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# Polyunsaturated fatty acid contents of some traditional fish and shrimp paste condiments of the Philippines

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**Abstract**

The fatty acid composition as well as the water, NaCl, ash and fat contents of six types of shrimp and fish paste condiments of the Philippines were determined. The condiments were prepared by incubating the fish or shrimp in high concentrations of salt and under high humidity at ambient temperature over several months. The primary objective was to assess the contents of polyunsaturated fatty acids, and in particular the content of all-*cis*-4,7,10,13,16,19-docosahexaenoic acid (DHA) in these products. The product derived from small shrimp fry (*Acetes* spp.) had the highest content of DHA. The contents of polyunsaturated fatty acids, including DHA, of this particular condiment, were not significantly different from that of fresh untreated shrimp fry, indicating the presence of mechanisms that protect against polyunsaturated fatty acid autoxidation during the preparation process. Also, the DHA contents of the condiment derived from juvenile *Siganus* approached that of the *Acetes* condiment because of its relatively high fat content. In conclusion, two of the fish and shrimp paste condiments tested may be sources of dietary DHA for those who consume this type of food. 

## 1. Introduction

The Philippines is a country in Southeast Asia composed of a group of islands situated 115–130°E longitude, and 5–22°N latitude. Its total population is 68.6 million as of 1995 (National Statistics Office, 2000). Among traditional foods that persist and remain part of the regular diet of a large segment of the population, is a type of condiment known locally as *bagoong*. Bagoong is prepared from the wet-treatment of fish or shrimp fry with salt over a time period of 3–12 months at ambient temperature (Espejo-Hermes, 1998). The finished product has a paste-like consistency and is consumed as is, or used as a flavour extender in the preparation of other types of indigenous food. Other population groups in Asia also consume similar products described as "salt-fermented fish" (Cha & Cadwallader, 1995; Ohshima, Yankah, Ushio, & Kiozumi, 1998). Currently, this product has become available in bottled format in speciality food shops outside of Asia, particularly in North America.

Fish and other types of marine-derived food are good sources of long-chain polyunsaturated fatty acids belonging to the $\omega$-3 (or *n*-3) family. $\omega$-3 Fatty acids are essential for neural development in the infant in utero and during the first few years after birth (Uauy, Mena, & Rojas, 2000). The particular $\omega$-3 fatty acid incorporated in the brain and retina of the developing infant is all-*cis*-4,7,10,13,16,19-docosahexaenoic acid or DHA. Since the human body lacks the enzymes to manufacture $\omega$-3 fatty acids, DHA must be derived from the diet, or produced in vivo from diet-derived $\omega$-3 fatty acid precursors such as $\alpha$-linolenic acid.

By virtue of its origin, fish and shrimp paste condiments might also be significant dietary sources of long-chain $\omega$-3 polyunsaturated fatty acids. However, the long periods of incubation in brine solution at an average ambient temperature of 30°C may result in the loss of these highly unsaturated fatty acids through oxidative deterioration. Our objective was to measure the fatty acid composition, as well as the moisture, NaCl, ash and fat contents of several varieties of fish and shrimp paste condiments obtained from local producers, with a view to assessing their potential contribution to dietary long-chain $\omega$-3 polyunsaturated fatty acids. The data obtained will be useful in nutritional evaluation of segments of the population consuming these food products.

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## 2. Materials and methods

Samples of fish paste and shrimp condiments were purchased from the main public market in Bolinao, a coastal town on the South China Sea in the central region of Luzon archipelago of the Philippines. Vendors were interviewed concerning the nature of their product, e.g. the starting material, percentage of salt used and the treatment time. The type of fish was verified visually where possible. It was impossible to verify the accuracy of reported preparation times or to ascertain the quality and physical characteristics of the salt used in the process. Samples were analyzed fresh or were lyophilized and stored at −20°C pending further analysis.

The salinity (salt content) of the samples was determined using the Mohr Method (Association of Official Analytical Chemists, 1990). The moisture content was calculated from the loss in weight of the sample after drying. Briefly, fresh samples were weighed to the nearest mg, then dried in a vacuum oven at 100±5°C at 1.3 psi for 2 h and cooled to ambient temperature in a desiccator for 1 h before weighing. Oven-drying was repeated for 1 h periods until constant weight was attained. The dried samples from the moisture determination were incinerated at 550°C to constant weight in porcelain crucibles to measure ash content (Food Chemical Codex, 1981). The water activities of model salt solutions were experimentally determined by equilibration in a sealed chamber. The water activity was assumed to be equal to the relative water vapour pressure over the salt solution in the sealed chamber after equilibration. The total fat content was measured by Soxhlet extraction of the lyophilized samples using low-boiling petroleum ether as solvent. The fatty acid composition was determined separately by an adaptation of a direct methylation procedure on the sample, followed by gas chromatography (Lepage and Roy, 1986). Briefly, 50 mg of lyophilized sample were suspended in 2 ml of a mixture of hexane/methanol (1:4) plus heptadecanoic acid as the internal standard in a 5 ml test tube with a magnetic stirrer and Teflon-lined screw cap. To the mixture was added 200 μl of acetyl chloride (Sigma Chemical Co., St. Louis MO). The tube was closed tightly and placed for 60 min on a heating block/magnetic stirrer at 100°C. Reflux occurred on the walls of the tube exposed to circulating air above the heating block. After the reaction period, the tube was cooled and excess of acid was neutralized with 5.0 ml aqueous 6% $K_2CO_3$, w/v. At this point, the mixture separated into two phases and the upper phase (hexane), containing the fatty acid methyl esters, was collected for analysis by gas chromatography.

Gas chromatography was performed using a Hewlett-Packard Model 5890 equipped with a 100 m, 0.2 mm internal diameter SP2380 fused silica capillary column (Supelco, Ontario Canada). Detection was by flame ionization and peaks were recorded and measured with a Hewlett-Packard 3396 electronic integrator. Commercial reference mixtures (NuChek Prep, Elysian MN, USA) were used to calibrate the instrument. Peaks were assigned based on comparison of retention times. Fatty acid composition is expressed as weight percent, calculated from the area (percent) corrected for instrument response. The total amount of fatty acids was calculated, based on the recovery of the heptadecanoic acid internal standard.

## 3. Results and discussion

Sample description is presented in Table 1. All samples are of marine origin, readily recognized by the indigenous population as edible species, and consumed fresh, unsalted, or processed into fish or shrimp paste condiments. Field interviews of local market vendors were conducted to obtain anecdotal descriptions of the methods of preparation. In general, fish or shrimp fry and salt are deposited in alternating layers in containers, at a weight ratio of three to four parts salt to one part of fish or shrimp fry (wet weight). The mixture is incubated for the prescribed period of time at ambient temperature as indicated in Table 1.

Table 1
Fish and shrimp paste condiment sample description

| Starting material[a] | | | Preparation time (months)[b] |
|---|---|---|---|
| Species | Local name | English name | |
| *Caesio* spp. | Dalagang bukid | Fusiliers | 12 |
| *Spratelloides gracillis* | Dilis | Silver sprat | 5 |
| *Siganus fuscescens* | Padas-1[c] | Fuscescens rabbitfish | 7 |
| *Siganus argenteus* | Padas-2[d] | Forktail rabbitfish | 3 |
| *Pterocaesio* spp. | Terong | Fusiliers | 12 |
| *Acetes* spp. | Alamang | Shrimp fry | 5 |

[a] Samples are all marine species.
[b] According to field interviews of local market vendors.
[c] Prepared from juvenile fish of uniform size.
[d] Prepared from fish of varying size, considered by locals to be of inferior quality.

Table 2
Water, salt and fat content of fish and shrimp paste condiments

| Sample | Water | Fat | Ash | NaCl |
|---|---|---|---|---|
| | Mean±S.D. ($n=3$)[a] | | | |
| Dalagang bukid | 39.6±0.4 | 0.76±0.07 | 37.1±0.9 | 22.3±0.0 |
| Dilis | 39.2±0.4 | 0.60±0.06 | 38.2±1.0 | 23.4±0.9 |
| Padas-1 | 34.5±0.2 | 1.57±0.5 | 46.0±2.0 | 19.0±0.0 |
| Padas-2 | 34.0±3.2 | 0.73±0.1 | 45.6±1.1 | 23.5±0.7 |
| Terong | 39.4±0.6 | 0.97±0.05 | 38.0±0.3 | 21.6±1.8 |
| Alamang | 33.2±0.4 | 0.91±0.08 | 43.9±0.4 | 24.4±0.0 |

[a] Grammes per 100 g wet weight.

Anecdotal accounts of product preparation are not reliable for obvious reasons, foremost of which are errors in measurement of the ingredients, and variability in the quality and moisture content of the salt ingredient. This is shown by Table 2 in that even though the vendors' account of the traditional process calls for up to 80% (w/w) of salt added to the fish, chemical analysis of NaCl in the finished product reveals that the actual salt concentration varies between 19 and 24% (w/w), based on wet weight of the product. These salt quantities are more consistent with the process described by the Korean Fisheries Society (Anonymous, 1998) describing an experiment in which anchovy (*Engraulis japonica*) was treated with 10 or 20% salt.

The salt to water weight ratio in samples, calculated from Table 2, ranged from 0.55 for Padas-1 to 0.74 for Alamang. Model salt solutions of the same proportions gave water activities of 0.66 and 0.68 at 30°C, respectively. The values are similar because the salt proportions tested both gave saturated solutions. Nevertheless, these figures are likely to represent the upper limits of water activities of the fish and shrimp paste condiments. The actual water activities of the samples would be lower because of the presence of water-binding solutes in the samples other than NaCl. Other water-binding solutes that would be present in the product are proteins, peptides and carbohydrates from the fish, as well as inorganic substances, other than NaCl, as indicated by the high ash content (Table 2). Low water activity limits microbial growth (Beauchat, 1981) which gives a shelf-life advantage in a country where refrigeration is not uniformly available and where ambient temperature is tropical. Water activity of $<0.7$ is permissive to the growth of xerophilic molds and osmophilic yeasts but not to bacterial growth even of halophilic species.

Table 3
Fatty acid composition of fish and shrimp paste condiments

| Fatty acid | Dalagang bukid | Dilis | Padas-1 | Padas-2 | Terong | Alamang | Untreated Alamang[a] |
|---|---|---|---|---|---|---|---|
| | Mean±S.D. ($n=3$) | | | | | | |
| 8:0[b] | 1.1±1.1 | 1.01±0.05 | 0.07±0.06 | 0.31±0.05 | 0.30±0.07 | 0.29±0.06 | nd |
| 10:0 | 1.8±1.1 | 1.1±0.3 | 0.20±0.05 | 0.29±0.05 | 0.6±0.2 | 1.4±0.1 | nd |
| 12:0 | 1.3±0.4 | 2.5±0.5 | 0.54±0.07 | 1.4±0.2 | 1.09±0.04 | 0.8±0.2 | nd |
| 14:0 | 11.3±0.2 | 7.5±0.9 | 8.2±1.1 | 7.6±0.3 | 10.9±0.5 | 4.1±0.6 | 4.8±0.3 |
| 16:0 | 38.4±1.5 | 37.2±1.3 | 41.5±2.2 | 39.9±1.1 | 40.4±0.2 | 25.0±2.9 | 27.7±0.4 |
| 18:0 | 16.6±0.2 | 17.8±1.4 | 15.3±0.3 | 14.5±0.7 | 15.8±0.1 | 11.0±0.8 | 10.1±0.1 |
| 20:0 | 1.08±0.07 | 0.77±0.03 | 0.6±0.2 | 0.65±0.01 | 1.01±0.05 | 1.47±0.05 | 1.01±0.02 |
| 22:0 | 0.9±0.1 | 0.9±0.1 | 0.61±0.07 | 0.53±0.06 | 0.77±0.01 | 2.8±0.4 | 1.94±0.03 |
| 24:0 | 1.0±0.2 | 2.6±1.0 | 0.6±0.1 | 0.6±0.2 | 0.78±0.03 | 0.7±0.2 | 0.52±0.01 |
| 16:1 | 8.3±0.2 | 5.8±0.3 | 9.9±1.1 | 7.3±0.6 | 8.5±0.4 | 6.6±0.7 | 11.7±0.2 |
| 18:1 | 12.3±0.4 | 13.5±1.0 | 10.24±0.08 | 12.6±0.6 | 12.1±0.3 | 10.8±0.7 | 10.1±0.1 |
| 20:1 | 1.0±0.2 | 1.0±0.2 | 0.59±0.07 | 0.80±0.07 | 0.90±0.04 | 0.4±0.1 | 0.34±0.02 |
| 22:1 | 0.04±0.07 | 0.04±0.07 | 0.058±0.001 | 0.06±0.05 | 0.125±0.004 | nd | nd |
| 24:1 | 1.3±0.1 | 0.84±0.08 | 0.58±0.04 | 0.91±0.02 | 1.08±0.02 | 0.28±0.04 | nd |
| 18:2$\omega$6 | 0.63±0.03 | 0.9±0.1 | 0.73±0.04 | 1.0±0.1 | 0.81±0.06 | 2.9±0.2 | 2.52±0.03 |
| 18:3$\omega$6 | nd | 0.08±0.07 | 0.02±0.04 | nd | 0.03±0.05 | 0.16±0.06 | 0.25±0.06 |
| 20:2$\omega$6 | 0.12±0.11 | 0.18±0.16 | 0.187±0.005 | 0.19±0.02 | 0.171±0.004 | 0.44±0.03 | 0.41±0.06 |
| 20:3$\omega$6 | 0.11±0.20 | 0.07±0.06 | 0.10±0.03 | 0.03±0.04 | nd | 0.1±0.1 | 0.19±0.04 |
| 20:4$\omega$[c] | 0.50±0.17a | 1.3±0.4a | 1.2±0.4a | 1.1±0.2a | 0.6±0.1a | 5.8±0.6b | 5.88±0.07b |
| 22:4$\omega$6 | nd | 0.1±0.1 | 0.12±0.03 | 0.11±0.02 | 0.01±0.02 | 0.04±0.08 | nd |
| 18:3$\omega$3 | 0.14±0.12a | 0.22±0.20a | 0.22±0.02a | 0.27±0.05a | 0.264±0.006a | 2.15±0.05b | 1.23±0.02c |
| 20:5$\omega$3 | 0.5±0.4a | 1.2±0.2a | 1.7±0.6a | 1.5±0.3a | 1.2±0.2a | 11.0±1.6b | 10.9±0.1b |
| 22:5$\omega$3 | 0.05±0.08 | 0.3±0.2 | 1.3±0.3 | 1.5±0.3 | 0.25±0.06 | 0.3±0.3 | 0.47±0.02 |
| 22:6$\omega$3 | 1.7±0.2a | 3.0±0.3ab | 5.4±2.5ab | 6.7±1.2bc | 2.4±0.6ab | 11.5±3.3cd | 10.1±0.5c |
| Total saturates | 73.4±1.4a | 71.5±2.0ab | 67.6±2.9bc | 66.0±1.1c | 71.6±0.5ab | 47.3±4.0d | 46.0±0.6d |
| Total mono-unsaturates | 22.9±0.7 | 21.3±0.5 | 21.3±1.1 | 21.7±1.3 | 22.7±0.3 | 18.0±1.4 | 22.1±0.1 |
| Total poly-unsaturates | 3.7±0.8a | 7.3±1.8ab | 11.1±4.0b | 12.3±2.1b | 5.8±0.9ab | 34.7±5.4c | 31.9±0.7c |
| Total $\omega$6 | 1.4±0.4a | 2.6±0.9a | 2.4±0.5a | 2.4±0.4a | 1.6±0.1a | 9.5±0.6b | 9.2±0.1b |
| Total $\omega$3 | 2.3±0.4a | 4.7±0.9ab | 8.7±3.5b | 9.9±1.8b | 4.1±0.7ab | 25.2±4.8c | 22.6±4.8c |

[a] Fresh untreated sample was also analyzed for comparison purposes with the corresponding salt-fermented product.

[b] Fatty acids are identified as *m*:*n*$\omega$x, where *m* is the carbon chain length, *n* is the number of *cis* double bonds and $\omega$x indicates the class of polyunsaturated fatty acid.

[c] One-way ANOVA was performed on selected fatty acids using the routines available in SPSS. Differences between means were declared statistically significant at the 0.05 level using Tukey's-*b* test. Values sharing the same letter are not statistically different from each other.

The fat contents, by Soxhlet, expressed as percent of wet weight, are shown in Table 2 and range from a low of about 0.60% for dilis to 1.57% for padas-1. Although Soxhlet extraction is an established and common procedure for the measurement of fat, its use in food analysis has been questioned (Greenfield and Southgate, 1992). This is principally because the fat is exposed to heat and that the organic solvent used may not extract all types of fat, particularly the polar lipids. We therefore used the internal standard procedure for the quantitation of fatty acids using gas chromatography, to serve as a verification of the Soxhlet data. Dalagang bukid, dilis, padas-1, padas-2, terong and alamang had $6.5\pm0.1$, $3.8\pm0.2$, $12.4\pm3.4$, $5.3\pm0.4$, $6.0\pm0.4$ and $7.26\pm0.08$ mg/g wet weight, respectively. When expressed as percentages, these values are similar to the Soxhlet data (Table 2), both in degree and relative to each other. Figures for total fatty acids are consistently lower than total fat by Soxhlet (Table 2) because the latter contains other lipid components besides fatty acids, such as sterols, non-saponifiable hydrocarbons and glycerol. The similarities of both sets of data indicate that the Soxhlet procedure is valid in this instance.

Table 3 shows the total fatty acid composition of the samples including calculated values for the various groups of fatty acids. Alamang contains from about 2 to 10-fold higher proportions of total $\omega$-3 polyunsaturated fatty acids than all the other types of fish paste condiments (Table 3). This difference is due to higher proportions of DHA and of 20:5$\omega$3 in alamang compared with the other types of fish paste condiments analyzed. Dalagang bukid had the lowest levels of DHA while padas-1 and padas-2 were intermediate. Conversely, alamang had the lowest proportion of saturated fat, dalagang bukid the highest and padas-1 and padas-2, intermediate. In terms of real amounts, the DHA content of padas-1 approaches that of alamang by virtue of this product's higher fat content (Table 2). It is interesting to note that padas-1 has twice as much fat as padas-2 even though both are derived from the same family of fish.

For comparison purposes, data from fresh untreated alamang are shown alongside the corresponding values for salt-treated alamang condiment. Remarkably, the fatty acid profile of alamang was largely unaffected by the salt incubation process. Except for $\alpha$-linolenic acid (18:3$\omega$), values for the long-chain polyunsaturated fatty acids were the same. This indicates that oxidative deterioration of lipids during the salt-incubation of the alamang, if it had occurred at all, must have been significantly suppressed, so that the highly unsaturated fatty acids remained intact. We do not have any data to show whether polyunsaturated fatty acids also remained intact in the other types of samples, particularly those that were salt-incubated for longer periods. Furthermore, there are no published data concerning the fatty acid composition of the species used in the preparation of the condiments studied, which would have provided a point of reference. It is possible that the high salt concentration contributed to the protective effect by the inhibition of oxygenase-type enzymes. It is also known that the rate of auto-oxidation of lipids diminishes as water activity drops to about 0.4 (Karel & Yong, 1981). It would be interesting, in future analyses, to measure antioxidant activity and oxygen tension in these preparations, to see whether they are also determining factors in the levels of polyunsaturated fatty acids in the final product.

In conclusion, all the fish and shrimp condiments analyzed contained measurable quantities of polyunsaturated fatty acids, including DHA. Alamang represents the highest source of DHA among the samples. Padas-1, because it has the highest fat content, has DHA levels that approach that of alamang. The question of whether alamang or padas-1 can be a source of dietary DHA can be answered only in conjunction with data on consumption patterns for these particular condiments.

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