# Chapter 2

# Methodology

# 2.1 Source of data

Data were obtained from two-survey cruises of MV.SEAFDEC in the Gulf of Thailand and East Coast of Peninsular Malaysia under the Interdepartmental Collaborative Research Program in the South China Sea area of the Southeast Asian Fisheries Development Center. The first cruise was between 3 September and 3 October 1995. The second cruise was from 23 April to 23 May 1996, with a total of 81 oceanographic stations (Fig. 2.1). Station no. 27 was surveying only the second cruise.



Figure 2.1 Oceanographic stations of MV.SEAFDEC between 3 September and 3 October 1995 and from 23 April to 23 May 1996.

#### 2.1.1. High resolution oceanographic data

All data were collected using the onboard Falmouth Integrated CTD instrument with conductivity, temperature and pressures sensors having an accuracy of  $\pm$  0.003 mmhn,  $\pm$  0.003 °c and  $\pm$  0.03%, respectively. There were two additional sensors: an FSI oxygen sensor ( $\pm$  0.1% accuracy) and a Sea Tech submersible fluorescence sensor. Raw counts of each variable were recorded and averaged at every 1 meter interval using the FSI post acquisition data analysis software.

#### 2.1.2 Low resolution oceanographic data

Low-resolution data comprise of nitrate ( $\mu$ M), Chlorophyll-a (mg/m<sup>3</sup>) and dissolved oxygen from titration (ml/l). All data were determined from the water sample collected using twelve 2.5 liter bottles equipped with The CTD. Water samples were collected on selected depth. The concentration of nitrate were determined by a two-channel auto-analyzer "TRAACS 2000" (Yasin et al., 1997). Dissolved oxygen concentration was determined by a modification of the Winkler procedure (Parsons et al., 1984) for the calibration of dissolved oxygen data. Chlorophyll-a concentration was determined by Spectrophotometric method (Parson et al., 1989).

#### 2.1.3. Biological data

1. Phytoplankton data (larger than 20 µm)

Phytoplankton data were available from Boonyapiwat (1999). Van Dorn water sampler was used to collect surface water then filter by 20  $\mu$ m plankton net. The data using in this study were the abundance in cell /m<sup>3</sup> of total phytoplankton, dominant and associate species of each station.

According to the fluorescence data, the maximum concentration of phytoplankton in this area should be at subsurface water (Fig. 2.2). However phytoplankton data was only available at surface layer, therefore chlorophyll-a which can be calculated from fluorescence data were assumed to be abundance of phytoplankton in the water column of this study.

#### 2. Zooplankton data

Total abundance (no/m<sup>3</sup>) and species composition of zooplankton from the study of Jivaluk (1999) were used in this study. Zooplankton samples were collected by Bongo net oblique haul.

3. Relative abundance of Pelagic fish (no/m<sup>3</sup>)

The Relative abundance of pelagic fish is from the study of Theparoonrat et al. (1999). The hydro-acoustic survey using The FURUNO FQ-70 scientific echo sounder, which was equipped with an echo integrator and two quasi-ideal beam transducers operated frequencies of 50 and 200 kHz.

The relative abundance of pelagic fish in this study was interpreted from the 200 kHz frequency, which is less effective for detecting plankton because at these frequencies the noise signal from plankton is decreased. The volume back scattering strength (SV) of the fish schools was collected from the sea surface to the depth of 10 meter above the sea bottom along the

cruise track. The SV data were used to calculate the relative abundance of pelagic fish (Theparoonrat et al., 1999).

In order to calculate the relative abundance, Theparoonrat et al. (1999) used the target strength (TS) of sardine, *Sadinella gibbosa* as the representative species of fish in the observation area, as they were the highest catch of pelagic species in the Gulf of Thailand in 1991-1994. *Sadinella gibbosa* feed on both phytoplankton and zooplankton (Fishbase, 2000)





#### 2.2 Data preparation

#### 2.2.1 Estimation of chlorophyll-a from fluorescence data

Chlorophyll-a data were estimated from the correlation equation between fluorescence (V) data from the SEA TECH fluorometer and the concentration of chlorophyll-a measured in the corresponding water samples. The regression of chlorophyll-a concentrations against the fluorometer output (in V) is a linear response as defined by manufacturer (Sea Tech Inc., 2001).

# 2.2.2 AOU and Preformed Nitrate

AOU is the difference between the oxygen concentration at equilibrium with the atmosphere at the *in situ* temperature and salinity, and the observed oxygen concentration (Redfield, Ketchum and Richard cited in Park, 1967(a))

- AOU =oxygen consumption by biological oxidation in the water
- $O_2$  = calculated concentration of dissolved oxygen at saturation with a wet atmosphere at the potential temperature of the sample
- $O_2$  = observed dissolved oxygen concentration in sample (ml/l)

The nitrate presence in seawater can be separated into two fractions. The first is nutrients of oxidative origin that have been regenerated from organic matter and the second is preformed nutrients that were present in the water at the time it sank from the surface (Redfield, Ketchum, and Richards cited in Park, 1967(a)). Therefore, the measured nitrate data can be expressed as follows (Park, 1967(a)):

$$N_{means} = N_p + N_{ox} \tag{2.2}$$

 $N_{means} = \text{Nitrate from measurement } (\mu M)$   $N_{p} = \text{Concentration of preformed nitrate } (\mu M)$   $N_{ox} = \text{Concentration of nitrate regenerated from or during the}$ biochemical oxidation of organic matter (  $\mu M$  )

The preformed nitrate is conservative in the same sense as which temperature and salinity are conservative; it is modified by run-off, precipitation, and the mixing of water types but is not affected by biological activity, which can be used to trace the water mass (Redfield, 1934).

From the assumed ratios in the biochemical changes in  $O_2$ : C : N : P of 138 : 106 : 16 : 1 by atoms (Richards et al. cited in Park 1967(a)), the nutrient concentrations of oxidative origin in equation 2.2 can be expressed as functions of apparent oxygen utilization (AOU). For instance, 16  $\mu$ M of oxidative nitrate will correspond to 138  $\mu$ M of AOU. Substituting these AOU functions in equations 2.2, we obtain:

$$N_{means} = N_p + 0.12AOU \tag{2.3}$$

### 2.2.3 Quality control of data

All oceanographic data were profiled and visualized, any remaining outliners were subsequently removed manually and using the excessive gradients check procedure of the National Oceanographic Data Center (NODC) to check the quality of data.

For each variable from the study, a check was made for excessive decreases and increases in value over a depth, or excessive gradients (NODC, 1999). A gradient was defined as:

$$gradient = \frac{Z_2 - Z_1}{V_2 - V_1}$$
(2.4)

where  $V_1$  = the value of the variable at the current depth level

 $V_2$  = the value of the variable at the next depth level

 $Z_1$  = the depth (meters) of the current depth level

 $Z_2$  = the depth (meters) of the next depth level

Two types of gradients were checked. The first was an Excessive Gradient, which was a negative gradient, i.e. an excessive decrease in the value over depth. The second was an Excessive Inversion, which was a positive gradient, i.e. an excessive increase in value over depth. The criteria used to define excessive as a function of variable were listed in table 2.1.

Data which exceeded the "maximum gradient value" (MGV) and "maximum inversion value"(MIV) was removed from further analysis.

Variable	Unit/Scale	MIV/meter	MGV/meter
Temperature	°C	0.300	0.700
Salinity	psu	9.000	9.000
Oxygen	ml/l	1.000	1.000
Nitrate	μΜ	1.000	1.000
Chlorophyll	μg/l	1.000	1.000

Table 2.1	Maximum gradient and inversion factors from NODC for the depth less than
	400 meter

#### 2.3 Water mass identification procedure

Water mass was defined as a body of water with a common formation history, which was usually based upon the observation that water renewal in the deep ocean was the result of water mass formation in contact with the atmosphere and spreading from the formation region. A point in the functional relationship of water mass was defined as a water type. Direction and quantity of the spreading and mixing of water mass with other water masses can be tracked by analyzing the distribution of conservative properties, that is they altered only by processes occurring at the boundaries of the ocean by mixing with the other water masses, such as temperature, salinity, etc. (Open University, 1989).

Several methods can be used to study water mass in the ocean. In this study, three methods; Temperature-Salinity diagram (TS-diagram), Temperature-Salinity-time diagram (TS-time diagram) and Optimum Multi-Parameter analysis (OMP analysis) were used to identify water mass in the Gulf of Thailand and East Coast of Peninsular Malaysia. Each method was applied in order to fit with biological data that were obtained independently.

Since stratification occurs over the whole study area especially in April 1996. In this study, water columns were divided into two layers: surface (well-mixed) layer and bottom layer (under the stratification layer). The sigma-theta gradient was used as criteria for identifying layer. The surface layer starting from surface down to the depth of sigma-theta gradient was equal or more than 0.5 kg/m<sup>3</sup> per one-meter depth. Below this gradient was the bottom layer. The percentages of mixed layer of each station were shown in Fig. 2.3.



September 1995

April - May 199



#### 2.3.1 TS-diagram

The temperature-salinity (TS-) diagram is a basic tool for water mass classification and analysis in deep-sea oceanography. It is the plot of two conservative properties against each other. A water mass with uniform temperature and salinity including water masses in mixed layers shows up in a TS-diagram as a single point. Other water masses, which present some variation of their properties with depth, will be shown in the TS-diagram as curves.

However, numbers of water masses were limited by the difference of these two properties, which may not be able to represent all water masses especially in coastal sea and mixing layer.

The temperature and salinity data of all depths were plotted against each other using the Ocean Data View Software (ODV). ODV presents depth as Z-axis with colors of each X, Y coordinate. Results from TS-diagram were presented as the range of temperature and salinity of each water mass.

# 2.3.2 TS-time diagram

In the coastal ocean, even where vertical stratification is present, a large part of the water column is still taken up by the surface mixed layer, which in a TS-diagram is represented

by a single water type. The properties of the mixed layers usually have large changes from season to season and do not fluctuate in a random, but in a seasonal pattern, which can be defined the water masses through the use of a so-called **"TS-time diagram"** (Tomczak, 1996). The values of both variables in the mixed layer were plotted against each other. The sequence of observations taken over the year defines a TS- relationship in time that reflects the weekly and seasonal changes of the two properties.

This technique combine with information from the environmental preferment of various marine life forms could give a fairly accurate variation of the physical environment over the year and how it might effect the coastal ecosystem.

Temperature and salinity data of the surface layer of both data sets were plotted against each other using the ODV software. Seasons were represented as a Z-axis by the color. The results were presented as the ranges of temperature and salinity in different season.

# 2.3.3 Optimum multi-parameter analysis (OMP)

OMP-analysis was developed to solve the situation that beyond the capacity of TSdiagram such as in coastal area, etc. Almost all available properties can be used but conservative ones will give better result (Tomczak and Large, 1989).

The basic assumption of the OMP analysis is that all water masses can be represented as "mixtures" of minimum water types (Tomczak and Large, 1989). Because the OMP analysis solves a linear system of mixing equations, all property relationships between the defined water types must be linear. However, non-linear property relationships in a water mass can be approximated by a series of linear property relationships connecting a finite number of defined water types (the degree of accuracy achieved by this procedure is limited only by the number of parameters available from observations).

An oceanic mixing situation can be expressed through the linear system of equations

$$W(Gx - d) = R \tag{2.5}$$

- $G \longrightarrow a$  matrix containing the parameter values that define the water types.
- d ---> a vector containing the data (observed parameter values that describe a water sample)
- x —> a vector containing the relative contributions, or mixing ratios, of the water types to the sample.
- $R \longrightarrow a vector of residuals.$
- $W \longrightarrow a$  diagonal weight matrix (weights are allocated to the various parameters according to the degree of conservation, instrumentation and accuracy of the method and instrument.

The OMP analysis solves the equation W(Gx-d) = R by minimizing R subject to certain conditions which commonly use the "least squares" approach with the condition of non negative values as the physical process of mixing requires that no element of the solution vector-x is negative.

Temperature, salinity, AOU and preformed nitrate data were used in OMP analysis of this study. In mixing layer, AOU and preformed nitrated cannot be expected to be a conservative parameter. In this area biological activity is significant, leading to high rate of nitrate consumption, and large exchanging of oxygen between water and atmosphere. Therefore, it was not including in OMP-analysis for decreasing error of calculation. The results from OMP analysis were presented as ratios of water type in each water density range. OMP analysis consists of the following steps (Tomczak and Large, 1989):

1. Plot all available parameters against temperature for the regions where only sources water masses were found for establishing G in equation 2.5.

Data of northern most, southern most and eastern most of the survey area were assumed as the information of source of water mass. The parameter's value of source of water mass, which correspond to the end points of the temperature range, the temperature end points themselves and mass conservation, define the matrix G.

2. Establish the matrix W (equation 2.5). Each parameter has its own weight which depends on the measurement accuracy, degree of conservation, and other processes which may cause some parameters to be less reliable than others. The weight are derived from

 $\sigma_i$  is the standard deviation of parameter *i* over the entire data set (a measure where of the ability of a parameter *i* to resolve differences in water mass content)

 $\delta_{i}$  is the smallest of the variances for the water masses.

$$\sigma_{j} = \sqrt{\frac{1}{n} \sum_{i=1}^{n} (G_{ji} - G_{j})^{2}} \qquad (2.7)$$

where  $G_j$  is a mean given by

$$G_{j} = \frac{1}{n} \sum_{i=1}^{n} G_{ji}$$
(2.8)

The largest of the weights W (usually that of temperature) is also assumed for the mass conservation, since equation 2.6 is not applicable to mass conservation.

3. Solve the matrix equation W(G'x-d')=R by minimizing the residual using iterative method.

#### 2.4 Relationship between water mass and biological data

Some biological data available at surface layer, some available at every meter, etc. Therefore, the water mass that will be used to relate with such biological data must be

identified by different method to match with those data.

# 2.4.1 Phytoplankton

# Phytoplankton data

Phytoplankton data were available only in the surface layer. Cluster analysis function of S-PLUS 2000 Software was applied to determine the group of stations with similar in dominant and associate species and percentage of blue green algae, diatom and dinoflagellate from total abundance of phytoplankton. The squared Euclidean distance algorithm was used to calculate the inter-individual distance for clustering.

# Chlorophyll-a

Chlorophyll-a data were estimated from fluorescence data, which was available at every meter. Contours of chlorophyll-a data were plotted to match the water masses identified by OMP analysis.

### 2.4.2 Zooplankton

Cluster analysis by the same software and algorithm that was used with phytoplankton was applied to determine the group of stations with similar in species composition and abundance of zooplankton. The available zooplankton data was represented for the whole water column, therefore results from cluster analysis were determine the relationship with water masses classified by TS-diagram, which applied for the whole water column data.

# 2.4.3 Relative abundance of pelagic fish

The available relative abundance of pelagic fish (no./ton) data was calculated from hydroacoustic data for 0.1 x 0.1 square nautical mile x 10 m depth represented at each station (Theparoonrat et al, 1999). Relative abundance of pelagic fish was plotted as a classed symbol to match the water masses identified by OMP analysis.