

## Development of a fish emulsion product (Pla Yor)

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**Abstract:** The objectives of this study were to select type of fish for developing fish emulsion product (Pla Yor) and to study the quality of the product kept at 5±2°C for 21 days. Three types of locally economic-driven fresh water fish, namely *Oreochromis niloticus* × *mossambicus* (a hybrid tilapia, ONM), *Oreochromis niloticus* (ON), and *Clarias batrachus* (CB), were selected for recipe optimisation. The fish fillets had L, a, and b values ranging from 51.85-58.45, 0.62-11.3 and 10.68-22.85, respectively. They had pH 6.26-6.71, 73.39-75.80% moisture, 1.05-1.89% fat, and 15.22-25.08 % protein. The type of fish and recipe were optimized to produce fish emulsion products and subjected to sensory evaluation by fifteen trained panelists using quantitative descriptive analysis approach and ratio profile test. The most preferred product included 72.10% mixed CB-ONM (1:1 weight basis), 10.82% ground lard, 8.65% crushed ice, 4.00% mixed garlic-pepper (1:1 weight basis), 2.88% egg, 1.00% salt, 0.37% glutinous flour, 0.18% phosphate. It was also found that the product did not significantly change ( $p>0.05$ ) in moisture, protein, fat, and microbial numbers during storage at 5±2°C for 21 days.

**Keywords:** Fish emulsion, Pla Yor, product development, *Oreochromis niloticus* × *mossambicus*, *Oreochromis niloticus*, *Clarias batrachus*

### Introduction

Oil-in-water pork emulsion product (Moo Yor) is a popular food in the North and North-eastern regions of Thailand. Conventionally, the product is made by mixing pork, lard, spice, and ice to obtain a homogenous mixture before tightly wrapping it in banana leaves forming cylindrical shapes (approx. 2-3 inches in diameter and 6-8 inches in length). The product is then steamed or boiled until the mix is cooked through (TISI, 2004). However, pork can be expensive, because of high consumer demand, or avoided, when outbreaks of diseases such as swine flu occur. Therefore, alternative, healthier, more cost-effective raw materials are highly sought after.

Freshwater fish is one of the most important, economically, agricultural products in Thailand. Freshwater fish farming has been promoted by many government bodies. The production of freshwater fish increased from approximately 377,000 tonnes in 1995 to 513,000 tonnes in 2005 (FAO, 2009). Fish in the genus *Siluridae*, *Cyprinidae*, *Clariidae*, *Ophicephalidae*, and *Cichlidae* are widely grown and cultivated (NICA, 2007). Fish utilization has been explored and many fish products have been developed. Fish emulsion (Pla Yor) is a product imitating pork emulsion. It was recently introduced to the Thai market and was accepted especially in the healthy food sector. White-flesh fish is normally used since the color of the finished product would

resemble the one made from pork. Only a few studies related to such product have been reported. Those studies included using hydrocolloids as fat replacer (Yamprayoon and Vicharnnikornkich, 2005), enhancing flavor by addition of herbs (Sukjuntra, 2006), and textural improvement using microbial transglutaminase (Long *et al.*, 2008).

However, a comparison study on the production of Pla Yor using different type of fish has never been reported and recipe optimization is under explored. This study aimed to identify the most suitable type of fish, using three fish species which are of economic importance, as well as optimize the recipe for Pla Yor production. Furthermore, chemical composition and microbiological changes during the product's storage in a refrigerator for 21 days were also assessed.

### Materials and Methods

#### Materials

All ingredients – fish, pork lard, ground garlic, ground pepper, salt, and eggs – were purchased from a local market in Loei Province, Thailand. All chemicals were obtained from Merck (Bangkok, Thailand) unless otherwise stated. All culture media were obtained from Difco (Bangkok, Thailand).

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## Methods

### Determination of quality attributes of fish

Fillets of three varieties of fish, namely *Oreochromis niloticus* × *mossambicus* (a hybrid tilapia, ONM), *Oreochromis niloticus* (ON), and *Clarias batrachus* (CB), were used. Fillet color was measured using a Hunter MiniScan XE, (Hunter Lab, Bangkok, Thailand) and fillet pH was measured using a CyberScan pH300 pH meter, (Eutech Instruments, Singapore). Chemical composition, (protein, fat and moisture) of the fillets was determined according to AOAC (1990).

### Optimization of type of fish

Fish were cleaned by rinsing in tap water. Fillets were separated and coarsely ground in a food processor. Three samples of fish emulsion were produced using a recipe, modified from typical recipe of Department of Fisheries, Minister of Agriculture and Cooperatives, Thailand (DoF, 2009). The ingredients included 750 g coarsely ground fish, 50 g ground lard, 20 g salt, 12 g ground pepper, 12 g ground garlic, 5 g glutinous flour, 1 egg, 2 g sodium triphosphate, and 30 g crushed ice. All ingredients were mixed in a bowl chopper while ice was gradually added into the mix. The mixture was blended for approximately 15 min until a homogenous mass was obtained. The emulsion mix was then transferred into bamboo containers (7-8 cm diameter, 15cm length). Both ends of the bamboo were closed with double sheets of aluminium foil and tightly tied with strings. The products were cooked in a steamer at 100°C for 45 min. Color values of the cooked products were measured as mentioned above.

All three samples of fish emulsion were subjected to sensory evaluation by 15 trained panelists using quantitative descriptive analysis (QDA), (Lawless and Heyman, 1999). Prior to the evaluation sessions, training sessions were conducted and a typical, commercial available fish emulsion was introduced to the panelists. They familiarized themselves to the nature of the product and made agreement upon the testing attributes – including color, size of air cells, fishy odor, spice odor, firmness, elasticity, taste, and overall liking. When evaluating the samples, panelists were asked to mark ideal score (I) and sample score (S) on a 10-cm scale line for each attribute according to their opinions. Ratio profile test (RPT), (Sintawalai, 1998) was performed to obtain the characteristics outline of the product, which was subsequently used as reference points for recipe optimization.

### Recipe optimization

The most accepted fish was selected to make the product and to be used for recipe optimization. The amounts of ingredients were adjusted according to the profile of the ideal product. The optimized recipe samples were analyzed for their color values (as described above), and firmness (using a Koehler K19500 penetrometer, (ALPE, Bangor, NSW, Australia)). Sensory evaluation (QDA) by 15 trained panelists was performed in the same manner as described earlier. Ratio profile test of each attribute and analysis of variance among sample means was subsequently carried out using randomized complete block design (RCBD) (O'Mahony, 1985). The mean difference between S/I and I/I ratio of each attribute was analysed using t-test (O'Mahony, 1985) and displayed on a spider chart. Recipe optimization was continued until there was no statistical difference between the mean values of S/I and I/I ratio of all attribute.

### Monitoring on quality changes during storage

The sample made from the optimized recipe was kept at 5±2°C for 3 weeks. Quality changes were monitored every 3 days. The product was measured for its color value, firmness, as well as protein, fat, and moisture content. Microbiological analysis, such as, total bacteria, *Salmonella* sp., *Escherichia coli*, and *Staphylococcus aureus* (AOAC, 1990), were also performed.

## Results and Discussion

### Physical and chemical properties of *Oreochromis niloticus* × *mossambicus*, *Oreochromis niloticus*, and *Clarias batrachus*

Three varieties of fish, including *Oreochromis niloticus* × *mossambicus* (ONM), *Oreochromis niloticus* (ON), and *Clarias batrachus* (CB), were selected for this study because of their significant importance to the local economy. In Table 1 the results of some physical and chemical analyses of those fish are shown.

The three fish had a pH range of 6.26-6.71 which was in the pH range 6-8 of fresh-water fish in general (Benjakul, 2005). They were categorized as low-fat fish because the fat contents were lower than 2% (Thongrueng, 2001). Statistical analysis showed that the color of the three fish were different ( $p \leq 0.05$ ). ONM fillet was whiter (L value) while CB fillet was redder (a value) and yellower (b value). The three fish were not different in moisture and fat content ( $p > 0.5$ ). However, CB contained significantly higher

protein than ONM.

**Table 1.** Analysis results<sup>1</sup> on some of physical and chemical properties of *Oreochromis niloticus* × *mossambicus* (ONM), *Oreochromis niloticus* (ON), and *Clarias batrachus* (CB)

Type of Fish	pH*	Color*		Moisture (%)	Fat (%)	Protein* (%)	
		L	a				b
<b>ONM</b>	6.71 ±0.02 <sup>a</sup> 6.39	58.45 ±0.14 <sup>a</sup> 56.56	0.062 ±0.02 <sup>a</sup> 4.73	10.68 ±0.03 <sup>a</sup> 15.47	75.80 ±1.40 73.39	1.89 ±0.99 1.43	15.22 ±2.41 <sup>b</sup> 20.77
<b>ON</b>	±0.01 <sup>b</sup> 6.26	±0.04 <sup>b</sup> 51.85	±0.03 <sup>b</sup> 11.30	±0.02 <sup>b</sup> 22.85	±1.97 75.33	±0.58 1.05	±2.29 <sup>ab</sup> 25.08
<b>CB</b>	±0.00 <sup>c</sup>	±0.07 <sup>c</sup>	±0.11 <sup>a</sup>	±0.03 <sup>a</sup>	±0.75	±0.22	±5.64 <sup>a</sup>

<sup>1</sup> Values were mean ± standard deviation of 3 replicates

\* Means with different letters within each column differed significantly (p≤0.05)

**Table 2.** Analysis results<sup>1</sup> on moisture content, firmness, and color of fish emulsion products made from *Oreochromis niloticus* × *mossambicus* (ONM), *Oreochromis niloticus* (ON), and *Clarias batrachus* (CB)

Type of Fish	Moisture (%)	Firmness* (N)	Color Value		
			L	a	b
<b>ONM</b>	72.12 ±7.11	0.30 ±0.03 <sup>b</sup>	74.14 ±0.00	-0.25 ±0.02	15.63 ±0.02
<b>ON</b>	68.62 ±4.53	0.34 ±0.00 <sup>a</sup>	69.84 ±0.00	0.02 ±0.01	17.12 ±0.02
<b>CB</b>	66.96 ±3.11	0.32 ±0.00 <sup>ab</sup>	68.16 ±0.04	1.00 ±0.03	18.59 ±0.07

<sup>1</sup> Values were mean ± standard deviation of 3 replicates

\* Means with different letters within each column differed significantly (p≤0.05)

**Table 3.** Sensory evaluation of emulsion fish products using from *Oreochromis niloticus* × *mossambicus* (ONM), *Oreochromis niloticus* (ON), and *Clarias batrachus* (CB)

Raw Material	Color*	Size of Air cells	Sample-to-ideal score ratio <sup>1</sup>					Overall Acceptance
			Fishy Odor	Spice Odor	Firmness	Elasticity	Taste	
<b>ONM</b>	0.84 ±0.39 <sup>b</sup>	1.32 ±0.52	1.62 ±0.88	0.65 ±0.23	0.85 ±0.22	1.04 ±0.16	0.88 ±0.30	0.91 ±0.23
<b>ON</b>	1.01 ±0.16 <sup>ab</sup>	1.71 ±0.76	1.69 ±0.84	0.72 ±0.27	0.80 ±0.29	0.96 ±0.24	0.92 ±0.24	0.93 ±0.15
<b>CB</b>	1.19 ±0.28 <sup>a</sup>	1.65 ±0.06	1.42 ±0.84	0.71 ±0.26	0.92 ±0.16	0.88 ±0.11	0.83 ±0.28	0.98 ±0.20

<sup>1</sup> Values were means of sample-to-ideal (S/I) score ratios ± standard deviation of 15 panelists

\* Means with different letters within each column differed significantly (p≤0.05)

### Selection of type of fish

Three fish emulsion samples using a typical recipe and the three fish varieties were produced. The finished samples were analyzed for their color values, firmness, and moisture contents (Table 2). It was found that the ON and CB sample were firmer than the ONM sample (p > 0.05). This could be because of their high protein content (Table 1). Proteins play significant role on emulsion forming in which they inhibit coalescence of fat globules and help binding water and fat (Hongprapas, 2002). The color of the samples was also different (p>0.05) according to the color values of the fish used to make the product.

The three fish emulsion samples were subjected to sensory evaluation by 15 trained panelists using the QDA approach. The closeness of S/I ratio of each attribute to 1 indicated that the sample met the expectation of the panelists on that particular attribute. From the results, the panelists pointed out that the CB sample was the darkest color which corresponded to the color of the fish (Table 1). However, the panelists did not find any difference between products on other attributes (p>0.05). However, the CB sample got S/I ratios on firmness and fishy odor, which were important characteristics of the product, closer

to ideal score than the other two samples. These as well reflected on the highest S/I ratio on overall acceptance. Regarding the acceptance on fishy odor, the CB sample's S/I ratio was the closest to I/I ratio (equals to 1). The less unpleasant fishy odor of the CB sample might be because the CB fillet had lower pH (6.26, Table 1) than the ONM and ON fillets. Therefore, lower amount of trimethylamine oxide in the flesh was hydrolyzed to volatile basic compounds (Benjakul, 2006). With regards to the cost of the three fish, the CB was the most economical (THB45/kg (whole fish), in 2008) than the ON (THB55/kg) and the ONM (THB65/kg). Therefore, based on those attributes, CB was selected for subsequent experiments on recipe optimization.

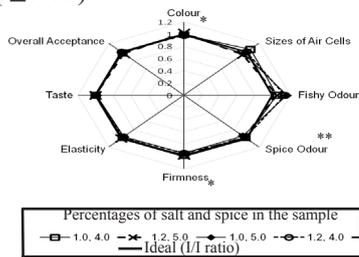
### Optimization of product's recipe

Recipe was optimized to improve color, spice odor, and taste of the CB sample according to the products' outline (Table 3). The CB sample was darker than the preferred product (S/I>1), therefore, four trials were made to lighten the color by mixing glutinous flour and CB (5:95, 10:90, and 15:85), and mixing glutinous flour, ONM and CB (5:47.5:47.5). The finished products were measured for their color values (Table 4). It was found that the product made from mixed glutinous flour-CB-ONM was significantly brighter than the other products (p≤0.05). Therefore, the mixed glutinous flour-CB-ONM was used in the next step.

The amounts of salt and spice (mixed garlic-pepper, 1:1 weight basis) were then adjusted to comply with the product characteristic outline. Four recipes, varying two concentrations of salt (1 and 1.2%) in combination with two concentrations of mixed garlic-pepper (4 and 5%), were used to make fish emulsion products. Table 5 illustrated firmness and color value of the four trials. It was found that the sample contained high amount of salt and spice (1.2% and 5%, respectively) was significantly firmer than the other samples (Table 5). This could be because salt helped dissolving myofibrils from the fish, in which they improved emulsion stability (Siripin, 2003). Furthermore, during the thermal cooking process, gelatinization of myofibrils might take place, which has been shown to enhance the firmness of the sample (Hongprapas, 2002). However, the high amount of salt and spice also resulted in darker color of the product.

Sensory evaluation of the samples showed that the mean S/I ratios of the four samples differed only on color and overall acceptance (p≤0.05, Table 6). When comparing the mean S/I ratio of each attribute to I/I ratio using t-test (Figure 1), it was found that

the samples with combination of 1.0, 5.0 and 1.2, 4.0 (salt, mixed garlic-pepper) did not meet the expectations of the panelists on spice odor, color, and firmness ( $p \leq 0.05$ ).



**Figure 1.** Acceptance profiles of fish emulsion samples made from mixed CB-ONM (1:1 weight basis) and different combination of salt and spice (mixed garlic-pepper, 1:1 weight basis) on each attribute in comparison with panelists' expectations (\* indicates the 1.0, 5.0 sample significantly differed from ideal, and \*\* indicates the 1.2, 4.0 sample significantly differed from ideal at  $p \leq 0.05$ )

**Table 4.** Color value of fish emulsion products using mixed CB-glutinous flour and mixed CB-ONM

Fish Mix	Color value <sup>1*</sup>		
	L	a	b
<b>Glutinous Flour:CB (5:95)</b>	67.69±0.33 <sup>b</sup>	0.94±0.12 <sup>a</sup>	17.17±0.12 <sup>b</sup>
<b>Glutinous Flour:CB (10:90)</b>	67.58±0.06 <sup>b</sup>	1.11±0.23 <sup>a</sup>	18.20±0.13 <sup>a</sup>
<b>Glutinous Flour:CB (15:85)</b>	68.64±0.85 <sup>b</sup>	0.91±0.49 <sup>a</sup>	17.34±0.02 <sup>b</sup>
<b>Glutinous Flour:ONM:CB (5:47.5:47.5)</b>	72.86±0.20 <sup>a</sup>	-0.59±0.04 <sup>b</sup>	17.00±0.36 <sup>b</sup>

<sup>1</sup> Values were mean ± standard deviation of 3 replicates.  
<sup>2</sup> Mixed garlic and pepper, 1:1 weight basis  
<sup>\*</sup> Means with different letters within each column differed significantly ( $p \leq 0.05$ ).

**Table 5.** Analysis results<sup>1</sup> of firmness and color value of fish emulsion made from different concentrations of salt and spice<sup>2</sup>

Salt and spice content in the sample (%)	Firmness* (N)	Color Value*		
		L	a	b
1.0, 4.0	0.20±0.03 <sup>b</sup>	64.93±1.44 <sup>a</sup>	0.82±0.33 <sup>a</sup>	17.43±0.70
1.2, 5.0	0.28±0.00 <sup>a</sup>	62.63±0.41 <sup>b</sup>	1.04±0.24 <sup>ab</sup>	17.92±0.10
1.0, 5.0	0.20±0.05 <sup>b</sup>	64.82±0.52 <sup>a</sup>	0.45±0.09 <sup>ac</sup>	17.84±0.27
1.2, 4.0	0.19±0.04 <sup>b</sup>	64.64±0.43 <sup>a</sup>	0.47±0.14 <sup>ab</sup>	17.26±0.18

<sup>1</sup> Values were mean ± standard deviation of 3 replicates  
<sup>2</sup> Mixed garlic and pepper, 1:1 weight basis  
<sup>\*</sup> Means with different letters within each column differed significantly ( $p \leq 0.05$ )

However, the samples with combination of 1.0, 4.0 and 1.2, 5.0 (salt, mixed garlic-pepper) did not differ from ideal scores for the product, which means those two samples satisfy the panelists' expectations on all agreed attributes. With regards to the mean S/I ratios of overall acceptance of the two samples on Table 6, it was found that the sample with the combination of 1.0, 4.0 (salt, mixed garlic-pepper) got a significantly higher score than that of the sample with the combination of 1.2, 5.0 (salt, mixed garlic-pepper). Therefore, the fish emulsion produced from mixed CB-ONM (1:1 weight basis) with 1.2% salt and 4% mixed garlic-pepper (1:1 weight basis) was selected as the most preferred recipe of the product and subsequently used for monitoring changes during storage.

*Monitoring changes of the developed fish emulsion product during storage*

The developed product was kept at  $5 \pm 2^\circ\text{C}$  for 3 weeks. Microbial changes of the product were monitored by taking samples every 3 days to check total bacteria count, *E. coli*, *S. aureus*, and *Salmonella* spp.. Chemical analyses were carried out on days 0 and 21.

After 21 days of storage at  $5 \pm 2^\circ\text{C}$ , it was found that the product did not significantly change on chemical composition and microbiological profile ( $p > 0.05$ ). However, the product slightly lost its moisture ( $61.35 \pm 0.13\%$  from the initial  $64.49 \pm 1.24\%$ ) and protein contents ( $15.51 \pm 0.38\%$  from the initial  $16.36 \pm 0.21\%$ ) after 21 days of storage. There were no differences observed for fat content of the product.

**Table 6.** Sensory evaluation<sup>1</sup> of fish emulsion samples produced from different concentrations of salt and spice<sup>2</sup>

Salt and spice content in the sample (%)	Sample-to-ideal score ratio							Overall Acceptance
	Color*	Size of Air cells	Fishy Odor	Spice Odor	Firmness	Elasticity	Taste	
1.0, 4.0	0.99 ±0.20 <sup>a</sup>	1.06 ±0.25	1.05 ±0.32	0.98 ±0.07	0.98 ±0.04	0.98 ±0.03	1.00 ±0.03	0.98 ±0.02 <sup>ab</sup>
1.2, 5.0	1.02 ±0.20 <sup>ab</sup>	0.94 ±0.19	1.05 ±0.15	0.97 ±0.04	0.98 ±0.08	1.00 ±0.03	1.00 ±0.05	0.96 ±0.07 <sup>b</sup>
1.0, 5.0	0.99 ±0.18 <sup>a</sup>	0.96 ±0.17	1.16 ±0.62	0.94 ±0.17	0.95 ±0.09	0.96 ±0.09	0.99 ±0.09	0.99 ±0.07 <sup>ab</sup>
1.2, 4.0	0.98 ±0.20 <sup>ac</sup>	1.00 ±0.16	1.14 ±0.44	0.98 ±0.04	0.95 ±0.06	0.97 ±0.09	1.00 ±0.05	1.00 ±0.04 <sup>a</sup>

<sup>1</sup> Values were means of sample-to-ideal (S/I) score ratios ± standard deviation of 15 panelists  
<sup>2</sup> Mixed garlic and pepper, 1:1 weight basis  
<sup>\*</sup> Means with different letters within each column differed significantly ( $p \leq 0.05$ )

With regards to the microbiological analysis results, no pathogenic bacteria were detected and total bacteria count of the 21-day old product was still within microbial safety standard specifications for fish emulsion product (less than 104 CFU/g, (TISI, 2004)).

**Conclusions**

Fish emulsion product (Pla Yor) was developed. The recipe for the most preferred product included 36% ground CB, 36% ground ONM, 10.8% ground lard, 8.6% crushed ice, 3% egg, 2% ground garlic, 2% ground pepper, 1% salt, 0.4% glutinous flour, 0.2% phosphate. Upon storage at  $5 \pm 2^\circ\text{C}$  for 21 days, it was found that the product did not significantly change in moisture, protein, fat, and microbial numbers ( $p > 0.05$ ). Small scale fish-farmers are expected to benefit from this newly established recipe, as it will provide a basis for a cheaper alternative to a popular pork based product.

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