# FISKERIDIREKTORATETS SKRIFTER

# SERIE TEKNOLÓGISKE UNDERSØKELSER

VOL. V, NO. 7

Report on Technological Recearch concerning Norwegian Fish Industry

# THE NUTRITIVE VALUE OF THAI FISH PRODUCTS I. THE VITAMIN CONTENT

By

TASANEE SORASUCHART

Government Vitamin Laboratory Norwegian Fisheries Research Institute Bergen — Norway

DIRECTORATE OF FISHERIES BERGEN, NORWAY 1971

#### INTRODUCTION

Although the vitamin contents of fish and fish products have been investigated for many years, there is still a lack of data from many parts of the world. The data in the literature on B-vitamins in fish and fish products have been reviewed by KUHNAU (1956), Love et al. (1959), BRAEKKAN (1962) and HIGASHI (1962). Except for an investigation of different fish sauces by SARKORNMONKOL et al. (1968), no study reports on the contents of B-vitamins in Thai fish and fish products. The present study reports the results of an investigation of B-vitamins in products from the market in Bangkok. The samples were sent to Bergen by airfreight and arrived in good conditions for nutritional investigations.

### MATERIALS

Altogether 19 samples of different fish products were investigated. As most products are unfamiliar to readers outside Thailand and the Far East, a brief description are given below.

Dried Snake-head (Ophicephalus striatus BLOCH) is produced by beheading, scaling and splitting of the fish along the back. It is then soaked in weak brine before being dried in the sun.

Dried Squid (Loligo sp.) is produced by removing the entrails and washing in sea water. The body is split lengthwise and soaked in weak brine before drying.

Dried Shrimps (Peneaus sp.) are produced by washing the shrimps in sea water, followed by half an hour boiling in fresh water with or without added salt. They are then dried in the sun, put in sacks and beaten against wooden boards to loosen and separate the shells from the meat.

Dried Mussels (Mytilus sp.) are produced by scalding of the shells to open them. The contents are taken out and boiled for 5 min in weak brine followed by sun-drying.

Salted dried pla tu or pedah Siam (Rastrelliger kanagurta CUVIER) is produced by removing gills and viscera through the throat of the fish. The gutted fish is washed and salted in brine. When buyers orders are received they are taken out and dried in the sun.

Mackerel (Cybium commersoni LACÉPÈDE) is gutted, salted in brine and sun dried.

Threadfin (Eleutheronema tetradactylum SHAW) is salted in the form of scaled and gutted fish. They are subsequently dried in the sun, sometimes with salt put in the belly cavity to prolong the storage life.

Pla slid (Trichogaster pectoralis REGAN) is scaled, beheaded, soaked in strong brine and sun-dried.

Leatherskin (Chorinemus lysan FORSKÅL) is scaled, split, salted and sun dried. The backbone is removed during dressing.

Carp (Puntius gonionotus BLEEKER). The fish is split through body and head, dry-salted and dried in the sun. Salted small carps are often fried and eaten with head and bones.

Ray (Dasyatis sp.). Only the flesh near the tail is used for salting. The skin is removed, the flesh cut longitudinally on both sides and dried spread wide out.

Anchovy (Stolephorus sp.). The whole fish is mixed with salt and subsequently dried in the sun. They are eaten whole with the bones after frying.

Mullet (Mugil sp.) is salted in the form of the split fish with the head on. The fish is fried and eaten whole.

With regard to the frying of the above species of salted dried products, the normal process is baking over a charcoal fire.

Smoked Catfish (Ompok bimaculatus BLOCH). The whole fish is smoked in bundles with a bamboo stick through their head and their tails stuck together by the slime of the fish.

*Featherback* (*Notopterus notopterus* PALLAS) is smoked whole. The smoked fish is mainly used as a flavoring additive to various dishes such as sauces, curries etc.

Seasoned dried catfish (Tachysurus sp.) is prepared by slicing the muscle longitudinally. The fillets are soaked in a mixture of nam pla (see below) and palm sugar, and subsequently dried in the sun. They are considered a delicacy when baked over the open fire.

Pla ra or fermented snake-head (Ophicephalus striatus BLOCH). Round fish is salted and fermented with roasted rice.

Nam pla or Fish sauce (Stolephorus spp.). Whole fish are mixed with salt and allowed to stand for 5 to 18 months. The clear liquid is drained off, filtered and exposed to the sun until ripe. The liquid varies in colour from yellow amber to dark brown. It is very salty and has a characteristic flavour. The sauce is mainly used as additive to rice.

Kapi or shrimp paste (Acetes sp; Myses sp.). The planktonic crustaceans

are mixed with salt, pounded and exposed to the sun until ripe. This class of products are generally used as condiments.

Description of some products and details of methods of preparation have been reported by Subba Rao (1967).

#### METHODS

For each treatment described below, an amount of sample corresponding to approximately 1 g dry matter (or 2—3 g wet sample) was weighed out. The analyses were carried out on the edible portions of the samples described above.

*Niacin* was extracted by autoclaving the sample with 100 ml 0.5 N  $H_2SO_4$  for 30 min at 120°C. After cooling the digest was adjusted to pH 4.5, diluted to a suitable concentration and filtered. Aliquots were adjusted to pH 6.8 before diluting to the final concentration.

*Biotin* was extracted by autoclaving the sample with 25 ml 3 N  $H_2SO_4$  for 3 hrs. at 120°C. The digest was adjusted to pH 4.5, diluted and filtered. Aliquotes were adjusted to pH 6.8 before final dilution.

Pantothenic acid was liberated by enzymatic digestion. The sample was dispersed in 10 ml 0.15 M sodium acetate buffer of pH 4.5 and 1 ml of a suspension of 20 mg takadiastase + 20 mg papain were added. The mixture was layered with toluene and incubated ca. 20 hrs. at 37°C. At the end of this time the digest was autoclaved for 5 min at 120°C, cooled and made up to volume and filtered. Aliquots were adjusted to pH 6.8 and diluted to a suitable concentration. The enzymes were practically free from pantothenic acid, thus a blank could be omitted.

*Riboflavin* was extracted by autoclaving the sample with 50 ml 0.1 N HCl for 30 min. After cooling the pH was adjusted to ca. 6.5 by 2 N NaOH and readjusted to pH 4.5 by 2 N CH<sub>3</sub>COOH, diluted to volume and filtered. The pH of aliquots were adjusted to 6.8 before final dilution and filtered if necessary.

Vitamin  $B_{12}$  was extracted by autoclaving the sample with 50 ml 0.066 M sodium acetate + 1 ml 1 % KCN solution for 15 min. The digest was made up to volume and filtered. The pH of aliquots taken were adjusted to 5.5 before dilution to suitable concentration.

Thiamine was extracted by steaming the sample with 50 ml 0.1 N  $H_2SO_4$  for 30 min. After cooling the pH was adjusted to 4.5 with 2.5 M sodium acetate solution. 2 ml of a suspension of 20 mg papain + 20 mg takadiastase pr. ml were added and the mixture was layered with toluene and incubated overnight at 37°C. The mixture was then steamed for 30 min made up to volume and filtered. Suitable aliquots were adjusted

to pH 5.5 before final dilution. The amount of thiamine in the enzymes were negligible, thus a blank could be omitted.

Vitamin  $B_6$  was extracted by autoclaving the sample with 200 ml 0.055 N  $H_2SO_4$  for 4 hrs. After cooling the extract was adjusted to pH 4.5 diluted to volume and filtered. Aliquots were diluted to suitable concentration.

Microorganisms employed, incubation temperatures and times, and methods for measurement of the growth response, are described in Table 1. In all assays  $16 \times 150$  mm Pyrex test tubes were used. The assay volumes were: 10 ml for thiamine, pantothenic acid, vitamin B<sub>12</sub> and biotin; 5 ml for vitamin B<sub>6</sub>; and 2 ml for riboflavin and niacin. Incubations were carried out in a water bath with the exception of vitamin B<sub>6</sub>, which was incubated under shaking in an air incubator. In the latter assay a standardized glass bead was added to each tube. The basal medium used for niacin, riboflavin, pantothenic acid and biotin were essentially as describibed in Pharmacopea Nordica (1960). For thiamine the medium described by MACIAS-R (1957), for vitamin B<sub>12</sub> the ready prepared medium from A/S Ferrosan, Denmark, and for vitamin B<sub>6</sub> the medium described by STORWICK et al. (1964), were employed.

The test organisms *Lactobacillus casei* and *Lactobacillus plantarum* were maintained as stab culture by weekly transfer on Bacto Micro Assay Culture Agar (Difco). After incubation for approximately 30 hrs. at the proper temperature (Table 1), the stab culture was stored at  $4-6^{\circ}$ C.

Lactobacillus leichmannii was maintained on liquid skim milk medium by daily transfer and incubation at 37°C. From Saturday to Monday the culture was stored in the cold.

Saccharomyces carlsbergensis was kept as slope culture on a maltose agar. The yeast was transferred biweekly and incubated for ca. 30 hrs. at  $30^{\circ}$ C and stored at  $4-6^{\circ}$ C.

Inoculum of yeast cells was prepared directly from a slope culture not older then five days. All other inocula were grown on Bacto Micro Inoculum Broth (Difco) over night, centrifuged and resuspended to suitable concentration in 0.9% saline solution.

Moisture was determined by drying in an oven at 105°C until constant weight.

Nitrogen was determined by the Kjeldahl method and the protein content calculated as N  $\times$  6.25.

# **RESULTS AND DISCUSSION**

In Table 2 are reported percentage edible portion, and the dry matter and protein (N  $\times$  6.25) contents of the edible portion of the products investigated.

Table 3 summarizes the results from the vitamin assays. When considering the values, it should be kept in mind that most products have been through methods of processing which may cause losses as well as increases in the vitamin contents. Thus salting and sundrying would make possible losses by extraction as well as by radiation, whereas in fermented product bacterial synthesis may have taken place. In the following discussion values for each vitamin have mainly been considered compared to the averages reported by BRAEKKAN (1962a, 1962b) in his compilations of literature values from all over the world.

The thiamine content was low in all products with the exception of dried snake-head and seasoned dried catfish, which showed moderate contents. PRADHAN & CHITRE (1951) and JOSHI et al. (1953) reported thiamine values from four different species of Indian salt water fish. Their results, in the order of  $2-3 \mu g$  thiamine per g wet weight, are unusually high compared with values from fish caught in other areas of the world, and should be reinvestigated. The low values in the present samples may reflect losses during processing, but the presence of thiaminases in some of the species is also most likely. DEOLALKAR and SOHONIE (1952 a and b) reported a common occurrence of thiaminases in Indian fishes from fresh, brackish and salt water. The present results showed that Thai fish products are low in thiamine and that these foods contribute very moderately to the daily nutritional requirement.

Riboflavin showed great variations between the different products, but generally lower values than reported on a world average by BRAEKKAN (1962a, 1962b). Boiled, dried mussel and pla slid with contents in the order of  $6.6 - 6.8 \ \mu g$  per g, are, however, important nutritional sources for riboflavin. JOSHI et al. (1953) also reported results for riboflavin in four Bombay species. As for thiamine, their finding are higher than usually reported for fish. Their extremely high values for the livers, 185, 164, 153 and 51  $\mu g$  per g are unique, and should be reinvestigated. If their results were valid, this food in small amounts would supply the total daily requirement.

The niacin contents varied considerably, and were usually lower than found in products from other areas (BRAEKKAN 1962a, 1962b). Especially pla slid and anchovy in the group of salted dried fish showed relatively low values, and the fermented product pla ra had a very low niacin content. Salted dried mackerel showed the highest value with 101  $\mu$ g per g followed by seasoned dried catfish and salted dried ray, with respectively 96 and 89  $\mu$ g per g. Previously have been reported some investigations on niacin in fish from India waters. KHORANA et al. (1942) reported 20—40  $\mu$ g per g flesh from marine fish. BRAGANCA (1944) found the highest value in the marine pomfret (*Stromateus* sp.). THAKUR and KARANDIKAR (1951) considered fish most commonly eaten in Bengal are low in niacin. The present results are generally in agreement with their conclusion. JOSHI et al. (1953) reported, however, high values also for niacin in their investigation of fresh fish.

Vitamin  $B_{12}$  showed the highest value in mussels, in agreement with the common concentration of this vitamin in most molluscs (BRÆKKAN, 1962). Salted dried carp, anchovy and mullet as well as seasoned dried catfish showed fairly high values, whereas the remaining products in general are good sources for vitamin  $B_{12}$ .

Vitamin  $B_6$  is usually present in fish in relatively high amounts (BRAEKKAN, 1969). The products investigated in the present study showed somewhat lower values than usually found in fish and fish products, indicating a loss during the different forms of processing.

Pantothenic acid showed values in the same range as those reported for other cured fish products of low moisture content (BRÆKKAN, 1962).

Biotin showed fairly high values compared with most food products, thus confirming that fish and fish products are good sources for this B-vitamin.

### SUMMARY

The contents of the B-vitamins: thiamine, riboflavin, niacin, vitamin  $B_{12}$ , vitamin  $B_6$ , pantothenic acid and biotin, were determined in 19 samples of Thai fish products from different or differently processed species bought at the market in Bangkok.

The vitamin determinations were carried out by microbiological methods, and the vitamins expressed as  $\mu g/g$  edible portion of the products.

The results showed the different B-vitamins to vary fairly extensively, but they were in general of the order reported for cured fish on a world basis.

# ACKNOWLEDGMENT

The author carried out this study on a fellowship from the Norwegian Agency for International Development (NORAD). I am very grateful to Dr. O. R. Brækkan and Mr. G. Boge for help and encouragement.

> The authors present address TASANEE SORASUCHART FACULTY OF FISHERIES KASETSART UNIVERSITY BANGKOK 9, THAILAND

#### REFERENCES

BRAGANCA, B. de M. (1944): Ann. Biochem. Exptl. Med. (Calcutta) 4, 41.

- BRAEKKAN, O. R. (1962a): In «Fish in Nutrition» Edited by HEEN and KREUZER, London. pp. 132—140.
  - (1962b): Ibid. pp. 141—145.
  - (1969): In «Fish ein zeitgemässe Lebensmittel». Edited by MEYER-WARDEN, Berlin pp. 105—116.
- DEOLALKAR, S. T. & SOHONIE, K. (1952a): Nature 173, 489.

— (1952b): Current Sci. (India) 21, 13.

- HIGASHI, H. (1962): In «Fish in Nutrition». Edited by HEEN and KREUZER. London pp. 125-131.
- JOSHI, S., MASTER, F. & MAGAS, N. G. (1953): Indian J. Med. Res. 41, 431.
- KHORANA, M. L., SARMA, M. L., & GIRI, K. V. (1942): Indian J. Med. 30, 315.
- KUHNAU, J. (1956): Arch. f. Fischereiwissenchaft 1. Beihefte, 7.
- Love, R. M., Lovern, J. A. & Jones, N. R. (1959): Food Investigation, Special Rep. No. 69, London.
- MACIAS-R, F. M. (1957): Appl. Microbiology 5, 249.
- PHARMACOPEA NORDICA. Edito Norvegica (1960): Bd. IV.
- PRADHAN, V. & CHITRE, R. G. (1951): Indian J. Med. Sci. 5, 645.
- SARKORNMONKOL, V., PANPRAPAI, P., SUKMEUN, A., SARKORNMONKOL, S., and KOMOL-PELIN, K. (1968): Res. Rep. No. 199. Dept. Sci. Ministry of Industry, Bangkok (In Thai).
- STORWICK, C. A., BENSON, E. M., EDWARDS, M. A., & WOODRING, M. J. (1964): In «Methods of Biochemical Analysis». Edited by GLICK. Interscience. Vol. XII, 183—276.
- SUBBA RAO, G. N. (1967): Indo-Pacific Fisheries Council Regional Study No. 4, FAO Regional Office for Asia and the Far East, Bangkok.

THAKUR, S. S. & KARANDIKAR, K. R. (1951): J. Univ. Bombay Sect. B 19, 64.

Vitamin	Test organism	ATCC*) No.	Standard range µg/tubes	Incubation tempera- ture °C	Incubation time hr.	Methods for measurement of the growth
Riboflavin Niacin Pantothenic acid Biotin Thiamine Vitamin B <sub>12</sub> Vitamin B.	Lactobacillus casei	(7469) (8014) (8014) (8014) (9338) (4797) (4228)	$\begin{array}{c} 0 & - 0.10 \\ 0 & - 0.10 \\ 0 & - 0.125 \\ 0 & - 0.0005 \\ 0 & - 0.025 \\ 0 & - 0.000125 \\ 0 & - 0.00625 \end{array}$	37 37 30 30 37 37 30	72 72 20 20 18 20 18	Titrimetric » Turbidimetric**) » » » »

-

Table 1. Microbiological methods employed for B-vitamin assay.

\*) American Type Culture Collection, 12301 Parklawn Drive, Rockville, Maryland, U.S.A.

\*\*) The growth was measured in a Beckman Model B Spectrophotometer.

9

Table 2. Percentage edible portion of the products and percentages dry matter and protein of the edible portion.

Sample		Edible portion %	Dry matter %	Protein (N×6.25) %	
Dried ma	ke head	70	75.0		
Dileu sila	:d	70	75.9	60.0	
» squ		100	74.9	65.6	
» shr		100	77.1	61.9	
» mu	ssel	100	58.6	39.5	
Salted drie	ed pla tu (pedah Siam)	57	53.8	25.6	
». »	mackerel	68	51.4	31.3	
» »	threadfin	57	50.9	32.6	
» »	pla slid	66	58.0	41.8	
» »	leatherskin	63	64.9	42.8	
» »	carp	90	72.8	43.4	
» »	ray	100	71.8	58.9	
» »	anchovy	100	68.5	45.9	
» »	mullet	100	53.4	28.4	
Smoked ca	utfish	62	88.4	71.3	
» fe	atherback	65	89.8	67.8	
Seasoned d	lried catfish	100	76.9	28.8	
Pla ra		39	40.9	20.8	
Nam pla		100	35.6	11.6	
Kapi		100	61.0	22.6	

Product	Thiamine	Ribo- flavin	Niacin	Vitamin B <sub>12</sub>	Vitamin B <sub>6</sub>	Panto- thenic acid	Biotin
	$\mu g/g$	$\mu { m g}/{ m g}$	$\mu { m g}/{ m g}$	$\mu { m g}/{ m g}$	$\mu \mathrm{g}/\mathrm{g}$	$\mu { m g}/{ m g}$	$\mu g/g$
Dried product							
Snake-head	2.4	2.03	51.4	0.036		16.5	0.021
Squid	0.4	0.90	65.7	0.084	0.66	9.5	0.115
Boiled dried product							
Shrimp	0.9	0.97	33.7	0.05	ca. 0.3	6.16	0.093
Mussel	0.3	6.80	19.6	0.37	ca. 2.5	7.54	0.26
Salted dried fish							
Pla tu		1.73	74.2	0.06	0.57	3.71	0.053
Mackerel	0.4	1.92	101.0	0.032	1.56	4.41	0.040
Threadfin	0.9	2.57	53.6	0.067	1.06	5.88	0.040
Pla slid	0.6	6.63	27.7	0.027	0.61	18.6	0.125
Leatherskin	0.5	1.50	46.4	0.04	0.87	4.17	0.051
Carp	0.1	1.57	38.6	0.096	1.02	20.4	0.082
Ray	1.6	1.39	89.3	0.053	1.89	10.2	0.059
Anchovy	0.1	1.10	23.1	0.11	ca. 0.6	10.2	0.145
Mullet	0.1	3.45	38.5	0.10	0.56	10.6	0.068
Smoked fish							
Catfish	0.1	1.92	65.5	0.15	0.59	17.7	0.102
Featherback	0.3	1.72	34.3	0.029	1.49	25.3	0.056
Seasoned dried fish							
Catfish	3.6	1.52	96.4	0.027	1.35	8.60	0.033
Fermented products							
Pla ra		0.99	6.4	0.023	0.4	2.25	0.016
Nam pla	traces	0.67	43.9	0.015	0.95	3.30	0.039
Kapi	0.1	1.73	24.7	0.052	ca. 0.5	10.1	0.105

Table 3. B-vitamin contents in edible portions of Thai fish products.