#### Chapter II

#### MATERIALS AND METHOD



### Materials

## 1. Collection of specimens

All the specimens were obtained from nature and from artificial breed at fishery station.

Young mullet (<u>Mugil dussumieri Val.</u>) were obtained by net near the shore at Ang-Sila in Cholburi Province on July 8, 1976. Sizes of the specimens ranged from 6.6 cm to 10.5 cm. The specimens were acclimated in a cement tank with aeration. After being acclimated at Ang-Sila for 2 days, there were transported to the laboratory at Chulalongkorn University.

Young seabass (<u>Lates calcarifer [Bloch.]</u>) were obtained from Songkla Fishery Station on November 17, 1976. They were of artificial breeding of the same brood. The size of them ranged from 1.45 cm to 4.00 cm. They had been transported to Bangkok by airfreight.

Young spinefoot (<u>Siganus virgatus</u> Cuv. & Val) were obtained from the coastal area of Ban Phay at Rayoung Province on December 29, 1976 and being acclimated in aerated polyethylene containers for two days. Then, they were transported to the laboratory at Chulalongkorn University. Their size

ranged from 1.90 cm to 3.50 cm.

The fish was transported to the laboratory at Chulalongkorn University by plastic bags filling with the well oxygenated sea water. One plastic bag of  $40 \times 65$  cm size can carry about 200 specimens of 1.45 - 4.00 cm size and about 25 - 30 of 6 - 10 cm size. The transportation time usually not exceed 4 hours.

## II Experimental Unit

There were thhree experimental units in an experiment. They were set up on a three-shelved iron stand, one unit on one shelf. An experimental unit consisted for six rectangular glass aquariums of 25 x 50 x 25 cm size. Each aquarium contained 30 litres of sea water. In order to set up a constant temperature, a 1100 w immersion circulator (heater and thermostatically controlled) was placed inside the aquarium located at one end of the experimental unit. A water pump was used to move water from the heating aquarium to another end of the experimental unit. Siphons were used to circulate the water successively, through each aquarium, to the heating aquarium for reheating the water and simultaneously balancing water level. In order to assure a constant temperature, the inlet and outlet siphons were placed on opposite sides of each aquarium. The temperature of water fluctuated about ± 0.2°C. Before being pumped from the

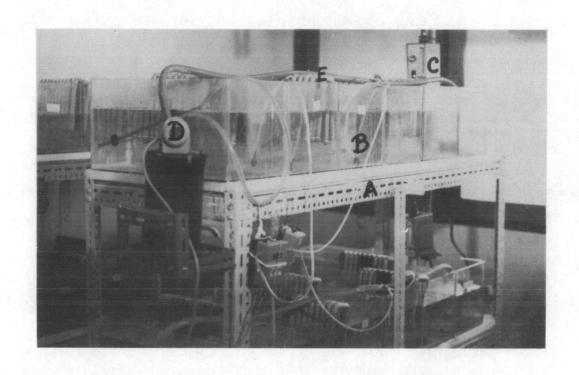


Figure 3 The experimental unit. A.metal stand., B rectangular glass aquariums  $25 \times 50 \times 25$  cm size. C. immersion circulator. D. water pump. E. siphons.

heating aquarium to another end of the experimental unit, the water would pass through a filter of charcoal and synthetic fibre. Each aquarium had one airstone and ten pieces of siphons. Since the temperature of the 23°C experiment was the same as water temperature in the room, the immersion circulator was not used in this particular experiment. All of the immersion circulator was changed every 48 hours. The fresh water was used for controlling a constant salinity.

#### Method

The three experimental units were used for one series of experiment.

Hec

The experiment were carried on at three different temperature 23.0, 28.0 and 33.0°C. Salinity was controlled at 28-30 ppt. Hence, each experimental unit possessed one of these temperature. Fishes were acclimated in the experimental unit for fourteen days before the beginning of the experiment. The initial size was recorded both in terms of weight and of total length.

In the first series of experiment, fifty young mullet each were put into each experimental unit. The fish were fed with fish meal at saturation. Food was given three times a day between 8.00 am and 8.00 pm. After the fish were completely acclimated for 14 days at 23.0°C, 28.0°C and 33.0°C, the experiment commenced.

In the second series of experiment, one hundred and fourteen of seabass were put into three experimental units. Forty-six were put into the 23.0°C experimental unit, forty-four into the 28.0°C experimental unit and fifty into the 33.0°C experimental unit. The fish were fed with shrimp meat at satiation. Food was given three times a day between 8.00 am and 8.00 pm. After they were completely acclimated, the experiment commenced.

In the third series of experiment, two hundred and thirteen of spinefood were put into three experimental units. Sixty-nine were put into the 23.0°C experimental unit, seventy-three into the 28.0°C unit and seventy-one into the 33.0°C unit. The fish were fed at satiation with red worm.

The experiments on mullet and spinefoot lasted 4 weeks, but that of seabass lasted 8 weeks.

# Measurement of growth

The increase of total length and weight were used to express the growth of fishes. The length and weight of mullet and spinefoot were measured every seven days but those of seabass, were measured every fourteen days.

To measure the total length and weight of fish, the fish were anasthetized and each specimen was then removed from the experimental unit into a polyethylene bow which had the diameter of 50 cm. Then, the measurement was taken by

putting each specimen on a plane and fine mech net, using divider with a scale, The total length of each specimen was measured to the nearest 0.05 millimeter. After measuring the total length, water was removed from their bodies by absorbent paper, then, they were put into a 15 cm diametered polyethylene bow containing water and placing on a balance and their weight were recorded.

## Statistical analysis

1. Mean value  $(\bar{X})$ . Since the measurements were taken for samples, the mean value could be calculated as followed:

 $\bar{\chi}$  = X1 + X2 + X3 ..... Xn/N Where X1, X2, X3 .... Xn = value of each sample N = total number of samples

2. Standard deviation ( d ). The standard deviation value could be calculated from each value of sample from the following relationship:

$$C = (X - \overline{X})^2 / N - 1$$

3. Analysis of covariance. The analysis of covariance was used to determine the different in mean value of length and weight of the samples. The test was form the hypothesis that all mean values were equal, assuming that the variances of each set were equal (Snedecor, 1967; Mendenhall, 1962).

Ho = 
$$\mu 1 = \mu 2 = \mu 3$$

mean value of each growth in different temperatures.

F = Mean square of sample mean
Mean square of individual

If the calculated value of F was less than the value of F from the covarian analysis Table at the same degree of freedom, the hypothesis was accepted. But, if the calculated value of F was greater than the value of F from the covarian analysis Table at the same degree of freedom, the hypothesis was rejected.

\* Symbol of covariance analysis

SV = Sources of variation

DF = Degree of freedom

SS = Sum square of treatment

MS = Mean square of treatment