# Study on the Species Composition and Abundance of Plankton, Water Quality and Stomach Contents of Pacific White Shrimp (*Litopenaeus vannamei*Boone,1931) Reared in Low Salinity Conditions

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### Abstract

This study analyzed the species composition and abundance of plankton and water quality in low salinity (2-3 parts per thousand (ppt)) ponds used for rearing Pacific white shrimp, Litopenaeus vannamei. A total of 62 genera of plankton were found from six groups: 12 genera in Division Cyanophyta, 28 genera in Division Chlorophyta, 10 genera in Division Chromophyta, two genera in Phylum Protozoa, eight genera in Phylum Rotifera and two genera in Phylum Arthropoda. The phytoplankton that were the most prolific were the diatoms, Cyclotella and Nitzschia. The most common blue-green algae were Merismopedia, Oscillatoria and Phormidium. The most common green algae were Dictyosphaerium, Oocystis and Scenedesmus. The numbers of plankton found near the bottoms of the ponds were not different from the numbers found near the water surface. The numbers of plankton were positively related to the dissolved oxygen (DO) and chlorophyll a contents in the water.

The plankton found in the stomachs and guts of the Pacific white shrimp consisted of seven genera in Division Cyanophyta, 19 genera in Division Chlorophyta, 10 genera in Division Chromophyta, one genus in Phylum Protozoa and two genera in Phylum Rotifera. The plankton found in both the stomachs and the guts were *Merismopedia*, *Oscillatoria*, *Phormidium*, *Scenedesmus* and *Oocystis* which were the same as the dominant species found in the water.

Keywords: Pacific white shrimp (Litopenaeus vannamei), plankton, stomach contents, water quality

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#### Introduction

Pacific white shrimp (*Litopenaeus vannamei*) are native to South America. They are raised commercially in Ecuador, Mexico, Peru, Panama, Honduras, Colombia, Brazil and other countries (Rosenberry, 1993; FAO, 1994). The Pacific white shrimp inhabits waters that range in salinity from 1 to 40 ppt (Bray *et al.*, 1994; Smith and Lawrence, 1990; Samocha *et al.*, 2001). Pacific white shrimp were first introduced to Asia in 1996 by farmers in Taiwan; they were introduced to China in 1998. Because of problems with raising black tiger shrimp that led to low harvests in Thailand, in 1998 some Pacific white shrimp were imported for testing. The first trials were not very successful. However, during March 2002 to February 2003 the Department of Fisheries, Ministry of Agriculture and Cooperatives the import of specific pathogen-free (SPF) broodstock of Pacific white shrimp for further

testing. During that period, farmers raising black tiger shrimp in Thailand experienced more serious problems with slow growth and a wide disparity in the sizes of shrimp at harvest time, with many shrimp growing to only 3-5 g. This was a problem that had been occurring for several years (Limsuwan, 1999). As a result some farmers decided to switch to raising Pacific white shrimp. Most of them found that the results were better than with black tiger shrimp. Subsequently, more shrimp farmers all over the country began raising Pacific white shrimp. The shrimp larvae that came from SPF broodstock grew quickly and were of a uniform size. They could be harvested and sold after rearing for just 90-100 days and the harvests were larger than those for black tiger shrimp.

The feeding habits and other behavior characteristics of white shrimp are quite different from those of black tiger shrimp. White shrimp are very active and keep swimming about, as well as eating almost constantly. This makes it quite difficult to feed them using the feeding tray method as is commonly done with black tiger shrimp. With white shrimp there is almost always some food left over in the feeding tray, but if one inspects the shrimps' digestive tracts they are almost always full of food. If the amount of prepared food that is provided is insufficient, the white shrimp will eat other things that they find in the pond. The food undergoing digestion in their intestines appears black, which is not the color of the pelleted food. To date, there has not been a study made of the contents of the digestive tracts of these shrimp to find out exactly what they eat besides the pelleted food. It might be animal organisms or phytoplankton found in the water.

In the rearing of all types of penaeid shrimp the farmer must try to control the water quality to keep it within the optimum range so the shrimp will be healthy and grow quickly (Chen, 1993; Philips *et al*, 1993). In particular, farmers pay attention to the amount of plankton in the water. In Thailand, the farmers refer to the plankton level as the "color" of the water. If the color remains constant, the shrimp will grow well. In fact, the populations of plankton in the pond vary all the time as they are affected by other factors such as the amount of food, the amount of sunlight, the temperature, pH and salinity of the water (Limsuwan, 2000).

The aim of this study is to determine the relationship between water quality and the species composition and abundance of plankton in the pond and the species composition of plankton in the stomach contents of Pacific white shrimp and their growth rate. The results will be beneficial, both as a source of basic data and in formulating guidelines for the successful commercial rearing of Pacific white shrimp.

#### Materials and Methods

The study was undertaken at a shrimp farm in Baan Sarng District, Prachinburi Province, Thailand. The study used four shrimp rearing ponds sized 4,448 m², 4,416 m², 4,208 m² and 4,096 m². There were four reservoirs of similar size, one for each rearing pond. A water recirculation system was used, changing water from the rearing ponds to the reservoirs. The ponds were 1.2-1.5 m deep. The soil type was sandy clay. To start the shrimp rearing process the shrimp farmer obtained fresh water from an irrigation canal and mixed it with concentrated (150-200 ppt) salt water from a paddy salt farm in a 150 m² plastic trough until the salinity was adjusted down to 8-10 ppt. The water outside the trough was diluted to just 3 ppt.

Postlarvae (PL) stage 12, that had been acclimated to a salinity level of 10 ppt and confirmed negative for the white spot syndrome virus (WSSV), Taura syndrome virus (TSV) and infectious hypodermal and hematopoietic necrosis virus (IHHNV) infections by the polymerase chain reaction (PCR) assay, were released into the plastic trough at a rate of 60 PL per square meter.

<u>Feeding program</u> Commercial feed pellets for Pacific white shrimp were fed four times daily at 07.00 a.m., 11.00 a.m., 04.00 p.m. and 10.00 p.m.

Water changing The water was not changed for the first 40 days, but some water from the reservoirs was added at intervals to replace water evaporation or soil seepage. The water was changed for the first time when the shrimp were 60 days old. All the water that was pumped out of the rearing pond was discharged back into the reservoirs. None was released outside the farm.

#### The species composition and abundance of plankton in ponds

Water samples were collected from the dock where the feeding tray was cleaned at two positions: 30 cm down from the surface and 30 cm up from the pond bottom. The surface samples were taken with a plastic bucket, for a volume of 8 liter per sample. The bottom samples were taken with a cylindrical container fitted with a plankton net with a mesh size of 20 microns. Each sample was 1 liter. The plankton samples were preserved in a 4% formalin solution and stored in 135-ml bottles. The plankton were identified and counted using Wongrat's method (Wongrat 1998, 1999) The water samples for measuring water quality were taken from the same area as the samples for observing the plankton.

#### Water quality analysis

Temperature (mercury thermometer), pH (ORION Model Sa520 pH meter), and dissolved oxygen (DO; YSI Model 51B DO meter) were measured daily at 06.00 a.m. and 01.00 p.m.. Measurements of the other water quality parameters were analyzed weekly from the time the PL were stocked to the time the shrimp were harvested, included salinity and electrical conductivity (YSI 30/10 FT), total alkalinity (titration method; APHA, 1995), hardness (EDTA titrimetric method; APHA, 1995), total ammonia nitrogen (TAN; phenolhypochloride method; APHA, 1995), nitrite nitrogen (colorimetric method; APHA, 1995), total suspended solid (TSS) and chlorophyll a (APHA, 1995).

#### Analysis of the stomach and intestine contents of Pacific white shrimp

The white shrimp that were used for the stomach and intestine contents study were reared on a intensive farm and were fed with commercial pelleted food throughout the culture period. For the convenience of analysis, the sample shrimp were ones that had been fed some time ago and were ready to be fed again. Thus they were sampled at 10:30-10:45, just before the 11:00 feeding.

After the shrimp were 30 days old, five shrimp were randomly selected as samples from each of the four ponds. Samples were then taken every two weeks until the shrimp were harvested. The sample shrimp were preserved in a 10% formalin solution. The stomachs and intestines were removed and crushed, then placed on slides to be examined under a high-resolution light microscope. The researchers observed and noted the species of plankton found.

#### **Results and Discussion**

The yields from the four test ponds ranged from 578-1,053 kg/1,600 m². The survival rate was 62-81%. These results are typical for white shrimp reared in low salinity conditions. However, the average weight of the shrimp in all four ponds was rather low at 14-16 g after 123-124 days. Usually the average weight is 20-25 g. The main reason for the small size of the shrimp was low temperatures because the study was undertaken in the cool season. In January and February the water temperature dropped to below 27 °C for several days and on some days it was as low as 23 °C. When the temperature is less than 27 °C the shrimp eat less

(Wyban et al., 1995; Villarreal et al. 1994). Normally farmers feed the shrimp 10% less for every 1-degree drop in temperature, but if the temperature drops to 24 °C they will reduce the amount by 50% and if it is lower than 23 °C they will suspend feeding for at least one meal time and feed again when the temperature rises in the afternoon.

Table 1 Data on raising Pacific white shrimp in low saline water

Parameter	Pond 1	Pond 2	Pond 3	Pond 4
Stocking date	24/10/2545	24/10/2545	24/10/2545	24/10/2545
Larval stage	PL12	PL12	PL12	PL12
Pond area (square meters)	4,448	4,416	4,096	4,208
Stocking density (shrimp/m²)	60	60	60	60
Harvest date	24/2/2546	24/2/2546	25/2/2546	25/2/2546
Culture period time (days)	123	123	124	124
Average weight (g)	16.39	14.08	15.15	14.71
Average size (shrimp/kg)	61	71	66	68
Yield (kg)	2,715.34	3,023.86	2,454.84	3,024.66
Yeild (shrimp)	165,671	214,763	162,036	205,620
Amount fed (kg)	4,751.85	4,989.37	4,173.23	4,869.70
Feed conversion rate	1.75	1.65	1.70	1.61
Survival rate (%)	62.08	81.05	65.93	81.44
Weight gain /day (g)	0.13	0.11	0.12	0.12
Production (kg/1,600 m <sup>2</sup> )	745.31	998.18	578.92	1,053.06

1. Species composition and abundance of plankton in ponds used to raise Pacific white shrimp

The species and numbers of plankton in the four test ponds were studied throughout the culture period. Fifty genera of plant plankton from three divisions (12 genera of Cyanophyta, 28 genera of Chlorophyta and 10 genera of Chromophyta), along with 12 genera of zooplankton from three phyla (two genera of Protozoa, eight genera of Rotifera and two genera of Arthropoda) were found.

The most abundant plant plankton found throughout the shrimp rearing period were diatoms, i.e. *Cyclotella* and *Nitzschia*, blue-green algae such as *Merismopedia*, *Oscillatoria*, and *Phormidium*; and green algae, especially *Dictyosphaerium*, *Oocystis* and *Scenedesmus*.

Table 2 Average numbers of plankton on the water surface and near the pond bottom

	Sample spot	Mean	Standard deviation	t-test for Equality of Means		
Month				t	df	Sig.(2-tailed)
1	Surface	361,446.75	195,088.87			
	Pond bottom	631,430.13	200,989.68	-3.856	30	P>0.05
2	Surface	1,810,141.25	2,515,651.23			
	Pond bottom	2,210,640.38	2,293,728.37	-0.471	30	P>0.05
3	Surface	1,426,937.50	702,623.40			
	Pond bottom	2,785,514.06	1,051,801.10	-4.296	30	P>0.05
4	Surface	3,676,398.35	3,556,163.15			17 0.05
	Pond bottom	5,488,315.35	3,911,462.66	-1.533	38	P>0.05

The numbers of plankton both in the samples taken near the water surface and near the bottom of the pond increased throughout the culture period. The lowest numbers were found in the very first samples taken and the highest numbers in the last samples.

In the beginning of the shrimp rearing period, the water appeared brown and the most abundant plankton found were diatoms. The color of the water changed as the shrimp grew and the types of plankton that were found in large numbers changed as well. When the water appeared brown the greatest number of plankton were the diatoms of genera *Cyclotella*, *Entomoneis* and *Nitzschia*. When the water changed to a more green color, the most abundant kinds of plankton were blue-green algae of genera such as *Merismopedia*, *Oscillatoria*, *Phormidium* and *Scenedesmus*. Blue-green algae were abundant throughout the duration of the experiment. The water color and types of plankton are summarized in Table 3 (Figure 1). Wongrat (1987) stated that the color of plankton depends on the amount of pigment in their cells. Plankton of the phylum Cyanophyta contain more chlorophyll and phycobilin than other pigments so they appear blue-green or green. Plankton in the phyla Bacillariophyta and Pyrrophyta contain more carotene and xanthophyll so they appear yellow or brown. The color of the water comes from the pigments found in the cells of the most abundant plant plankton.

**Table 3** Relationship between water color and species of plankton

Figure	Water color	Most abundant plankton		
1	Clear brown	Cyclotella, Nitzschia		
2	Yellowish brown	Scenedesmus, Nitzschia, Phormidium, Kirchneriella		
3	Dark brown	Entomoneis, Merismopedia, Oscillatoria, Scenedesmus		
4	Light green	Merismopedia, Dictyosphaerium, Phormidium		
5	Yellowish green	Merismopedia, Nitzschia		
6	Bright green	Scenedesmus, Spirulina, Oocystis		



Figure 1 Relationship between water color and species of plankton during the shrimp rearing period

6) Water with large numbers of Scenedesmus

# 2. Water quality in the ponds

5) Water with large numbers of Merismopedia

The water quality parameters that were measured and analyzed were: salinity, EC, pH, DO, TAN, nitrite, total alkalinity, hardness, TSS, and chlorophyll a, as shown in Table 4.

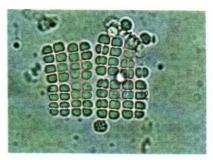
Table 4 Highest and lowest readings for water quality throughout the shrimp culture period

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Water quality parameter	Pond 1	Pond 2	Pond 3	Pond 4	Mean
1.salinity (ppt)	2.3-3.2	2.4-3.4	2.3-3.3	1.9-3.1	2.66±0.35
2.electrical conductivity (mmhos/cm)	4.35-5.67	4.47-5.79	4.44-5.75	3.72-5.69	4.95±0.53
3.temperature (°C)					
- morning	23.0-29.0	23.5-29.5	23.0-29.0	23.0-29.0	26.46±1.88
- afternoon	25.0-31.0	25.0-31.0	25.0-31.5	25.0-31.0	28.58±1.74
4.pH					
- morning	7.3-8.2	7.3-8.3	7.3-8.2	7.3-8.2	$7.79 \pm 0.23$
- afternoon	7.9-8.8	7.6-8.5	8.2-8.8	8.2-8.8	8.37±0.15
5.dissolved oxygen (mg/l)					
- morning	5.0-7.5	5.0-7.0	5.0-7.5	5.0-7.0	6.13±0.62
- afternoon	7.5-12.0	6.5-12.0	7.5-11.0	7.5-11.5	9.32±1.27
6.Total ammonia nitrogen (mg/l)	0.05-1.5	0.04-1	0.02-0.4	0.05-1.5	0.30±0.34
7.nitrite nitrogen (mg/l)	0.01-0.15	0.01-0.17	0-0.15	0-1.2	0.10±0.17
8.total alkalinity (mg/l)	119-198	87-180	82-160	103-176	140.85 <u>+</u> 22.77
9.hardness (mg/l)	594-1282	641-1064	554-946	585-768	741.28±118.25
10.total suspended solids (mg/l)	16-158	38-224	24-130	17-266	112.79 <u>+</u> 60.81
11.cholorphyll a (mg/m²)	44.49-552.2	20.23-408.37	8.2-434.46	8.96-419.15	210.16±129.06

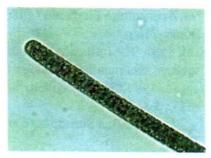
#### 3. The stomach and intestine contents of Pacific white shrimp

The types of plankton found in the stomachs and intestines of the shrimp were not different. This shows that Pacific white shrimp do not selectively eat specific types of plankton. They eat any kinds of plankton that are abundant in the water. The most common types of plankton found in the stomachs and intestines of the test shrimp were of the genera *Merismopedia*, *Oscillatoria*, *Phormidium*, and *Scenedesmus*. These were the same types of plankton that were most abundant in the ponds, both from the surface samples and the pond bottom samples, throughout the culture period. The most common genera of plankton found in the water were *Cyclotella*, *Nitzschia*, *Merismopedia*, *Oscillatoria*, *Phormidium*, *Dictyosphaerium* and *Scenedesmus*.

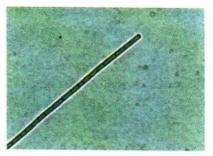
Pacific white shrimp eat plankton throughout their life spans, not only when they are young. Wassenberg and Hill (1987) stated that Pacific white shrimp can eat plants, animals and dead organisms found in the water. They can also eat living things and the remains of dead organisms found on the surface of the ocean floor. Decapod crustaceans are usually opportunistic omnivores, taking their food from the bottom of their habitats or from the fauna associated with submersed and shore vegetation in the water bodies (Williams, 1981; Marte, 1980; Nelson, 1981; Laber, 1985). Therefore, if shrimp farmers maintain the water quality so that there is a sufficient amount of natural food, they could decrease their expenditure on food.



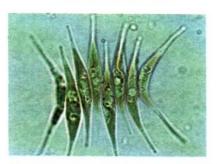
Merismopedia (x1,000)



Oscillatoria (x1,000)



Phormidium (x1,000)



Scenedesmus acuminatus (x1,000)



Scenedesmus bernardii (x1,000)

Figure 2 Genera of plankton most commonly found in the stomachs and intestines of the shrimp samples

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