS SUBSEQUENT TREATMENTS

Subsequent treatment	pH	Specific gravity	Total solids (g-%)	Sodium chloride (g-%)	Total N (g-%)	Organic N (g-%)	Formal-dehyde N (g-%)	Amino acid N (g-%)	Ammonia N (g-%)	Liq. portion from 150 ml (ml)	Aroma	Colour
None	6.3	1.1384	29.23	12.59	2.05	1.54	1.35	0.84	0.51	95	Fair, sour	Yellow
1 month in sun	6.4	1.1386	29.15	12.64	2.23	1.66	1.43	0.86	0.57	100	Fair, sour	Yellowish brown
15% NaCl added, 1 month in sun	6.3	1.2136	34.57	22.38	2.06	1.56	1.27	0.77	0.50	95	Rather good	Yellow
15% NaCl added, 1 month at 37°C	6.4	1.2207	33.39	23.41	1.91	1.40	1.25	0.74	0.51	90	Fair, not sour	Yellow
15% raw salt added, 1 month in sun	6.2	1.1822	33.44	21.49	1.67	1.26	1.09	0.68	0.41	100	Fair, not sour	Brownish yellow
15% raw salt added, 1 month at 37°C	6.1	1.2137	29.32	22.48	1.89	1.40	1.18	0.69	0.49	100	Fair, not sour	Brownish yellow

TABLE 7

RESULTS OF SUN LEACHING OF RESIDUES FROM DIFFERENT TYPES OF PLA KRATAK FERMENTATIONS (30 PER CENT SALT)

Original fermentation	Leaching time	pH	Specific gravity	Total solids (g-%)	Sodium chloride (g-%)	Total N (g-%)	Organic N (g-%)	Formal-dehyde N (g-%)	Amino acid N (g-%)	Ammonia N (g-%)	Aroma	Colour
2 days at 49°C,												
2 weeks at 37°C,	1 month	5.7	1.1832	27.08	22.89	0.77	0.71	0.37	0.31	0.06	Good	Brownish yellow
1 month in sun	2 months	5.4	1.1893	26.76	22.50	0.66	0.62	0.35	0.31	0.04	Good	Brownish yellow
2 days at 49°C, 2												
weeks at room temp.,	1 month	5.7	1.1949	27.07	22.93	0.78	0.69	0.45	0.36	0.09	Good	Brownish yellow
1 month in sun	2 months	5.5	1.1900	28.76	23.27	0.74	0.68	0.39	0.33	0.06	Good	Yellow
2 weeks at 49°C,	1 month	5.7	1.1843	26.77	21.87	0.74	0.66	0.37	0.25	0.08	Good	Brownish yellow
1 month in sun	2 months	5.5	1.1940	27.09	22.00	0.62	0.57	0.33	0.28	0.05	Good	Brown

TABLE 8
RESULTS OF LEACHING FOR ONE MONTH OF RESIDUES FROM VARIOUS ORIGINAL FERMENTATIONS

Original fermentation	Leaching	pH	Specific gravity	Total N (g-%)	Aroma	Colour
			SUN LEACHING			
Pla kratak, 20% salt, 1 month at 37°C, 1 month in sun	Leaching 1	7.7	1.1243	0.89	Fair	Brownish yellow
	Leaching 2	7.7	1.1174	0.34	Rather bad	Brownish yellow
	Leaching 3	7.6	1.1155	0.10	Bad	Yellow
Pla kratak, 25% salt, 1 month at 37°C, 1 month in sun	Leaching 1	6.8	1.1559	0.75	Fair	Yellowish brown
	Leaching 2	7.4	1.1438	0.43	Fair	Yellow
	Leaching 3	7.2	1.1378	0.12	Fair	Yellow
Pla kratak, 30% salt, 1 month at 37°C, 1 month in sun	Leaching 1	6.3	1.1950	0.77	Good	Brown
	Leaching 2	6.7	1.1812	0.32	Fair	Yellow
	Leaching 3	7.5	1.1744	0.15	Fair	Yellow
			INCUBATOR LEACHING AT 37°C			
Pla thu, 20% salt, 1 day at 49°C, 2 months at 37°C	Leaching 1	6.4	1.1564	1.06	Fair	Brown
	Leaching 2	6.2	1.1476	0.45	Fair	Brownish yellow
	Leaching 3	7.6	1.1447	0.18	Bad	Brownish yellow
Pla thu, 25% salt, 1 day at 49°C, 2 months at 37°C	Leaching 1	6.2	1.1665	0.97	Fair	Yellowish brown
	Leaching 2	6.2	1.1623	0.52	Fair	Brownish yellow
	Leaching 3	6.5	1.1631	0.29	Bad	Yellowish brown
Pla thu, 30% salt, 2 days at 49°C, 2 months at 37°C	Leaching 1	5.3	1.2150	1.06	Good	Yellowish brown
	Leaching 2	5.2	1.1947	0.62	Fair	Yellowish brown
	Leaching 3	5.6	1.1795	0.35	Fair	Brown
Pla sai daeng, whole, 25% salt, 4 days at 49°C, 2 months at 37°C	Leaching 1	6.1	1.1738	0.91	Fair	Brownish yellow
	Leaching 2	7.8	1.1524	0.48	Fair	Brownish yellow
	Leaching 3	7.4	1.1521	0.22	Fair	Yellow
Pla sai daeng, finely chopped, 25% salt, 4 days @ 49°C, 2mos.@ 37°C	Leaching 1	6.1	1.1768	1.20	Fair	Brownish yellow
	Leaching 2	7.2	1.1607	0.69	Fair	Brown
	Leaching 3	7.4	1.1517	0.26	Bad	Brownish yellow
Pla sai daeng, finely chopped, 20% salt, 4 days @ 49°C, 2 mos.@ 37°C	Leaching 1	6.9	1.1387	1.00	Fair	Brown
	Leaching 2	6.3	1.1264	0.43	Fair	Brownish yellow
	Leaching 3	6.3	1.1191	0.15	Bad	Brownish yellow
Pla sai daeng, finely chopped, 25% salt, 2 months at 37°C	Leaching 1	6.8	1.1704	0.87	Fair	Yellowish brown
	Leaching 2	6.8	1.1345	0.37	Fair	Yellowish brown
	Leaching 3	7.4	1.1271	0.14	Rather bad	Yellow
Pla sai daeng, finely chopped, 20% salt, 2 months at 37°C	Leaching 1	7.2	1.1407	0.88	Fair	Yellowish brown
	Leaching 2	7.2	1.1163	0.25	Fair	Brownish yellow
	Leaching 3	7.5	1.1209	0.10	Bad	Yellow

TABLE 9
RESULTS OF CHEMICAL ANALYSIS OF FISH RESIDUES FROM PLA THU FERMENTATIONS AFTER THE THIRD LEACHING

Original fermentation	Per cent sodium chloride (wt./wt.)	Per cent total N (wt./wt.)
20% salt, coarsely chopped, 1 day at 49°C, 2 months at 37°C	17.54	1.60
25% salt, coarsely chopped, 1 day at 49°C, 2 months at 37°C	18.29	2.05
30% salt, coarsely chopped, 2 days at 49°C, 2 months at 37°C	16.57	2.08
30% salt, coarsely chopped, 2 days at 49°C, 3 months at 37°C	16.88	2.14

DISCUSSION

From Table 1, it is evident that good quality fish sauce from pla kratak can be produced with the original salt content varying from 20 to 30 per cent by weight of the fish after incubation at 37°C for one month followed by sun exposure for another month. We noted that the higher the salt content, the better the aroma. Nitrogen retention in the primary extract is better at the lower salt concentrations. Sufficient breakdown of nitrogenous materials, as judged by the formal-

dehyde nitrogen content of the product according to the regulation of the Government of French Indochina (1943), can be obtained with the above salt concentrations in every case. The extra month of sun exposure helps to increase further the amounts of total nitrogen, organic nitrogen, formaldehyde nitrogen, and amino acid nitrogen. This is especially true with the product processed with 30 per cent salt, and very little so with the one processed with 20 per cent salt. The main advantage of sun exposure is in the improvement in colour and possibly the flavour of the finished product.

Continuing the incubation at 37°C to two months (Table 2) does not appreciably increase the amounts of total nitrogen and organic nitrogen in the primary extract of pla kratak fermentation. There are significant increases in the formaldehyde nitrogen and amino acid nitrogen in the products made with 20 and 25 per cent salt, however, but not quite so much with the product made with 30 per cent salt. This means that the treatment has some advantage in causing further breakdown of the fish protein, but it has to be weighed against the increase in the production cost. The degree of liquefaction with prolonged incubation at 37°C is not much better than with ordinary sun exposure (Table 1). Raising the initial incubation to 49°C for two days with this group of products was aimed at better disintegration of the fish and hence a better yield. This proved not to be the case. Liquefaction during the first month was retarded in all cases, more so with products made with 20 and 25 per cent salt.

For pla thu fermentations (Table 3), the coarsely chopped fish disintegrated well after initial incubation at 49°C for one-two days with all the four salt concentrations used. After removal to 37°C at least two more months were required for sufficient breakdown of nitrogenous materials and development of acceptable aroma. For the product processed with 30 per cent salt, three months at 37°C were needed. Product processed with 15 per cent salt developed sour odour and contained large amount of ammonia nitrogen and is not recommended.

Fine chopping and machine grinding of the salt and fish mixture after desliming can substitute for, or is even better than, primary incubation at 49°C. Good products could be obtained with 25 per cent salt after two months of incubation at 37°C.

Pla sai daeng (Table 4) lends itself to nampla fermentation more readily than pla thu. Good quality primary extract could be obtained from whole fish with 25 per cent salt after incubation at 49°C for four days and subsequently at 37°C for two months. Fine chopping and the same two-stage incubation also yielded good quality products with both 20 and 25 per cent salt. Fine chopping with only one-stage incubation at 37°C for two months and using 25 per cent salt also yielded acceptable product. Product obtained with the latter treatment but using 20 per cent salt was not good, containing high ammonia nitrogen and not so good aroma, however.

This is quite contrary to expectation. Pla sai daeng, because of its demersal nature, is presumed to contain less enzyme in the flesh and entrails than the pelagic pla thu (Amano 1962), and should be more difficult to ferment. Furthermore, pla sai daeng is the leanest of the three species so far as the contents of total solids, total nitrogen, and crude fat are concerned (Table 5). In this respect it should make poorer nampla than the other two species according to the conventional belief that the richer the species during the season, the better the nampla quality. Yet pla sai daeng makes better nampla than pla thu, and not far inferior to pla kratak. It would appear then that the suitability of a particular species for making nampla depends on something else besides the richness in enzyme and nutritious nature of the fish.

Conducting the initial fermentation of pla thu at a lower salt concentration (15%) to speed up the disintegration of the fish and the breakdown of nitrogenous compounds, and then adding more salt later to improve the aroma did not work out. In Table 6, with all four types of treatment, the aroma improved only slightly and acceptable product was not obtained. Moreover, addition of salt, whether in the form of pure sodium chloride or raw salt, caused reduction in all forms of nitrogen examined. Raw salt appeared to be worse than sodium chloride in this respect. Sun exposure alone caused slight further increase in all forms of nitrogen, but the aroma did not improve.

The failure to improve the aroma of the product by the addition of more salt in this case could be attributed to the fact that during the initial fermentation with low salt content there was too much putrefaction already, and it was not possible to get rid of the soluble putre-

factive products by mere addition of more salt. It is to be noticed from Table 6 that ammonia nitrogen content still remains high after the treatment. This is quite in contrast with the products of good aroma originally but which have gone bad during storage. In these cases we found that the addition of 5-10 per cent more salt together with sun exposure for two weeks did greatly improve the aroma of the product, whether it was primary extract or leaching.

For leaching (Table 8), it appears that generally it can be repeated once, and the second leachings in all cases still contain a fair amount of total nitrogen. Products originally fermented with the higher concentrations of salt seem to give better leachings than those made with 20 per cent salt. With pla thu, a third leaching can still be made, especially if the original fermentation has been carried out with 30 per cent salt. The third leaching still contains 0.35 gramme-per cent of total nitrogen and the aroma is fair. This is not true with pla kratak. Its third leachings contain only 0.10-0.15 gramme-per cent of total nitrogen irrespective of the salt concentration of the original fermentation, even though the aroma is still fair. For pla sai daeng, third leaching can still be made from the residue of the original fermentation carried out with whole fish and 25 per cent salt. The third leaching contains 0.22 per cent total nitrogen and has fair aroma. Pla sai daeng was not processed with 30 per cent salt, but if it had been, the third leaching would certainly be good.

It is to be noticed from Table 9 that the higher the percentage of the salt used in the original fermentation (and hence in the brine used for leaching), the higher the amount of total nitrogen retained in the final residue after the third leaching.

Since profit from commercial nampla production comes mainly from the sale of lower grade products, it would appear then that the production by the new quick process had better be carried out with 30 per cent salt. This would ensure a primary product of good aroma and the production of a larger amount of acceptable leachings.

Leaching studies being reported were all made with the same salt concentration as the original fermentation. Leaching with saturated brine has not yet been attempted. It seems probable that this procedure

might improve the quality and increase the number of good leachings of each fermentation.

CONCLUSIONS

(1) Effective salt concentrations for nampla fermentation are from 20 to 30 per cent by weight of the raw fish. This is true for all three species of fish tested.

(2) Lower salt concentration favours more rapid disintegration of the fish and better breakdown of nitrogenous materials, but the higher the salt content the better the aroma.

(3) It is sufficient to incubate pla kratak fermentation at 37°C only and for one month, to be followed by sun exposure for another month. Raising the initial fermentation temperature to 49°C and then prolonging the 37°C incubation to two months is only slightly more advantageous, but would not be worth the extra cost of prolonged incubation.

(4) For pla thu and pla sai daeng, the larger-sized fishes, it is necessary to conduct the initial fermentation at 49°C until the fish can be well disintegrated by stirring and to continue the rest of the fermentation at 37°C for two to three months. Fine chopping and machine grinding of the salt and fish mixture after desliming can substitute for the initial incubation at 49°C where the fermentation is carried out with 25 per cent salt. In the case of pla thu, better products are obtained with the latter treatments.

(5) Pla sai daeng lends itself to nampla fermentation more readily than pla thu.

(6) Leaching can be made with salt solution of the same concentration as that of the original fermentation. In general two acceptable leachings are obtained from the residue of the original fermentation, but for the larger-sized fishes, if processed with 30 per cent salt, one more leaching can be obtained.

(7) For the economics of commercial production, the fermentation with 30 per cent salt originally is recommended.

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