

**EFFECT OF LOW SALINITIES ON THE
GROWTH AND SURVIVAL OF**

PENAEUS MONODON **FABRICIUS**

BY

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THESIS

Submitted in partial fulfilment of the
requirement for the degree

MASTER OF FISHERIES SCIENCE

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Panangad - Cochin

1988

To My Parents

DECLARATION

I hereby declare that this thesis entitled "EFFECT OF LOW SALINITIES ON THE GROWTH AND SURVIVAL OF PENAEUS MONODON (FABRICIUS)" is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

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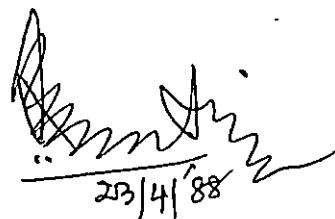
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ACKNOWLEDGEMENTS

Many persons have contributed in one way or another to the completion of my thesis and I am grateful to all of them. Some of them have made critical and constructive contributions that warrant special mention.

I am fortunate to have benefitted from the incisive comments and friendly encouragement of my Adviser Dr. M.J. Sebastian (Dean of Faculty) at all stages of the research. I wish to thank him for his concern and support.

Dr. D.M. Thampy (Professor and Head of the Department of Aquaculture) provided invaluable technical advice and assistance for which I am most grateful.

The recommendations of Shri P.M. Sherief, Shri P.S. Mrithunjayan, Shri Syed Ismail Koya, Shri S. Krishnan, members of the Advisory Committee are greatly appreciated.

For helpful discussions on aspects of the research while in progress I would like to thank Drs. J. Rajasekharan Nair, Sajan George, I.S. Bright Singh (Faculty members) and Laxminarayanan (Scientist and Officer-in-charge, CIBA, Cochin).

My thanks are also due to Shri K.J. Joseph (Joint-Director, Regional Shrimp Hatchery, Azhicode) for the help in the procurement of the samples for the work and M/s Coastal Impex for typing this manuscript.

Sincere thanks go to my friends who were always there when needed. And finally I would like to thank my parents for their encouragement and for always having faith in me.

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INTRODUCTION

INTRODUCTION

1.1 General

In India, prawn and prawn products account for about 83% of the foreign exchange earnings from the seafood export (Anonymous, 1986). However, the inconsistent trends in capture fisheries necessitate the identification and adoption of alternative methods for enhancing prawn production. Scientific farming of prawns appears to be the best alternative in the present context. This is important not only due to its potential for increasing production but also due to its scope for generating employment opportunities for the rural poor.

In the recent years, there has been an increased interest in the culture of marine prawns. Among the 74 species of penaeid prawns known to exist in the Indian region (Mohamed, 1973) Penaeus monodon (Fabricius) is considered the best species for culture purpose (Silas, 1980). This is by virtue of its fast growth, large size at maturity, high fecundity, hardiness to withstand environmental stress, wider distribution and capability to co-exist successfully under high population densities, in both mono and polyculture systems. For tropical countries with higher temperature of water, ranging from 25 to 35°C, it is one of the best species for culture (Aquacop, 1977). It is one of the most important commercial prawns in the West Indo-Pacific region. Successful intensive farming of this species is practised in countries like the Philippines (Villadolid and Villaluz, 1950; Delmendo and Rasalan, 1956; Borja and Rasalan, 1968) Taiwan (Kubo, 1956; Chen, 1972; Liao and Huang 1972) and Indonesia (AlIkunhi et al., 1975). In India selective culture of this species is highly recommended (Ghosh et al.,

1973; Subrahmanyam, 1973; Verghese et al., 1975). P. monodon, along with other species, support a traditional capture fishery in the paddy-fields and bheries of West Bengal. Cultural activities have been extended to a large number of private farms. Recently, attempts have been made to improve their culture practices by introducing modern technologies. Considerable progress has also been made in the country in the intensive culture of this species by experimental trials (Swaminathan, 1980).

1.2 Salinity and Biological Activity

The postlarvae of P. monodon as is the case with most of the commercially important penaeid prawns migrate into the estuaries and backwaters. In the brackishwaters, they grow rapidly into subadults and return to the sea. During their postlarval development, they encounter wide diurnal and seasonal variations in salinity of the environment.

The effect of salinity on marine and brackishwater organisms is of special importance. First, it represents an ecological master factor for most of the aquatic organisms. In tropical countries like India, salinity assumes greater importance as it influences the survival, growth, maturation, spawning and distribution of the animal. Salinity affects functional and structural responses of the animal through changes in

- 1) total osmo-concentration
- 2) relative proportion of solutes
- 3) co-efficient of absorption and saturation of dissolved gases
- 4) density and viscosity, through changes in surface tension and related parameters (Kinne, 1971).

Salinity also exerts an indirect effect by modifying the species composition of the ecosystem. This changes the biotic background for the

remaining forms (Kinne, 1964). However, salinity tolerance tend to decrease as test temperatures, concentrations of dissolved gases and other environmental factors become suboptimal. Besides, salinity tolerance may be subjected to significant intraspecific (inter-populations) differences, when populations occupy habitats with different salinity conditions viz, average salinities, extent and speed of salinity fluctuation (Kinne, 1971).

There are only few systematic studies and reports on the survival and growth of P. monodon along the South West Coast of India. Lack of prior knowledge on the levels and rates at which this species can adopt to lower salinities, has led in the past to unrealistic conclusions. Knowledge on these aspects has great implications in the culture of the species. While planning to undertake commercial farming of P. monodon, it was found that detailed information on the salinity tolerance of the postlarvae and juveniles, optimum salinity for maximum rate of survival and growth, effect of sudden fluctuation in salinity on the survival rate etc. would be very much useful.

Another reason for undertaking such a study was that postlarvae of P. monodon appear in the wild collections of prawn seed during the months from February to June (Thampy et al., 1980). The monsoon rains start atleast during the month of June in Kerala and as such the postlarvae collected in June could not be used in commercial farming. Seed collected in earlier months and grown in ponds would be in danger if salinity is such an important factor in the early development of this species. Moreover, for the proper maintenance of P. monodon postlarvae in nurseries and other holdings, it is necessary to have a clear idea of the environmental requirements, of which salinity tolerance is one of the most important.

In view of the above facts and In view of a lack of clear information on the aspects in question, a laboratory study was undertaken on the effect of low salinities on the growth and survival of postlarvae and juveniles of this prawn.

REVIEW OF LITERATURE

REVIEW OF LITERATURE

2.1 Natural Distribution

Salinity is an important factor influencing the distribution of penaeids. Penaeus setiferus is more abundant at lower salinities than P. aztecus and P. duorarum (Williams, 1955; Gunter et al., 1964). All these three species have been found at salinities from less than 1 ppt (Gunter and Shell, 1958; Tabb et al., 1962; Gunter and Hall, 1963). The survival and growth of P. aztecus postlarvae in Barataria Bay were related to salinity (Amant et al., 1965). Preference for reduced salinities over seawater in postlarvae and early juveniles has been demonstrated experimentally for four species of penaeids from Western Mexico (Mair, 1980).

The availability of postlarvae of P. monodon fluctuated to a great extent according to change in the physico-chemical factors of the environment. The occurrence and distribution of postlarvae of this species with reference to salinity in the various estuarine and brackishwater systems in India such as Hoogly-Matlah estuary (Verghese et al., 1979; Chakraborti et al., 1982) Kovelong backwaters (Rajendran and Sampath, 1975) Marakanam estuary (Bose and Venkatesan, 1982) Vellar estuary, (Ramaswamy and Pandian, 1985a) Ennore estuary (Vasudevan and Subrahmoniam, 1985) Pichavaran (Chandrasekaran and Natarajan, 1984) off Orissa coast (Rajyalakshmi et al., 1985) Pulicat lake (Raj, 1980; Raj and Raj, 1982a) and Cochin backwaters (Sebastian et al., 1980; Thampy et al., 1982) has been reported. Gunter et al. (1964) reported that increase in rainfall and consequent dilution of coastal waters might prove more favourable to the adult white shrimp. The heavy prawn catches in New

South Wales and Queensland during monsoon or immediately after monsoon may probably be due to the mass migration consequent upon the lowering of salinity (Racek, 1956; 1959). A similar observation of inverse correlation between the catches and salinity was also recorded from Pulicat lake. Declining salinity was conducive for survival and growth of postlarvae of P. monodon resulting in better catches (Rao and Gopalakrishnayya, 1974). A regression model, catch/net hour with salinity, show that salinity is a prominent factor in the prediction of P. monodon catch (Karmakar, 1982). Pearse and Gunter (1957) stated that young of many animals usually thought of as marine, required areas of low salinity for nursery grounds. The early stages required oceanic water, but the older larvae must reach bay water or perish.

However, some authors have felt salinity to be of less importance (Hoese, 1960; Broad, 1962). According to Lindner and Anderson (1956) size of young shrimp was correlated more with locality than salinity and concluded that salinity within a broad range was not important.

2.2 Osmoregulation

Crustacean osmoregulation studies have dealt primarily with salinity effects on the isosmotic and ionic properties of the haemolymph (Mantel and Farmer, 1983). Changes in water and ion concentration of haemolymph with the environment have been studied on a variety of penaeid species; P. indicus (Parado-Esteva et al., 1987) P. aztecus (Bishop et al., 1980; Howe et al., 1982) P. monodon (Cawthorne et al., 1983; Ferraris et al., 1986, 1987) Penaeids exposed to low salinities maintain blood osmolarity and ionic concentrations hyperosmotic and hypertonic to the external medium (Mangum

et al., 1976). Upon transfer to high salinity environments, blood ionic and osmotic concentrations increase due to efflux of water and inward movement of ions. Cell volume during hypersaline stress is regulated by an increase in intra-cellular free amino-acids, resulting in a decrease in ammonia excretion, indicating the greater use of endogenous ammonia to synthesize amino acids. (Mangum and Towle, 1977; Gilles, 1979) Crustaceans experience least osmotic stress and undergo best growth when placed in waters of isosmotic salinity (Panikkar, 1968).

In Crustaceans increased metabolic rate has been reported at salinities differing from the isosmotic point as indicative of the increased energy cost due to osmoregulation (Beadle, 1931; Lofts, 1956; Dehnel and Mc Caughran, 1964; Kutty et al., 1971). However, numerous reports that indicated either a decrease in metabolic rate in supra/subnormal salinities (Simmons and Knight, 1975) or no correlation between metabolic rate and salinity (Elfringham 1965; Mc Farland and Pickens, 1965) has led to difficulty in interpreting results of environmental changes on metabolic rate. Gunter et al. (1964) observed that adaptation to low salinity in penaeids is highly developed in younger stages while Panikkar (1968) has indicated that osmoregulation in dilute media is less effective in full-sized individuals especially in their reproductive phase. He considers this as one of the principal factors which forces them to migrate back to the sea. Mc Farland and Lee (1963) demonstrated that brown shrimp adults were better osmoregulators at higher salinity than at lower, with a greater tendency to isosmoticity when the external medium was below 18 ppt. Moulting interacted with salinity in inducing stress in penaeids. In culture systems high mortalities were observed when moulting coincided with large fluctuations in salinity. (Ferraris et al., 1987). However, dependence on external factors gradually declined in older moult stages. This suggests there is a reduction in integument

permeability and greater development of ion absorption or secretion mechanism as the exoskeleton hardened (Ferraris et al., 1987).

2.3 Growth and Survival

Zein-Eldin (1963) carried out pioneering works regarding the influence of salinity on the growth in prawns. He concluded that under conditions of constant temperature and restricted food supply Penaeus postlarvae survived and grew over a wide range of salinity (2-40 ppt). Zein Eldin and Aldrich (1965) observed salinity had little effect on either survival or growth of postlarvae of P. aztecus except at extreme temperatures. Jhonson and Fielding (1956) in attempts to rear shrimps in ponds noted that P. setiferus juveniles could be raised at 18.5 and 34 ppt salinity levels. Williams (1960) in experiments concerning the osmotic relationship of brown and pink shrimp found that temperature apparently had far more effect upon survival than did salinity. However, several experimental studies that followed revealed that salinity influenced the survival and growth of penaeid postlarvae and juveniles. (Grajcer and Neal, 1972; Nair and Kutty, 1975; Bhattacharya and Kewalramani, 1976; Kuttyamma, 1982; Lakshmikantham, 1982). Many others focussed on the cumulative effect of temperature and salinity on the survival, growth, food intake and food conversion efficiency (Venkataramaiah et al., 1975).

Kramer (1975) reported the influence of salinity on the lethal dissolved oxygen levels in prawns. Exposure to subnormal salinities resulted in reduced tolerance to other environmental factors like temperature, dissolved gases or hydrostatic pressure (Kinne, 1971). However, studies on the effect of salinity on the survival and growth of P. monodon are limited (Subrahmanyam, 1973; Verghese et al., 1975; Liao, 1977; Hideo, 1979; Motoh, 1981; Raj and Raj 1982b; Chakraborti et al., 1985a). There are few reports on the growth of P. monodon

adult in ponds of fluctuating physico-chemical conditions (Verghese et al., 1982; Pillai et al., 1984; Venkatesan and Joshua, 1984; Chakraborti et al., 1985b, 1986; Chen, 1985). Sebastian et al. (1980) reported that sudden drop in salinity of pond-waters was harmful for adults of P. monodon. Adults cultivated in the inundated paddy-fields of Kuttanad in Kerala under less than 1 ppt salinity grew well until the onset of the first monsoon rain with which all of them showed great stress and mass mortality of this species was recorded. On the contrary Das et al. (1982) reported that gradual increase or decrease in salinity did not affect the survival of the adults of this species. In culture systems, increasing salinity at ambient temperature was found to initiate moulting in P. monodon adults while decreasing salinity did not (Hag, 1984). Reyes (1985) reported the influence of salinity on hatching rate of eggs and survival from nauplius to zoea stage in this species.

There are only few systematic studies and reports on the survival and growth of the early stages of P. monodon under low saline conditions. Studies on tolerance of postlarvae/juveniles of this species to wide fluctuations in low salinities and lowest lethal salinity are lacking. In this study an attempt is made to improve the chances of postlarval survival by different levels of acclimation.

MATERIALS AND METHODS

MATERIALS AND METHODS

3.1 Collection and Holding of Experimental Animals

Postlarvae/juveniles of P. monodon raised in the Regional Shrimp Hatchery of Department of Fisheries of Kerala at Azhicode were used as experimental material for the present study. The postlarvae were brought to the lab in oxygen-packed polythene seed transport bags. They ranged in size from 10-12 mm in total length and 3 - 4 mg in weight. A set of juveniles ranging from 25 - 35 mm in total length and weighing 180 - 260 mg was used for the growth experiments. These experiments were conducted in circular cement cisterns of 1 m diameter and 1 m depth (holding capacity 700 l). Tolerance experiments were carried out in cylindrical glass troughs of 10 l capacity (30 cm dia. x 15 cm ht).

The postlarvae/juveniles brought from the hatchery were acclimated to 25 ppt salinity in cement cisterns (pH 7.40 ± 0.36 and temperature of water $28^{\circ}\text{C} \pm 0.5$). The animals (buffer stock) in the acclimation tanks were fed with Tata's pelleted commercial prawn feed of 3 mm diameter ad libitum (Carbohydrate 47% ; Protein 30% ; Fat 7% ; Crude fibre 4-5% ; Moisture 10-12% ; Sand/Silica 2-3% ; Vitamins/Minerals trace). The leftover feed and faecal matter were siphoned out everyday.

3.2 Preliminary Tolerance Experiments

Five sets of experiments were conducted in this study. Animals were pre-acclimated to a particular salinity before releasing them to different test salinities.

3.2.1 Acclimation

Five different concentrations of acclimation salinity were selected (25, 20, 15, 10 and 5 ppt). The animals were transferred from the stock

tank and slowly acclimated to the respective acclimation salinity and then maintained for five days. Animals were fed ad libitum. Mortality during acclimation was negligible.

3.2.2 Tolerance in Test Salinity

From each acclimation salinity, fifteen numbers of postlarvae were transferred to cylindrical glass troughs containing 8 l of water of different test salinities (1.5 ± 0.5 : 4.5 ± 0.5 : 9.5 ± 0.5 : 14.5 ± 0.5 and 19.5 ± 0.5 ppt). The ratio of weight of the animals to the volume of medium was kept well below 2g/l (Duodoroff et al., 1951). The troughs were aerated intermittently and removable hideouts were provided to avoid cannibalism (Rajyalakshmi, 1982). Feed was given in small quantities for maintenance. Leftover feed and faecal matter were siphoned out everyday.

The animals were reared for 120 h at each test salinity. Frequent observations were made to record survival percentage. Dead animals were removed immediately. The experiments were carried out in duplicate.

3.3 Tolerance In Salinity Below 5 ppt

Tolerance of the postlarvae to very low salinities (4, 3, 2, 1 and 1 ppt) was studied after step-wise acclimation. The approach was to evaluate the improvement in survival of postlarvae at these very low salinities by a gradual change in salinity. Fifteen numbers of postlarvae were transferred to cylindrical glass troughs containing 8 l of water at lower salinities (4, 3, 2, 1 and 1 ppt) after step-wise acclimation. The animals were reared for 120 h at each salinity. Frequent observations were made to record survival. LC 50 after 120 h was recorded.

3.4 Lethal Salinity Studies

Three batches of ten animals each were subjected to gradually decreasing salinity from a pre-acclimation salinity of 20 ppt down to 2 ppt at 2 ppt intervals and from 2 ppt at still smaller intervals. The step-wise decrease in salinity was achieved by adding low saline water to the experimental containers. The animals were maintained at each salinity for a period of 24 h before the salinity was changed. Cumulative mortality at each salinity after a period of 24 h was observed. Dead animals were removed immediately. The salinity in which atleast 50 percent of the animals survived at the end of 24 h was considered within the tolerable range and rest as lethal.

3.5 Growth Experiment

Juveniles of P. monodon ranging from 180-250 mg from the buffer stock maintained at 25 ppt salinity were used for the study. The animals were slowly acclimated and released to different test salinities (1.5 ± 0.5 : 4.5 ± 0.5 : 9.5 ± 0.5 : 14.5 ± 0.5 and 19.5 ± 0.5 ppt as control). Lower salinities were prepared by diluting sea water with well water. About 120 l of water was maintained in each of these tanks. Twenty numbers of juveniles were introduced in each of these cement cisterns. After release, they were allowed 48 h to settle before the start of the experiment and monitoring of different parameters. The tanks were aerated intermittently to maintain sufficient oxygen concentration and provided with removable hideouts. Water in these tanks was changed partially every fortnight in order to prevent the lethal accumulation of metabolites and bacteria. Feeding was done with Tata's pelleted

commercial prawn feed of 3 mm diameter, ad libitum in the evening. The leftover feed and faecal matter were siphoned out everyday.

The prawns were reared for 56 days in water of the same salinity to which they were acclimated. Every fortnight, the animals were netted and weighed and the live weight was determined using a K-15 super analytical single pan balance (K. Roy & Co.) having a sensitivity of 0.01 mg. Minor variations that occurred in the salinity during the course of the experiment were adjusted to specified levels. A duplicate experiment was also carried out.

All the above experiments were conducted at room temperature which ranged from 28 to 32°C. The temperature of water in the experimental containers during the course of the experiment ranged from 27 to 29°C.

3.6 Methods of Water Analysis

| | | |
|------------------|---|---|
| Salinity | : | Standard Argentotitric Method (Strickland and Parsons, 1968) |
| Dissolved Oxygen | : | Winkler's Method (Strickland and Parsons, 1968) Analysis was done within an hour of sampling. |
| Temperature | : | Mercury bulb Thermometer (0 to 50°C) |
| pH | : | Potentiometric Method using a Digital pH meter (ELICO - Model L1-122) |
| Alkalinity | : | Titrometric Method to Potentiometric end point (Strickland and Parsons, 1968) |

Fig. A1 Cylindrical glass troughs used as experimental containers in tolerance studies. Size : 30x15 cm : Capacity : 10 l Troughs were filled with 8 l of test medium and aerated intermittently. Temperature of water in the troughs ranged from 27 to 29⁰C.



Fig. A2 Cement cisterns used as experimental containers in growth studies. Size : 1x1 m : Capacity : 700 l. Cement cisterns were filled with 120 l of test medium and aerated intermittently. Temperature of water in the cisterns ranged from 27 to 29°C.



Fig. A2

3.7 Sensory Evaluation

3.7.1 Sample Preparation

Animals of the growth experiments were used in this study. Prawns reared in different test salinities were harvested, peeled, deveined and cleaned. The samples were steam cooked separately for ten minutes under normal pressure. Cooked samples were coded and served on white plastic plates. The cooked samples were held at 28°C until evaluated by the taste panel.

3.7.2 Taste Panel Evaluation

A ten member taste panel was selected. Sensory evaluation utilizing an hedonic scale was used to evaluate the sensory characteristics (texture of flesh, flavour and overall quality). The scale consists of 6 scores from 0 to 6 in the ascending order (Table 15). Panelists were asked to make a judgement on the basis of their preference. Panelists were asked to rinse their mouth with distilled water between samples.

3.8 Statistical Analysis

Tolerance and growth data were subjected to Analysis of Variance (ANOVA). An F-test was performed to determine if difference between treatment (test salinity) means existed. If an F-value was found to be statistically significant, data was analysed by a Least Significance Difference (LSD) test. All possible differences between the means of each treatment were computed and compared to LSD. If the absolute value of the difference (d) was greater than the LSD ($|d| \geq \text{LSD}$), the difference was found to be significant at $P < 0.05$.

Results of experiment to determine Lethal Salinity (3.4) was verified using Probit analysis.

Analysis of the data was performed by a pre-written statistical RBD analysis on "VERSA IWS" of CP/M-86 Model.

RESULTS

RESULTS

4.1 Preliminary Tolerance Studies

Survival percentage of 10-12 mm size postlarvae in the different test salinities after pre-acclimation at indicated levels of salinity over 120 h exposure period is given in Table 6.

Postlarvae pre-acclimated at 25 ppt recorded a survival between 86.6 and 56.6% on sudden transfer (Table 1). Maximum survival was recorded at 14.5 ± 0.5 ppt and minimum at 4.5 ± 0.5 ppt. At 20 ppt pre-acclimation, survival ranged between 50 and 86.65% (Table 2). Maximum and minimum survival were recorded at 19.5 ± 0.5 and 4.5 ± 0.5 ppt respectively. Postlarvae pre-acclimated at 15 ppt recorded a maximum survival of 93.3% in 4.5 ± 0.5 and 19.5 ± 0.5 ppt and minimum of 79.95% at 14.5 ± 0.5 ppt (Table 3). Survival was uniform (93.3%) in all the tested conditions when the postlarvae were pre-acclimated at 10 ppt (Table 4). At 5 ppt pre-acclimation, survival of postlarvae ranged from 89.95 to 96.65% (Table 5). Survival was maximum at 9.5 ± 0.5 ppt and minimum at 4.5 ± 0.5 ppt and 14.5 ± 0.5 ppt. In all the above trials, complete mortality of the postlarvae was observed in less than 1 ppt test salinity within 2 h of their transfer, regardless of pre-acclimation.

4.1.1 Analysis

Survival values less than 1 ppt were not considered for analysis. There was no significant difference ($P \geq 0.05$) in the survival of postlarvae in different test salinities. (ANOVA - Table 6). Postlarvae acclimated at 25 ppt (ANOVA - Table 1) and 20 ppt (ANOVA - Table 2) showed highly significant variation in survival ($P < 0.001$) especially between 4 and 15 ppt.

Postlarvae showed less significant survival ($P < 0.05$) when pre-acclimated at 15 ppt (ANOVA - Table 3). There was no significant difference in survival ($P \geq 0.05$) when the postlarvae were pre-acclimated at 5 ppt (ANOVA - Table 5) and 10 ppt (ANOVA - Table 4).

4.2 Tolerance in Salinity Below 5 ppt

Postlarvae exhibited marked variation in survival when subjected to progressive dilutions below 5 ppt (Table 7). Survival percentage decreased with reduction in salinity levels, recording 93.3, 86.6, 86.65 and 59.5% in 4, 3, 2 and 1 ppt respectively (Fig. 1). In less than 1 ppt, the lowest survival was recorded (46.6%). This salinity was considered lethal (LC 50) for the postlarvae since mortality at the end of 120 h was more than 50%.

Analysis of variance showed highly significant difference in survival ($P < 0.001$) in less than 5 ppt levels of salinity (ANOVA - Table 7).

4.3 Lethal Salinity Studies

Postlarvae recorded cent percent survival upto the third day when salinity was gradually decreased from 20 to 16 ppt (Fig. 2). Postlarvae showed signs of mortality when the salinity reached 14 ppt (96.7% survival). Survival percentage decreased with step-wise reduction in salinity, recording 90, 86.7, 76.7, 66.7 and 53.34% at 8, 6, 2, 1 and 0.75 ppt respectively. At 0.5 ppt the survival was 53.34%. Salinity less than 0.5 ppt proved to be lethal for the postlarvae.

The above results were verified using probit analysis (Table 9). A probit regression line for the relation between the logarithmic salinity (dose = X) and mortality (Probit response = Y) was determined to be $Y = 5.982023 + 1.332194 X$. The proportion responded among controls was

zero. LC 50 for the postlarvae was determined to be 0.5459 ppt with its upper and lower limits being 0.377 and 0.790 ppt respectively. LC 30 and LC 70 values determined were 1.3514 and 0.22055 ppt respectively.

4.4 Behavioural Observation

Animals exhibited abnormal behaviour in salinity less than 1 ppt. On sudden transfer, some of the postlarvae were seen jumping out of water to the sides of the containers. The postlarvae exhibited abnormal swimming pattern, swimming side-wise, with their body inclined to one side. These animals exhibited spiral movements and circular movements at the bottom of the container as well as in the water column. The stress conditions in animals at sub-optimal salinities were evident from the low rate of feeding and excretion of faecal matter. A high rate of cannibalism was observed at the extreme lower salinities.

4.5 Growth Studies

The growth of juveniles in different test salinities are given in Table 10 and Figure 3. Variation in the growth rate of juveniles in different test salinities are given in Table 11. Survival of juveniles recorded during the course of study is given in Table 12. Water quality parameters recorded are given in Table 14.

Graphical representations of growth rate variations in 1.5 ± 0.5 ppt reveal that from an initial increase of 4.43 mg/day to 8.95 mg/day, it dropped to 6.44 mg/day and remained steady, recording 6.63 mg/day (Fig. 5). In test salinities between 4 and 15 ppt, growth rate variations generally followed an increasing trend. The initial growth rate of 10.48 mg/day increased to 14.86 mg/day and later to 14.85 mg/day and

finally recorded 16.11 mg/day in 4.5 ± 0.5 ppt salinity. There was a gradual increase from an initial growth rate of 7.53 mg/day to 10.59 mg/day and then increased to 11.44 mg/day and finally recorded 13.30 mg/day in 9.5 ± 0.5 ppt salinity. In 14.5 ± 0.5 ppt there was a steady increase from 7.0 mg/day to 9.44 mg/day, 13.05 mg/day and 13.41 mg/day. However, in 19.5 ± 0.5 ppt, the initial increase in growth rate from 10.29 mg/day to 13.22 mg/day and 13.94 mg/day later declined to 12.74 mg/day towards the end of the experiment.

Biomass increase* New (1971) of the juveniles in the different test salinities are given in Table 13. Maximum biomass increase was recorded in 14.5 ± 0.5 ppt (18.77 g/m^2). See Figure 7. This was followed by 9.5 ± 0.5 ppt (16.15 g/m^2), 4.5 ± 0.5 ppt (15.03 g/m^2) and 19.5 ± 0.5 ppt (14.93 g/m^2). 1.5 ± 0.5 ppt salinity recorded the least of 1.54 g/m^2 .

Summing up the whole results, overall growth rate was highest in 4.5 ± 0.5 ppt (16.11 mg/day/72.5% survival) followed by 14.5 ± 0.5 ppt (13.41 mg/day/100%) 9.5 ± 0.5 ppt (13.296 mg/day/90%) and 19.5 ± 0.5 ppt (12.74 mg/day/87.5%). See Figure 6. Overall growth rate was least in 1.5 ± 0.5 ppt (6.63 mg/day/50%).

4.5.1 Analysis

There was highly significant difference ($P < 0.001$) in the growth rate of the juveniles in different test salinities (ANOVA - Table 8). Mean values of growth rates of juveniles between 4 and 15 ppt did not differ

*C = $\frac{azy}{100} - bz$ where, a = av. final weight (g), b = av. initial weight (g),
y = Survival percentage, z = Stocking density (nos/m^2), c = Biomass increase (g/m^2)

significantly ($P \leq 0.05$) from the control (19.5 ± 0.5 ppt). Variation in growth rate was highly significant ($P < 0.001$) among the different exposure periods. Variation in the 56-day survival of juveniles was also highly significant (ANOVA - Table 9). Mean values of survival between 1 and 5 ppt differed significantly ($P < 0.05$) from the control (19.5 ± 0.5 ppt).

4.6 Sensory Evaluation

Results of sensory evaluation at different salinities are shown in Tables 16, 18 and 20. The data involved were subjected to primary analysis to determine the Kendall's co-efficient of concordance (Sidney Siegel, 1956) to test the significant association in the judgements (Tables 17, 19 and 21). Only data of those sensory characteristics which were significant were subjected to analysis of variance.

Only in the case of flavour of cooked meat was there a significant association ($P < 0.05$) in the judgements (Table 17). The overall score of shrimps reared at 9.5 ± 0.5 ppt was better than those reared at other salinity levels (Table 17). A significant difference ($P < 0.01$) in cooked flavour was found between shrimps held at 1.5 ± 0.5 and 9.5 ± 0.5 ppt : 9.5 ± 0.5 and 14.5 ± 0.5 ppt and 14.5 ± 0.5 and 19.5 ± 0.5 ppt (ANOVA - Table 10).

Table 1. Survival percentage* of 10-12 mm size postlarvae of P. monodon in different test salinities over different exposure periods after acclimation at 25 ppt.

| Test salinity levels (ppt) | Exposure periods (h) | | | | |
|----------------------------|----------------------|-------|-------|-------|-------|
| | 24 | 48 | 72 | 96 | 120 |
| Less than 1 | - | - | - | - | - |
| 4.5 ± 0.5 | 79 | 69.95 | 59.95 | 56.6 | 56.6 |
| 9.5 ± 0.5 | 96.65 | 93.3 | 76.65 | 76.65 | 69.95 |
| 14.5 ± 0.5 | 100 | 96.65 | 93.3 | 89.95 | 86.6 |
| 19.5 ± 0.5 | 100 | 96.65 | 93.3 | 89.95 | 83.3 |

* Each representative value is the average of two determinations for 15 animals per treatment.

ANOVA - TABLE 1

ANOVA for testing significance of survival of 10-12 mm size postlarvae of P. monodon in different test salinities (TS) over different exposure periods (EP) after acclimation at 25 ppt.

| Source | S.S. | d.o.f. | M.S. | F-ratio |
|-----------------|---------|--------|--------|------------|
| Test Salinity | 1774.59 | 3 | 591.53 | 115.67 *** |
| Exposure Period | 1103.45 | 4 | 275.86 | 53.94 *** |
| Error | 61.37 | 12 | 5.11 | |

| TS Means: | | EP Means: | |
|-------------------------------|---------|-----------|---------|
| TS1 | 53.696 | EP1 | 80.7125 |
| TS2 | 66.6816 | EP2 | 72.6645 |
| TS3 | 76.9396 | EP3 | 65.459 |
| TS4 | 76.3696 | EP4 | 63.2325 |
| | | EP5 | 59.99 |
| CD 5% for TS means comparison | | 3.116 | |
| CD 5% for EP means comparison | | 3.434 | |

*** denotes significant at 0.1%

TS1 = 4.5 ± 0.5 ppt : TS2 = 9.5 ± 0.5 ppt

TS3 = 14.5 ± 0.5 ppt : TS4 = 19.5 ± 0.5 ppt

EP1 = 24 h : EP2 = 48 h : EP3 = 72 h : EP4 = 96 h

EP5 = 120 h

Since complete mortality of the postlarvae was observed in less than 1 ppt test salinity level, it was not considered for analysis.

The individual percentages (x) were transformed into arc size values $(\text{Sin}^{-1} \sqrt{\frac{X}{100}})$ for analysis.

Table 2. Survival percentage* of 10-12 mm size postlarvae of *P. monodon* in different test salinities levels over different exposure periods after acclimation at 20 ppt.

| Test salinity levels (ppt) | Exposure periods (h) | | | | |
|----------------------------|----------------------|-------|-------|-------|-------|
| | 24 | 48 | 72 | 96 | 120 |
| Less than 1 | - | - | - | - | - |
| 4.5 ± 0.5 | 69.95 | 69.95 | 50 | 50 | 50 |
| 9.5 ± 0.5 | 93.3 | 93.3 | 89.95 | 86.6 | 80 |
| 14.5 ± 0.5 | 100 | 96.65 | 96.65 | 89.95 | 83.3 |
| 19.5 ± 0.5 | 100 | 100 | 96.65 | 89.95 | 86.65 |

* Each representative value is the average of two determinations for 15 animals per treatment.

ANOVA - TABLE 2

ANOVA for testing significance of survival of 10-12 mm size postlarvae of P. monodon in different test salinities (TS) over different exposure periods (EP) after acclimation at 20 ppt.

| Source | S.S. | d.o.f. | M.S. | F-ratio |
|-----------------|---------|--------|--------|-----------|
| Test salinity | 2814.56 | 3 | 938.19 | 73.07 *** |
| Exposure period | 847.18 | 4 | 211.79 | 16.50 *** |
| Error | 154.07 | 12 | 12.84 | |

| TS Means: | | EP Means: | |
|-------------------------------|---------|-----------|---------|
| TS1 | 49.704 | EP1 | 77.9395 |
| TS2 | 70.6952 | EP2 | 75.352 |
| TS3 | 77.33 | EP3 | 68.905 |
| TS4 | 79.948 | EP4 | 64.1425 |
| | | EP5 | 60.7075 |
| CD 5% for TS means comparison | | 4.9405 | |
| CD 5% for EP means comparison | | 5.5209 | |

*** denotes significant at 0.1%

TS1 = 4.5 ± 0.5 ppt : TS2 = 9.5 ± 0.5 ppt

TS3 = 14.5 ± 0.5 ppt : TS4 = 19.5 ± 0.5 ppt

EP1 = 24 h : EP2 = 48 h : EP3 = 72 h

EP4 = 96 h : EP5 = 120 h

Since complete mortality of the postlarvae was observed in less than 1 ppt test salinity level, it was not considered for analysis.

The individual percentages (x) were transformed into arc sine values $(\text{Sin}^{-1} \sqrt{\frac{x}{100}})$ for analysis

Table 3. Survival percentage* of 10-12 mm size postlarvae of P. monodon in different test salinities over different exposure periods after acclimation at 15 ppt.

| Test Salinity levels (ppt) | Exposure periods (h) | | | | |
|----------------------------|----------------------|-------|-------|-------|-------|
| | 24 | 48 | 72 | 96 | 120 |
| Less than 1 | - | - | - | - | - |
| 4.5 ± 0.5 | 96.65 | 93.3 | 93.3 | 93.3 | 93.3 |
| 9.5 ± 0.5 | 100 | 89.95 | 89.95 | 89.95 | 89.95 |
| 14.5 ± 0.5 | 100 | 86.65 | 86.65 | 79.95 | 79.95 |
| 19.5 ± 0.5 | 100 | 100 | 96.65 | 96.65 | 93.3 |

* Each representative value is the average of two determinations for 15 animals per treatment.

ANOVA - TABLE 3

ANOVA for testing significance of survival of 10-12 mm size postlarvae of P. monodon in different test salinities (TS) over different exposure periods (EP) after acclimation at 15 ppt.

| Source | S.S. | d.o.f. | M.S. | F-ratio |
|-----------------|--------|--------|--------|----------|
| Test salinity | 376.39 | 3 | 125.46 | 4.59 *** |
| Exposure period | 619.60 | 4 | 154.90 | 5.66 *** |
| Error | 328.30 | 12 | 27.36 | |

| TS Means: | | EP Means: | |
|-------------------------------|--------|-----------|---------|
| TS1 | 75.33 | EP1 | 86.6625 |
| TS2 | 75.216 | EP2 | 76.272 |
| TS3 | 70.788 | EP3 | 73.6845 |
| TS4 | 82.859 | EP4 | 72.392 |
| | | EP5 | 71.2290 |
| CD 5% for TS means comparison | | 7.2118 | |
| CD 5% for EP means comparison | | 8.0591 | |

* denotes significant at 5%

** denotes significant at 1%

TS1 = 4.5 ± 0.5 ppt : TS2 = 9.5 ± 0.5 ppt

TS3 = 14.5 ± 0.5 ppt : TS4 = 19.5 ± 0.5 ppt

EP1 = 24 h EP2 = 48 h EP3 = 72 h

EP4 = 96 h EP5 = 120 h

Since complete mortality of the postlarvae was observed in less than 1 ppt test salinity level, it was not considered for analysis.

The individual percentages (x) were transformed into arc sine values ($\text{Sin}^{-1} \sqrt{\frac{x}{100}}$) for analysis.

Table 4. Survival percentage* of 10-12 mm size postlarvae of P. monodon in different test salinities over different exposure periods after acclimation at 10 ppt.

| Test Salinity levels (ppt) | Exposure periods (h) | | | | |
|----------------------------|----------------------|-------|-------|------|------|
| | 24 | 48 | 72 | 96 | 120 |
| Less than 1 | - | - | - | - | - |
| 4.5 ± 0.5 | 100 | 96.65 | 93.3 | 93.3 | 93.3 |
| 9.5 ± 0.5 | 100 | 100 | 96.65 | 93.3 | 93.3 |
| 14.5 ± 0.5 | 100 | 96.65 | 93.3 | 93.3 | 93.3 |
| 19.5 ± 0.5 | 100 | 96.65 | 93.3 | 93.3 | 93.3 |

* Each representative value is the average of two determinations for 15 animals per treatment.

ANOVA - TABLE 4

ANOVA for testing significance of survival of 10-12 mm size postlarvae of P. monodon in different test salinities over different exposure periods after acclimation at 10 ppt.

| Source | S.S. | d.o.f. | M.S. | F-ratio |
|-----------------|--------|--------|--------|-----------|
| Test salinity | 33.76 | 3 | 11.25 | 2.09 NS |
| Exposure period | 675.48 | 4 | 168.87 | 31.36 *** |
| Error | 64.61 | 12 | 5.38 | |

| TS Means: | | EP Means: | |
|-------------------------------|--------|-----------|---------|
| TS1 | 78.888 | EP1 | 90.0 |
| TS2 | 81.889 | EP2 | 82.0875 |
| TS3 | 78.888 | EP3 | 76.111 |
| TS4 | 78.888 | EP4 | 74.998 |
| | | EP5 | 74.998 |
| CD 5% for TS means comparison | | 3.1979 | |
| CD 5% for EP means comparison | | 3.5759 | |

NS = Non-significant

*** denotes significant at 0.1%

TS1 = 4.5 ± 0.5 ppt : TS2 = 9.5 ± 0.5 ppt

TS3 = 14.5 ± 0.5 ppt : TS4 = 19.5 ± 0.5 ppt

EP1 = 24 h EP2 = 48 h EP3 = 72 h

EP4 = 96 h EP5 = 120 h

Since complete mortality of the postlarvae was observed in less than 1 ppt test salinity level, it was not considered for analysis.

The individual percentages (x) were transformed into arc sine values ($\text{Sin}^{-1} \sqrt{\frac{x}{100}}$) for analysis.

Table 5. Survival percentage* of 10-12 mm size postlarvae of P. monodon in different test salinities over different exposure periods after acclimation at 5 ppt.

| Test Salinity levels (ppt) | Exposure periods (h) | | | | |
|----------------------------|----------------------|-------|-------|-------|-------|
| | 24 | 48 | 72 | 96 | 120 |
| Less than 1 | - | - | - | - | - |
| 4.5 ± 0.5 | 100 | 96.65 | 96.65 | 93.3 | 89.95 |
| 9.5 ± 0.5 | 100 | 96.65 | 96.65 | 96.65 | 96.65 |
| 14.5 ± 0.5 | 100 | 96.65 | 93.3 | 93.3 | 89.95 |
| 19.5 ± 0.5 | 96.65 | 96.65 | 93.3 | 93.3 | 93.3 |

* Each representative value is the average of two determinations for 15 animals per treatment.

ANOVA - TABLE 5

ANOVA for testing significance of survival of 10-12 mm size postlarvae of P. monodon in different test salinities (TS) over different exposure (EP) periods after acclimation at 5 ppt.

| Source | S.S. | d.o.f. | M.S. | F-ratio |
|-----------------|--------|--------|--------|-----------|
| Test Salinity | 60.56 | 3 | 20.19 | 2.42 NS |
| Exposure Period | 412.05 | 4 | 103.01 | 12.35 *** |
| Error | 100.06 | 12 | 8.34 | |

| TS Means: | | EP Means: | |
|-------------------------------|-------|-----------|---------|
| TS1 | 79.08 | EP1 | 87.3625 |
| TS2 | 81.56 | EP2 | 79.45 |
| TS3 | 78.19 | EP3 | 77.224 |
| TS4 | 76.78 | EP4 | 76.111 |
| | | EP5 | 74.372 |
| CD 5% for TS means comparison | | 3.9817 | |
| CD 5% for EP means comparison | | 4.44925 | |

NS denotes non-significant

*** denotes significant at 0.1%

TS1 = 4.5 ± 0.5 ppt : TS2 = 9.5 ± 0.5 ppt

TS3 = 14.5 ± 0.5 ppt : TS4 = 19.5 ± 0.5 ppt

EP1 = 24 h EP2 = 48 h EP3 = 72 h

EP4 = 96 h EP5 = 120 h

Since complete mortality of the postlarvae was observed in less than 1 ppt test salinity level it was not considered for analysis.

The individual percentages (x) were transformed into arc sine values ($\sin^{-1} \sqrt{\frac{x}{100}}$) for analysis.

Table 6. Survival percentage* of 10-12 mm size postlarvae of P. monodon in different test salinities over 120 h exposure period after acclimation at indicated salinities (compiled from the results of Tables 1, 2, 3, 4 and 5).

| Acclimation salinity (ppt) | Test Salinity levels (ppt) | | | |
|----------------------------|----------------------------|-----------|------------|------------|
| | 4.5 + 0.5 | 9.5 ± 0.5 | 14.5 ± 0.5 | 19.5 ± 0.5 |
| 5 | 89.95 | 96.65 | 89.95 | 93.3 |
| 10 | 93.3 | 93.3 | 93.3 | 93.3 |
| 15 | 93.3 | 89.95 | 79.95 | 93.3 |
| 20 | 50 | 80 | 83.3 | 86.65 |
| 25 | 56.6 | 69.95 | 86.6 | 83.3 |

* Each representative value is the average of two determinations for 15 animals per treatment.

ANOVA - TABLE 6

ANOVA for testing significance of 10-12 mm size P. monodon postlarvae surviving 120 h exposure period in different test salinities (TS) after acclimation at indicated salinities (AS)

| Source | S.S. | d.o.f. | M.S. | F-ratio |
|----------------------|--------|--------|--------|---------|
| Test salinity | 184.44 | 3 | 61.43 | 1.21 NS |
| Acclimation salinity | 256.63 | 4 | 189.16 | 3.73 * |
| Error | 609.27 | 12 | 50.77 | |

| TS Means: | | AS Means: | |
|-------------------------------|-------|-----------|---------|
| TS1 | 63.06 | AS1 | 74.37 |
| TS2 | 69.23 | AS2 | 74.99 |
| TS3 | 68.87 | AS3 | 71.23 |
| TS4 | 71.89 | AS4 | 60.72 |
| | | AS5 | 59.99 |
| CD 5% for TS means comparison | | | 9.8198 |
| CD 5% for AS means comparison | | | 10.9836 |

NS denotes non-significant

* denotes significant at 5%

TS1 = 4.5 ± 0.5 ppt : TS2 = 9.5 ± 0.5 ppt
 TS4 = 14.5 ± 0.5 ppt : TS4 = 19.5 ± 0.5 ppt
 AS1 = 5 ppt AS2 = 10 ppt AS3 = 15 ppt
 AS4 = 20 ppt AS5 = 25 ppt

Since complete mortality of the postlarvae was observed in less than 1 ppt test salinity level, it was not considered for analysis.

The individual percentage (x) were transformed into arc sine values ($\text{Sin}^{-1} \sqrt{\frac{x}{100}}$) for analysis.

Table 7. Survival percentage* of 10-12 mm size postlarvae of P. monodon in salinities below 5 ppt over different exposure periods after step-wise acclimation.

| Test Salinity levels (ppt) | Exposure periods (h) | | | | |
|----------------------------|----------------------|-------|-------|-------|-------|
| | 24 | 48 | 72 | 96 | 120 |
| Less than 1 | 93.3 | 66.6 | 63.3 | 59.9 | 46.6 |
| 1 | 93.3 | 86.6 | 83.3 | 73.3 | 59.5 |
| 2 | 96.65 | 96.65 | 93.3 | 90 | 86.65 |
| 3 | 100 | 93.3 | 89.95 | 86.6 | 86.6 |
| 4 | 96.65 | 96.65 | 96.65 | 96.65 | 93.3 |

* Each representative value is the average of two determinations for 15 animals per treatment.

ANOVA - TABLE 7

ANOVA for testing significance of survival of 10-12 mm size of postlarvae of P. monodon in salinity levels (TS) below 5 ppt at different exposure periods (EP) after step-wise acclimation.

| Source | S.S. | d.o.f. | M.S. | F-ratio |
|-----------------|---------|--------|--------|-----------|
| Salinity level | 1868.87 | 4 | 467.22 | 19.58 *** |
| Exposure period | 968.24 | 4 | 242.06 | 10.14 *** |
| Error | 381.87 | 16 | 23.87 | |

TS Means:

| | |
|-----|----------|
| TS1 | 55.23359 |
| TS2 | 63.73681 |
| TS3 | 74.8076 |
| TS4 | 74.6796 |
| TS5 | 73.5596 |

EP Means:

| | |
|-----|---------|
| EP1 | 79.77 |
| EP2 | 71.4076 |
| EP3 | 68.916 |
| EP4 | 65.81 |
| EP5 | 61.1068 |

| | |
|-------------------------------|----------|
| CD 5% for TS means comparison | 6.736164 |
| CD 5% for EP means comparison | 6.736164 |

*** denotes significant at 0.1%

| | | |
|-----------------------|---|-------------|
| TS1 = less than 1 ppt | : | TS2 = 1 ppt |
| TS3 = 2 ppt | : | TS4 = 3 ppt |
| TS5 = 4 ppt | | |

The individual percentages (x) were transformed into arc sine values ($\text{Sin}^{-1} \sqrt{\frac{x}{100}}$) for analysis.

Table 8. Cumulative mortality of postlarvae of *P. monodon* exposed to salinity gradually decreasing from 20 ppt to 0.2 ppt.
(10 numbers per replicate)

| Experimental period (days) | Salinity (ppt) | Replicates | | | Total number of animals present | Cumulative survival % | Cumulative mortality % |
|----------------------------|----------------|------------|----|----|---------------------------------|-----------------------|------------------------|
| | | 1 | 2 | 3 | | | |
| 1 | 20 | 10 | 10 | 10 | 30 | 100 | 0 |
| 2 | 18 | 10 | 10 | 10 | 30 | 100 | 0 |
| 3 | 16 | 10 | 10 | 10 | 30 | 100 | 0 |
| 4 | 14 | 10 | 10 | 9 | 29 | 96.7 | 3.33 |
| 5 | 12 | 10 | 10 | 9 | 29 | 96.7 | 3.33 |
| 6 | 10 | 10 | 10 | 9 | 29 | 96.7 | 3.33 |
| 7 | 8 | 10 | 9 | 8 | 27 | 90.0 | 10 |
| 8 | 6 | 9 | 9 | 8 | 26 | 86.7 | 13.33 |
| 9 | 4 | 8 | 8 | 8 | 24 | 80 | 20 |
| 10 | 2 | 7 | 8 | 8 | 23 | 76.6 | 23.3 |
| 11 | 1 | 6 | 7 | 7 | 20 | 66.7 | 33.3 |
| 12 | 0.75 | 5 | 7 | 7 | 19 | 63.34 | 36.66 |
| 13 | 0.5 | 4 | 5 | 7 | 16 | 53.34 | 46.66 |
| 14 | 0.2 | 3 | 3 | 1 | 7 | 23.34 | 76.66 |

The animals were maintained at each salinity for 24 hours before the salinity was changed and cumulative mortality at each salinity after 24 h was observed.

Table 9. Probit analysis to determine the relation between logarithmic salinity level (dose = X) and mortality (probit response = Y) for the postlarvae of *P. monodon* for Exp. 3.2.4.

| | | |
|--------------------------------------|---|------------|
| Proportion responded among controls | = | 0 |
| Intercept of probit regression line | = | 5.982823 |
| Slope of probit regression line | = | - 1.332194 |
| Weighted mean of log dose (x) | = | 1.262063 |
| Weighted mean of probit response (y) | = | 4.30071 |
| Standard error | = | 1 |
| Standard error of slope | = | .1349906 |

| Percent | Lower limit (ppt) | Lethal dose (ppt) | Upper limit (ppt) |
|---------|-------------------|-------------------|-------------------|
| 30 | 1.801389 | 1.3514 | 1.013819 |
| 50 | 0.7900891 | 0.5459431 | 0.3772408 |
| 70 | 0.3656071 | 0.2205521 | 0.1330478 |
| 90 | 0.1243583 | 0.0595834 | 2.854801E-02 |

Table 10. Mean individual weight* (g) of juveniles of P. monodon surviving 56 days in different test salinities recorded at fortnightly intervals

| Experimental period (days) | Test salinity levels (ppt) | | | | |
|----------------------------|----------------------------|-----------|-----------|------------|------------|
| | 1.5 ± 0.5 | 4.5 ± 0.5 | 9.5 ± 0.5 | 14.5 ± 0.5 | 19.5 ± 0.5 |
| Initial | 0.2483 | 0.1917 | 0.2412 | 0.1974 | 0.2143 |
| 14 | 0.3130 | 0.3384 | 0.3466 | 0.2954 | 0.3583 |
| 28 | 0.4990 | 0.6077 | 0.5376 | 0.4618 | 0.5486 |
| 42 | 0.5186 | 0.804 | 0.7117 | 0.7457 | 0.7998 |
| 56 | 0.6194 | 1.094 | 0.9858 | 0.9485 | 0.9275 |

* Individual values are the means of two determinations

Table 11. Growth rate variations (mg/day) of juveniles of P. monodon surviving 56 days in different test salinities recorded at fortnightly intervals.

| Experimental period (days) | Test salinity levels (ppt) | | | | |
|----------------------------------|----------------------------|-----------|-----------|------------|------------|
| | 1.5 ± 0.5 | 4.5 ± 0.5 | 9.5 ± 0.5 | 14.5 ± 0.5 | 19.5 ± 0.5 |
| 14 | 4.43 | 10.48 | 7.53 | 7.0 | 10.29 |
| 28 | 8.95 | 14.86 | 10.59 | 9.44 | 13.22 |
| 42 | 6.44 | 14.58 | 11.44 | 13.05 | 13.94 |
| 56 | 6.63 | 16.11 | 13.30 | 13.41 | 12.74 |

ANOVA - TABLE 8

ANOVA for testing significance of variations in growth rate of juveniles of P. monodon reared in different test salinities (TS)

| Source | S.S. | d.o.f. | M.S. | F-ratio |
|-----------------|--------|--------|--------|-----------|
| Test salinity | 123.26 | 4 | 30.815 | 18.96 *** |
| Exposure period | 61.66 | 3 | 20.55 | 12.64 *** |
| Error | 19.55 | 12 | 1.625 | |

TS Means:

| | |
|-----|-------|
| TS1 | 6.61 |
| TS2 | 14.01 |
| TS3 | 10.72 |
| TS4 | 10.73 |
| TS5 | 12.55 |

EP Means:

| | |
|-----|-------|
| EP1 | 7.93 |
| EP2 | 14.41 |
| EP3 | 11.89 |
| EP4 | 12.44 |

CD 5% for comparison of TS means 1.965

CD 5% for comparison of EP means 1.7575

*** denotes significant at 0.1%

TS1 = 1.5 ± 0.5 ppt : TS2 = 4.5 ± 0.5 ppt

TS3 = 9.5 ± 0.5 ppt : TS4 = 14.5 ± 0.5 ppt

TS5 = 19.5 ± 0.5 ppt (control)

EP1 = 14 days : EP2 = 28 days : EP3 = 42 days

EP4 = 56 days

Table 12. 56-days survival (%)* of juveniles of P. monodon in different test salinities recorded at fortnightly intervals.

| Experimental period (days) | Test salinity levels (ppt) | | | | |
|----------------------------|----------------------------|-----------|-----------|------------|------------|
| | 1.5 ± 0.5 | 4.5 ± 0.5 | 9.5 ± 0.5 | 14.5 ± 0.5 | 19.5 ± 0.5 |
| Initial | 100 | 100 | 100 | 100 | 100 |
| 14 | 80 | 85 | 100 | 100 | 100 |
| 28 | 70 | 55 | 95 | 100 | 100 |
| 42 | 50 | 55 | 90 | 100 | 95 |
| 56 | 50 | 50 | 90 | 100 | 75 |

* Individual values are the means of two determinations.

ANOVA - TABLE 9

ANOVA for testing significance of 56-day survival of juveniles of P. monodon in different test salinities (TS)

| Source | S.S. | d.o.f. | M.S. | F-ratio |
|--------|----------|--------|---------|-----------|
| TS | 3599.760 | 4 | 899.94 | 9.108 *** |
| EP | 2310.805 | 4 | 577.70 | 5.8468 ** |
| ERR | 1580.895 | 16 | 98.8050 | |

| TS Means: | | EP Means: | |
|-------------------------------|--------|-----------|-------|
| TS1 | 60.046 | EP1 | 90.0 |
| TS2 | 59.59 | EP2 | 80.13 |
| TS3 | 80.04 | EP3 | 72.34 |
| TS4 | 90.00 | EP4 | 66.30 |
| TS5 | 81.416 | EP5 | 62.31 |
| CD 5% for TS means comparison | | 13.39 | |
| CD 5% for EP means comparison | | 13.39 | |

*** denotes significant at 0.10%

** denotes significant at 1%

TS1 = 1.5 ± 0.5 ppt : TS2 = 4.5 ± 0.5 ppt

TS3 = 9.5 ± 0.5 ppt : TS4 = 14.5 ± 0.5 ppt

TS5 = 19.5 ± 0.5 ppt (control)

EP1 = 14 days : EP2 = 28 days : EP3 = 42 days

EP4 = 56 days

The individual percentages (x) were transformed into arc sine values ($\text{Sin}^{-1} \sqrt{\frac{x}{100}}$) for analysis.

Table 13. Experimental values* for initial weight, stocking density, final weight and biomass increase of juveniles of *P.monodon* in growth experiment.

| | Test salinity levels (ppt) | | | | |
|---|----------------------------|---------|---------|----------|----------|
| | 1.5±0.5 | 4.5±0.5 | 9.5±0.5 | 14.5±0.5 | 19.5±0.5 |
| Average initial wt. (g) | 0.2483 | 0.1917 | 0.2412 | 0.1974 | 0.2143 |
| Average final wt. (g) | 0.6194 | 1.094 | 0.9858 | 0.9485 | 0.9275 |
| Survival percentage | 50 | 72.5 | 90 | 100 | 87.5 |
| Stocking density (initial) nos/m ² | 25 | 25 | 25 | 25 | 25 |
| **Biomass increase (g/m ²) | 1.535 | 15.0363 | 16.0505 | 18.7775 | 14.9316 |
| Biomass increase/ day (mg/m ² /day) | 27.410 | 268.504 | 288.40 | 335.31 | 266.635 |
| Average daily gain (mg/day) | 6.6267 | 16.1125 | 13.2964 | 13.4125 | 12.7357 |

* Individual values are the means of two determinations

** Biomass increase (New, 1971) $c = \frac{azy}{100} - bz$

Where a = av. final wt. (g)
 b = av. initial wt. (g)
 y = Survival percentage
 z = Stocking density (nos/m²)
 c = Biomass increase (g/m²)

Table 14. Average (\bar{x}) and standard deviation (S.D.) of water quality parameters monitored during 56-day survival of juveniles of P. monodon in cement cisterns (n = s)

| Parameters | 1.5 ± 0.5 ppt $\bar{X} \pm$ S.D. | 4.5 ± 0.5 ppt $\bar{X} \pm$ S.D. | 9.5 ± 0.5 ppt $\bar{X} \pm$ S.D. | 14.5 ± 0.5 ppt $\bar{X} \pm$ S.D. | 19.5 ± 0.5 ppt $\bar{X} \pm$ S.D. |
|------------------------|-------------------------------------|-------------------------------------|-------------------------------------|--------------------------------------|--------------------------------------|
| Air temperature (°C) | 30±0.63 | 30±0.63 | 30±0.63 | 30±0.63 | 30±0.63 |
| Water temperature (°C) | 27.4±0.49 | 27.6±0.58 | 27.7±0.748 | 27.5±0.45 | 28.75±1.40 |
| pH | 7.44±0.32 | 7.544±0.31 | 7.57±0.32 | 7.402±0.33 | 7.26±0.24 |
| Dissolved Oxygen (ppm) | 5.46±0.70 | 4.64±2.37 | 5.54±0.79 | 4.942±0.68 | 6.48±1.40 |
| Alkalinity (ppm) | 34.75±2.86 | 67.8±17.79 | 65±12.537 | 67.6±15.12 | 65.2±14.96 |
| Salinity (ppt) | 1.202±0.34 | 4.89±0.46 | 10.62±0.67 | 15.16±0.88 | 19.43±0.63 |

Fig. 1 120-h survival (%) of postlarvae of P. monodon in salinities below 5 ppt. The lethal salinity (LC 50) for the postlarvae at a temperature of water between 27 to 29°C is indicated by.*

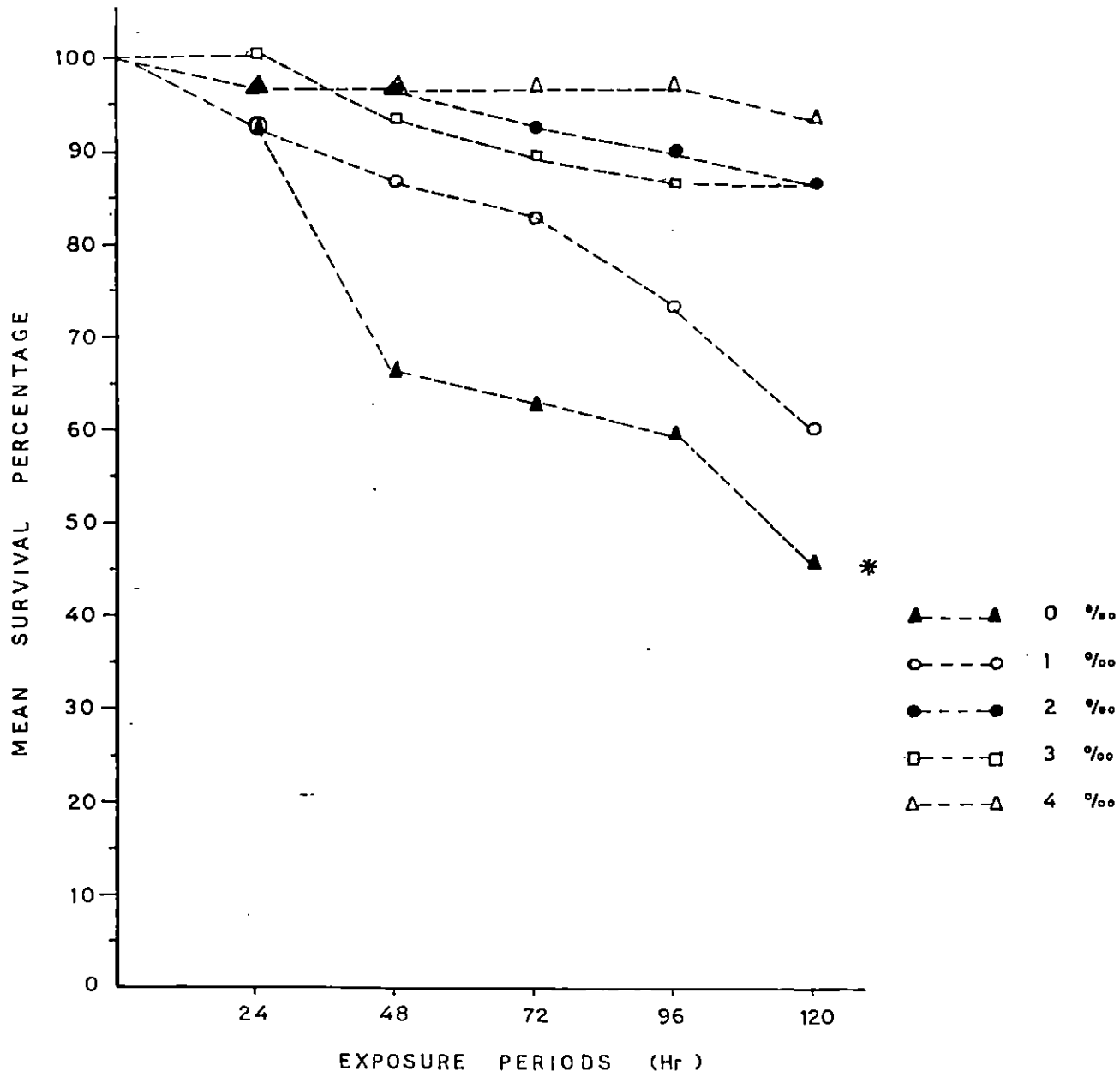


Fig. *1.

Fig. 2 Cumulative mortality (%) of postlarvae of P. monodon exposed to salinity gradually decreasing from 20 to 0.2 ppt.

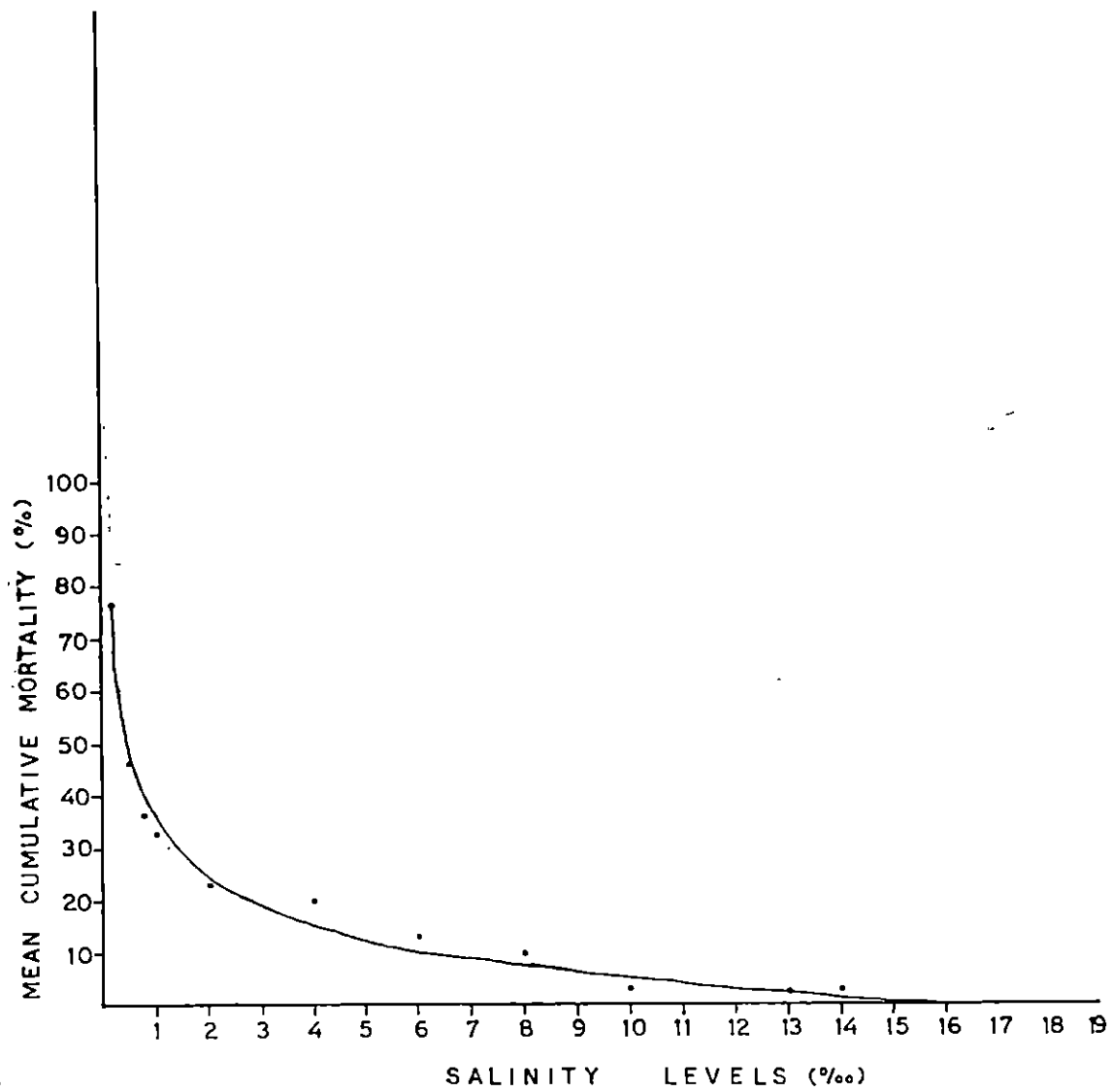


Fig. 2.

Fig. 3 Growth of juveniles of P. monodon in different test salinities (length of experiment : 56 days). Under unrestricted food and constant levels of other growth factors.

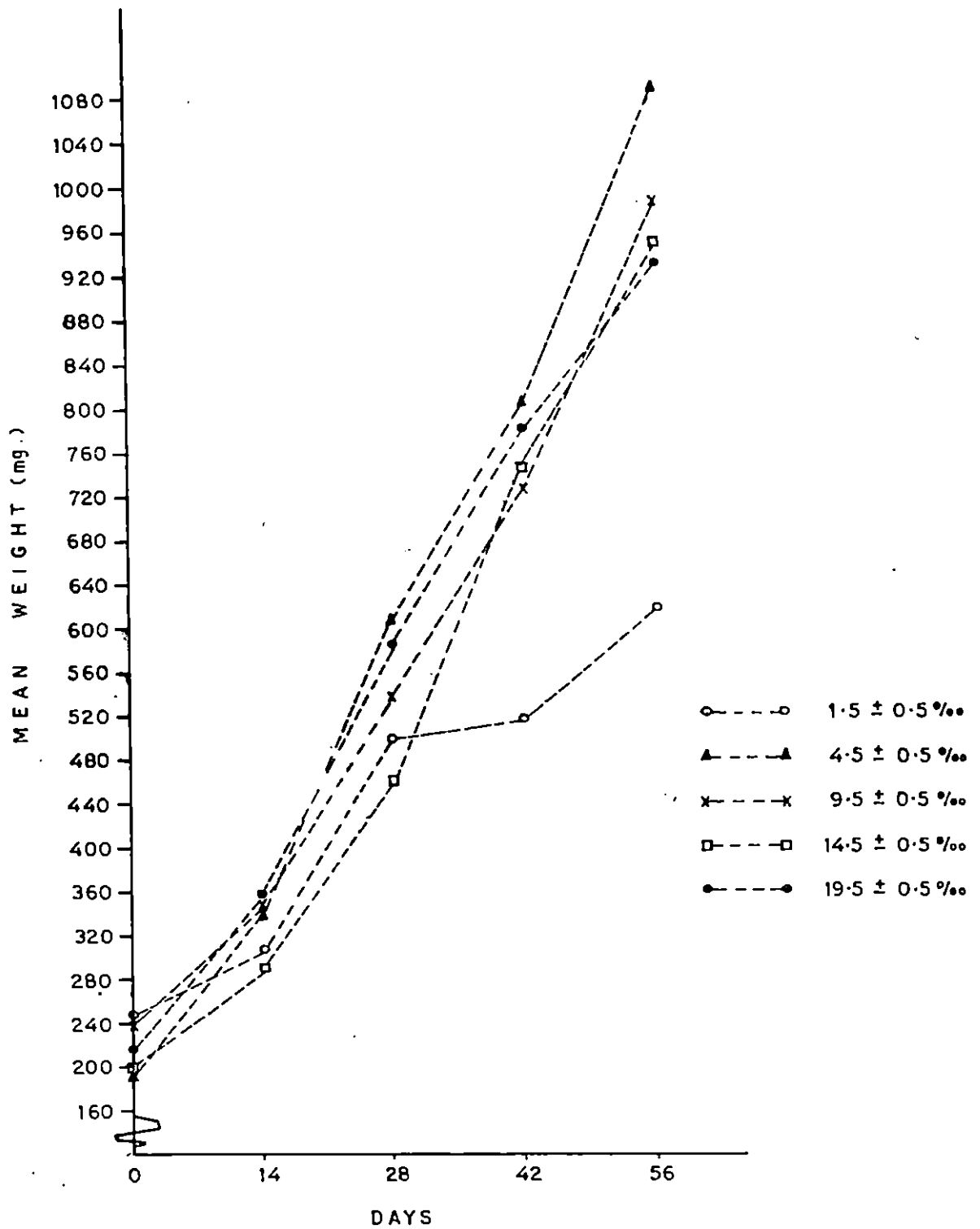


Fig. 3.

Fig. 4 56 - day survival (%) of juveniles of P. monodon in different test salinities. Under unrestricted food and constant levels of other factors. Mean values that differ significantly from the control (19.5 ± 0.5 ppt) are indicated by*

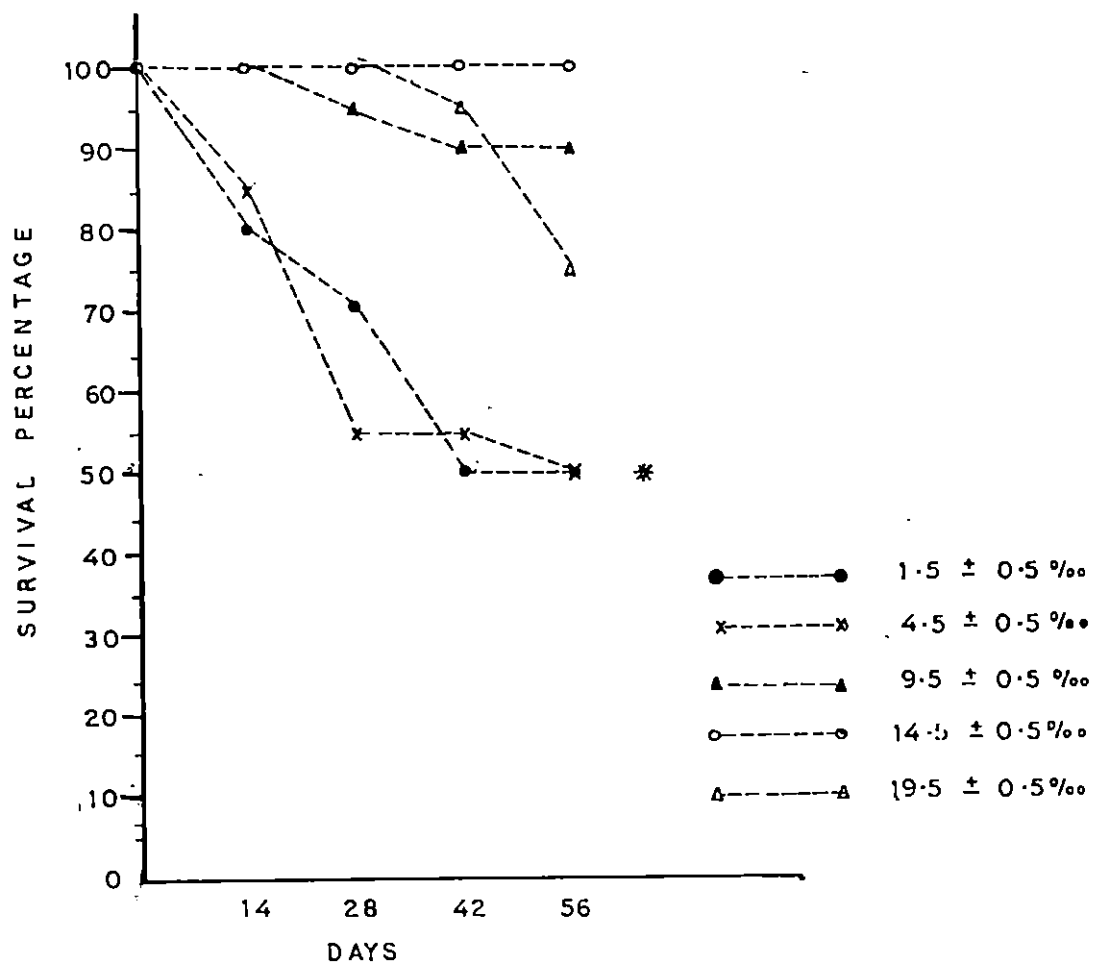


Fig. 4.

Fig. 5 Effect of low salinity on the growth rate of juveniles of P. monodon under laboratory conditions (length of experiment : 56 days). Under unrestricted food and constant levels of other growth factors. Mean values that differ significantly from the control (19.5 ± 0.5 ppt) are indicated by*

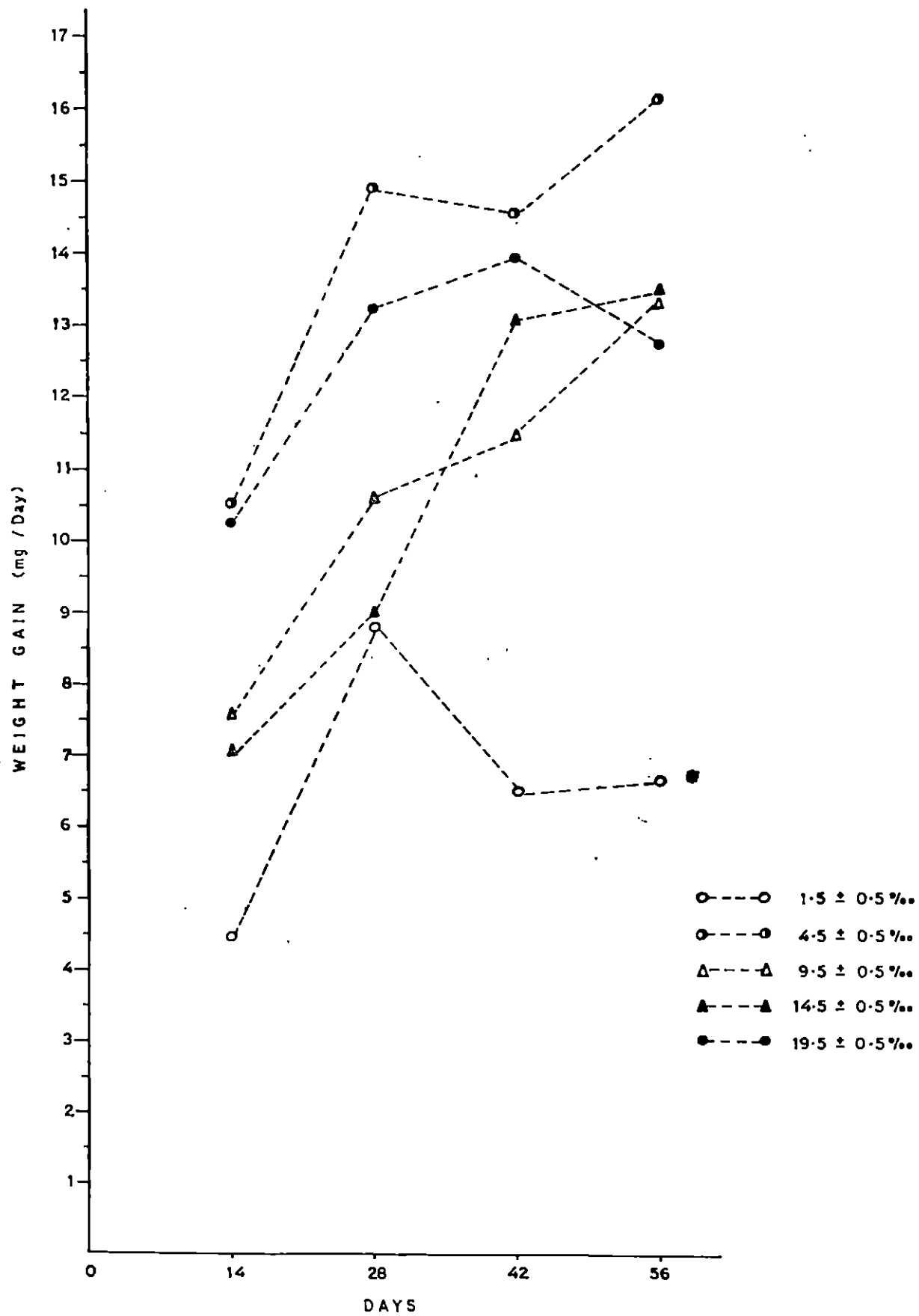


Fig. 5.

Fig. 6 Average growth rate of juveniles of P. monodon surviving 56 days in different test salinities. Mean values that differ significantly from the control (19.5 ± 0.5 ppt) are indicated by*

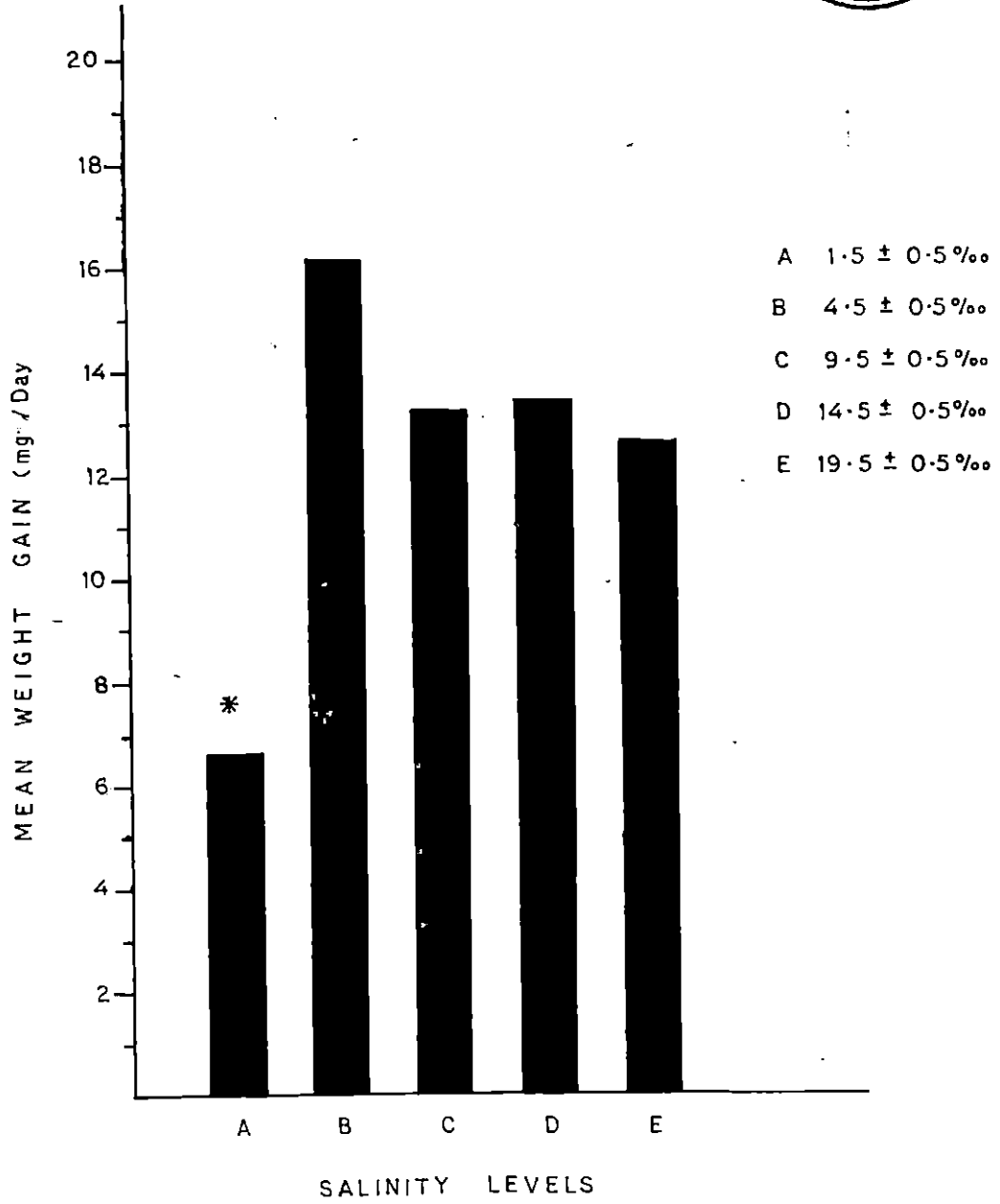


Fig. 6.

Fig. 7 Effect of low salinity on the biomass increase of juveniles of P. monodon in the laboratory (length of experiment : 56 days).

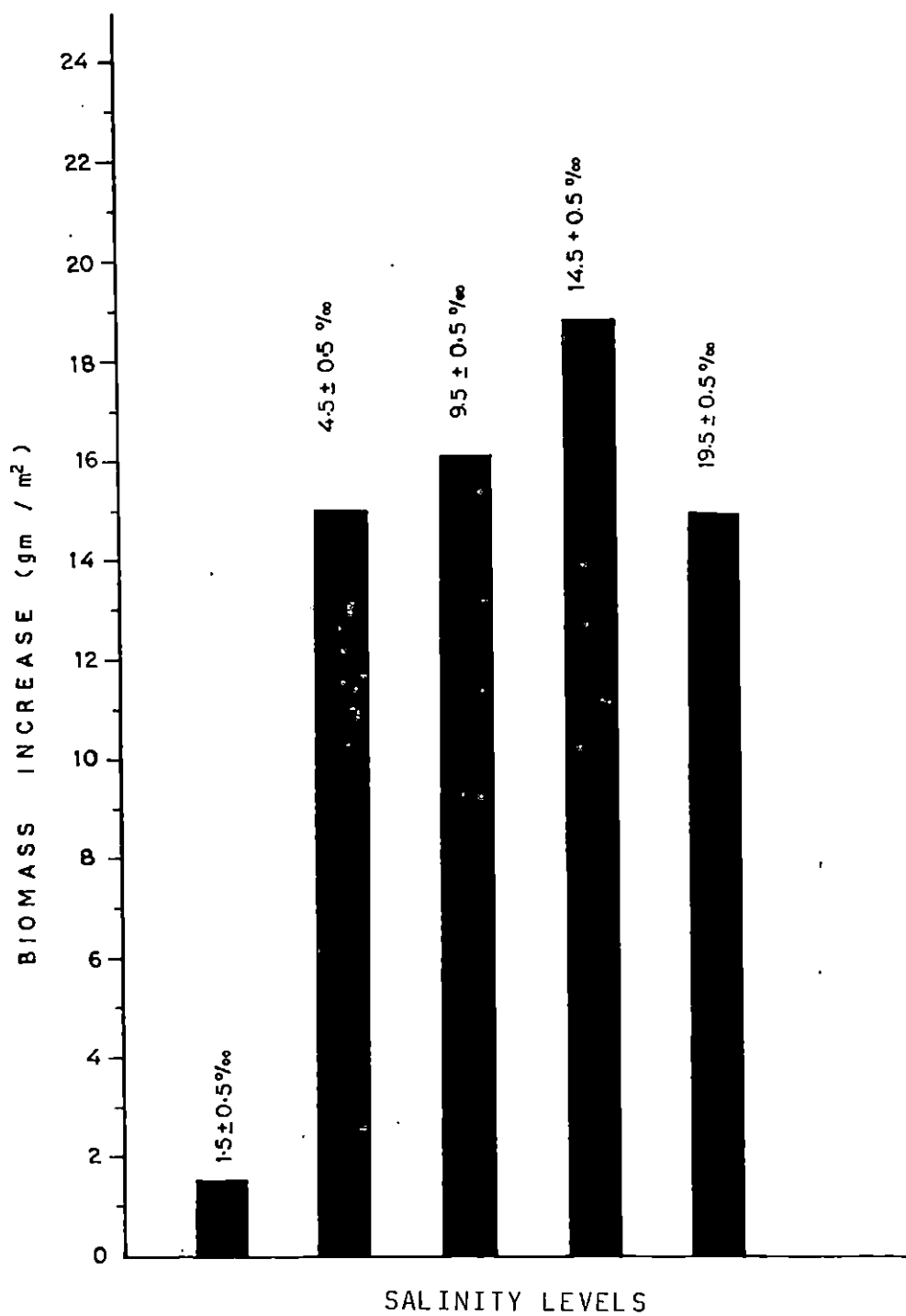


Fig: 7.

Table 15. Model of taste panel evaluation sheet

Sample Date.....

Name of the judge

Kindly evaluate the given coded samples on the basis of your preference by placing the appropriate score against each quality characteristic.

| <u>Quality rating</u> | <u>Scale</u> |
|-----------------------|--------------|
| Excellent | 6 |
| Good | 5 |
| Moderate | 4 |
| Fair | 3 |
| Slightly poor | 2 |
| Poor | 1 |
| Very bad | 0 |

| Quality characteristics | Sample code | | | | |
|-------------------------|-------------|---|---|---|---|
| | A | B | C | D | E |
| Cooked flavour | | | | | |
| Cooked texture | | | | | |
| Overall quality | | | | | |

Table 16. Taste panel evaluation sheet for flavour of cooked meat of P. monodon reared in different test salinities.

| Taste Panel | Samples | | | | |
|-------------|--------------------|------------------|------------------|-------------------|-------------------|
| | A | B | C | D | D |
| | 1.5 ± 0.5 (ppt) | 4.5±0.5 (ppt) | 9.5±0.5 (ppt) | 14.5±0.5 (ppt) | 19.5±0.5 (ppt) |
| 1 | 4 | 5 | 5 | 3 | 2 |
| 2 | 5 | 5 | 5 | 4 | 4 |
| 3 | 4 | 4 | 5 | 2 | 5 |
| 4 | 4 | 4 | 4 | 4 | 3 |
| 5 | 3 | 5 | 6 | 3 | 6 |
| 6 | 2 | 2 | 5 | 1 | 5 |
| 7 | 2 | 1 | 5 | 2 | 4 |
| 8 | 4 | 4 | 4 | 4 | 5 |
| 9 | 4 | 5 | 4 | 4 | 5 |
| 10 | 4 | 4 | 4 | 3 | 5 |

Table 17. Kendall's co-efficient of concordance (Sidney Segel, 1956) to test significance in association among judgements for flavour of cooked meat of shrimps reared in different test salinities.

| Taste panel | Samples | | | | |
|-------------|--------------------|--------------------|--------------------|---------------------|---------------------|
| | A | B | C | D | E |
| | 1.5 ± 0.5 (ppt) | 4.5 ± 0.5 (ppt) | 9.5 ± 0.5 (ppt) | 14.5 ± 0.5 (ppt) | 19.5 ± 0.5 (ppt) |
| 1 | 3 | 4.5 | 4.5 | 2 | 1 |
| 2 | 4 | 4 | 4 | 1.5 | 1.5 |
| 3 | 2.5 | 2.5 | 9.5 | 1 | 4.5 |
| 4 | 3.5 | 3.5 | 3.5 | 3.5 | 1 |
| 5 | 1.5 | 3 | 4.5 | 1.5 | 4.5 |
| 6 | 2.5 | 2.5 | 4.5 | 1 | 4.5 |
| 7 | 2.5 | 1 | 5 | 2.5 | 4 |
| 8 | 2.5 | 2.5 | 2.5 | 2.5 | 5 |
| 9 | 2 | 4.5 | 2 | 2 | 4.5 |
| 10 | 3 | 3 | 3 | 1 | 5 |
| ** | 27 | 31 | 38 | 18.5 | 35.5 |

Kendall's coefficient of concordance (W) = 0.2365*

*denotes significant at 5%

Individual values given are the ranks in ascending order of preference.

**denotes the total of individual ranks in a particular sample.

ANOVA - TABLE 10

ANOVA for testing significance of variation in flavour of cooked meat of P. monodon samples reared in different test salinities.

| Source | S.S. | d.o.f. | M.S. | F-ratio |
|-------------|-------|--------|------|---------|
| Between Tr. | 17.88 | 4 | 4.47 | 3.89 * |
| Error | 51.8 | 45 | 1.15 | |

TR Means

| | |
|----|-----|
| T1 | 3.6 |
| T2 | 3.9 |
| T3 | 4.7 |
| T4 | 3.0 |
| T5 | 4.4 |

CD 5% for Tr means comparison 0.97

* denotes significant at 5%

- T1 = mean panel score for samples at 1.5 ± 0.5 ppt (n=10)
 T2 = mean panel score for samples at 4.5 ± 0.5 ppt (n=10)
 T3 = mean panel score for samples at 9.5 ± 0.5 ppt (n=10)
 T4 = mean panel score for samples at 14.5 ± 0.5 ppt (n=10)
 T5 = mean panel score for samples at 19.5 ± 0.5 ppt (n=10)

Table 18. Taste panel evaluation sheet for texture of cooked meat of P. monodon reared in different test salinities.

| Taste Panel | Samples | | | | |
|-------------|------------------|------------------|------------------|-------------------|-------------------|
| | A | B | C | D | E |
| | 1.5±0.5 (ppt) | 4.5±0.5 (ppt) | 9.5±0.5 (ppt) | 14.5±0.5 (ppt) | 19.5±0.5 (ppt) |
| 1 | 5 | 5 | 5 | 3 | 2 |
| 2 | 4 | 4 | 4 | 2 | 3 |
| 3 | 4 | 4 | 4 | 5 | 5 |
| 4 | 5 | 5 | 4 | 5 | 3 |
| 5 | 4 | 3 | 5 | 3 | 5 |
| 6 | 3 | 3 | 5 | 1 | 4 |
| 7 | 1 | 2 | 5 | 1 | 3 |
| 8 | 5 | 4 | 4 | 4 | 4 |
| 9 | 3 | 4 | 5 | 2 | 4 |
| 10 | 4 | 5 | 4 | 2 | 5 |

Table 19. Kendall's co-efficient of concordance to test significance in association among judgements for texture cooked meat of shrimp reared in different salinities.

| Taste panel | Samples | | | | |
|-------------|--------------------|--------------------|--------------------|---------------------|---------------------|
| | A | B | C | D | E |
| | 1.5 ± 0.5 (ppt) | 4.5 ± 0.5 (ppt) | 9.5 ± 0.5 (ppt) | 14.5 ± 0.5 (ppt) | 19.5 ± 0.5 (ppt) |
| 1 | 4 | 4 | 4 | 2 | 1 |
| 2 | 4 | 4 | 4 | 1 | 2 |
| 3 | 2 | 2 | 2 | 4.5 | 4.5 |
| 4 | 4 | 4 | 2 | 4 | 1 |
| 5 | 3 | 1.5 | 4.5 | 1.5 | 4.5 |
| 6 | 2.5 | 2.5 | 5 | 1 | 4 |
| 7 | 1.5 | 3 | 5 | 1.5 | 4 |
| 8 | 5 | 2.5 | 2.5 | 2.5 | 2.5 |
| 9 | 2 | 3.5 | 5 | 1 | 3.5 |
| 10 | 2.5 | 4.5 | 2.5 | 1 | 4.5 |
| ** | 30.5 | 31.5 | 36.5 | 20 | 31.5 |

Kendall's co-efficient of concordance (W) = 0.147 NS

NS denotes non-significant.

Individual values given are the ranks in ascending order of preference.

**denotes the total of individual ranks in a particular sample.

Table 20. Taste panel evaluation sheet for overall quality of cooked meat of P. monodon reared in different test salinities.

| Taste panel | Samples | | | | |
|-------------|--------------------|--------------------|--------------------|---------------------|---------------------|
| | A | B | C | D | E |
| | 1.5 ± 0.5 (ppt) | 4.5 ± 0.5 (ppt) | 9.5 ± 0.5 (ppt) | 14.5 ± 0.5 (ppt) | 19.5 ± 0.5 (ppt) |
| 1 | 4 | 5 | 5 | 3 | 2 |
| 2 | 5 | 5 | 5 | 3 | 4 |
| 3 | 4 | 4 | 4 | 3 | 5 |
| 4 | 4 | 5 | 4 | 5 | 3 |
| 5 | 3 | 3 | 5 | 3 | 5 |
| 6 | 3 | 2 | 5 | 1 | 4 |
| 7 | 2 | 1 | 5 | 2 | 4 |
| 8 | 4 | 4 | 4 | 4 | 5 |
| 9 | 4 | 5 | 4 | 4 | 5 |
| 10 | 4 | 4 | 4 | 3 | 5 |

Table 21. Kendall's co-efficient of concordance to test significance in association among judgements for overall quality of cooked meat of shrimps reared in different salinities.

| Taste Panel | Samples | | | | |
|-------------|--------------------|--------------------|--------------------|---------------------|---------------------|
| | A | B | C | D | E |
| | 1.5 ± 0.5 (ppt) | 4.5 ± 0.5 (ppt) | 9.5 ± 0.5 (ppt) | 14.5 ± 0.5 (ppt) | 19.5 ± 0.5 (ppt) |
| 1 | 3 | 4.5 | 4.5 | 2 | 1 |
| 2 | 4 | 4 | 4 | 1 | 2 |
| 3 | 3 | 3 | 3 | 1 | 5 |
| 4 | 2.5 | 4.5 | 2.5 | 4.5 | 1 |
| 5 | 2 | 2 | 4.5 | 2 | 4.5 |
| 6 | 3 | 2 | 5 | 1 | 4 |
| 7 | 2.5 | 1 | 5 | 2.5 | 4 |
| 8 | 2.5 | 2.5 | 2.5 | 2.5 | 5 |
| 9 | 2 | 4.5 | 2 | 2 | 4.5 |
| 10 | 3 | 3 | 3 | 1 | 5 |
| ** | 27.5 | 31 | 36 | 19.5 | 36 |

Kendall's co-efficient of concordance (W) = 0.1535 NS

NS denotes non-significant

Individual values given are the ranks in ascending order of preference.

**denotes the total of individual ranks in a particular sample.

Table 22. Salinity preference of some penaeids

| Species | Stage | Salinity (ppt) | Author |
|-------------------------------------|----------------|----------------|-------------------------------------|
| <u>Penaeus aztecus</u> | larvae | 28-20 | Cook (1970) |
| - do - | Juvenile | 32 | Grajcer and Neal (1973) |
| - do - | (13-20 mm) | 8.5-17 | Venkataramalah <u>et al.</u> (1975) |
| <u>P. indicus</u> | Postlarvae | 10 | Nair and Kutty (1975) |
| | Juvenile | 30 | -do- |
| | Postlarvae | 21-28 | Bhattacharya and Kewalramani (1976) |
| | 8-19 mm | 10-25 | Lakshmikantham (1982) |
| | 13-15 mm | 5-35 | -do- |
| | 15-19 mm | 5-40 | -do- |
| | Postlarvae | 20 | Kuttyamma (1982) |
| | Juvenile | 25 | -do- |
| | Juvenile | 5-15 | Raj and Raj (1982) |
| <u>P. japonicus</u> | Embryonic | 27-39 | Takeo Imai (1977) |
| | Early juvenile | 23-47 | -do- |
| <u>P. mergulensis</u> | Postlarvae | 25 | Beard <u>et al.</u> (1977) |
| <u>P. (Melicertus) latisulcatus</u> | Juvenile | 25-45 | Ramaswamy and Pandian (1985b) |
| <u>P. monodon</u> | Juveniles | 15-20 | Hideo (1979) |
| | Juveniles | 10-25 | Manik <u>et al.</u> (1979) |
| | Juveniles | 5-15 | Raj and Raj (1982) |
| | Postlarvae | 7-10 | Rajyalakshmi (1982) |
| | Juveniles | 15-25 | Chen (1984) |
| | Juveniles | 4-20 | Present study |

DISCUSSION

DISCUSSION

5.1. Salinity Tolerance

The interpretations of observations on salinity and shrimp abundance in nature is rather difficult because some factors like temperature, light, substrate, food supply, vegetative cover and pollution vary frequently independent of changes in salinity (Zein-Eldin and Aldrich, 1965). For this reason, controlled environment studies in the laboratory were employed for salinity tolerance studies.

It was observed that for the postlarvae, fluctuations in salinities ranging from 4 to 20 ppt, were not a significant factor ($P \geq 0.05$) influencing the survival (ANOVA Table 6). However, a marked reduction in tolerance to salinity below 4 ppt was observed. A general reduction in survival in salinity levels below 1 ppt on sudden transfer regardless of acclimation salinities suggest a strong salinity effect. (Table 6). The excellent survival of the postlarvae upto 24 h in salinity ranging from 1 to 20 ppt suggested a short term tolerance to this salinity range while the survival in different test salinities for 120 h regardless of acclimation salinities confirmed the wide range of tolerance to salinity ranging from 4 to 20 ppt. Complete mortality was observed at salinity less than 1 ppt. Mortality at this test salinity was presumably the result of the immediate stress caused by changes in the salinity levels. However, the stress of salinity acclimation do not seem to explain the poorer survival (56.6 to 69.95%) of postlarvae between 4 and 10 ppt when they were acclimated at 25 ppt (Table 1).

The higher rate of mortality at low salinity levels can be due to lethal salinity level. According to Manik *et al.* (1979) at low salinity, the shrimps could maintain their osmotic and ionic balance by decreasing the rate of water transport across the gut, as well as the rate of sodium and chloride extrusion through gills. The animal can survive only when the cells are able to tolerate the decreased haemolymph sodium and chloride concentrations. When this becomes too little for

the cells to maintain ionic balance, the shrimps would die. This may be a factor that caused mortality of postlarvae in this trial. Moreover, the stress would make some of the animals weak and susceptible to predation. In culture systems, Ferraris et al. (1987) observed that mortality of the juveniles of P. monodon occurred when moulting coincided with salinity fluctuations. When moulting occurred at very low salinity levels, more energy and time were required to normalize haemolymph osmolarity. This long time interval increased the vulnerability of the animal to cannibalism and prolongs their inability to forage for food.

Chakraborti et al. (1985a) reported a 76 to 100% survival for postlarvae of P. monodon when acclimated to different salinities, viz. 25, 20, 15, 10 and 5 ppt and directly transferred to salinities ranging between 3 to 30 ppt. Survival was between 2 to 4% below 2 ppt. He suggested salinity above 3 ppt should be maintained in brackishwater impoundments during the monsoon since long exposure to salinity lower than 3 ppt causes heavy mortality. Result of the present study also demonstrated a similar effect.

The effect of step-wise acclimation in extending ranges of postlarval tolerance is clearly shown in fig. 1. When the changing rate of salinity was 2 ppt per day while from 2 ppt onwards, it was done at still smaller intervals, 53.34% of postlarvae survived in 0.5 ppt salinity (Fig. 2). Comparing this result with those, when the postlarvae were pre-acclimated at different salinities and then introduced suddenly into the different test salinities (Table 6), it can be seen that a gradual changing condition permitted better survival than did sudden changes. There are similar reports of 40-64% survival of postlarvae of P. monodon in salinity less than 1 ppt when the changing rate of salinity was done 1-2 ppt per day (Motoh, 1981; Chakraborti et al., 1985a). Analysis of probit regression line for the relation between the logarithmic salinity level (dose = X) and mortality (probit response = Y) was found to be $Y = 5.982023 + 1.332194 X$ (Table 9). This implies that lowest lethal salinity (LC 50) for the postlarvae is 0.5459 ppt with its upper and lower

limit being 0.3772 and 0.7901 ppt respectively. LC 30 and LC 70 lethal salinities were 1.3514 and 0.2206 ppt respectively. These results can explain how postlarvae of P. monodon can penetrate into and survive extreme low salinities in the natural environment.

5.1.1. Biological and Ecological Implications.

Commercially important shrimps of the genus Penaeus spawn at the sea, migrate into the estuarine areas as postlarvae, remain in less saline estuaries until they approach maturity and then return to the sea (Subrahmanyam and Rao, 1968; Subrahmanyam and Ganapati, 1975). A number of authors have emphasized the requirement of low salinities by postlarvae and juveniles of Penaeus for better survival and growth in nursery areas (Gunter, 1950; Pearse and Gunter, 1957; Gunter et al., 1964). Preference for reduced salinities over seawater in postlarvae and early juveniles had been demonstrated experimentally for Penaeus vannamei, P. stylirostris, P. californiensis and P. brevisrostris from Western Mexico (Mair, 1980). A few penaeids sp. like P. aztecus, P. setiferus and P. duorarum have been recorded at salinities less than 1 ppt (Gunter and Shell 1959; Tabb et al., 1962; Gunter and Hall, 1963). Large numbers of the postlarvae of P. aztecus were collected from Louisiana waters within 0.8 to 5.99 ppt (Wengert, 1972). Reports on the occurrence of the postlarvae of P. monodon in very low salinity (less than 1 ppt) are available (Manik et al., 1979; Raj and Raj, 1982a). In Marakanam estuary, Bose and Venkatesan (1982) encountered postlarvae of P. monodon in salinity ranging from 1.3 to 2.0 ppt. According to Thampy et al. (1982) owing to its capacity to tolerate low salinity conditions the percentage availability of the postlarvae of P. monodon in the Cochin backwaters could have been more in the monsoon but for the turbidity of water and silting of beds during the rainy season which could be adversely affecting the survival of the postlarval recruitment just prior to the start of the South-West monsoon. George

and Suseelan (1982) reported the postlarvae of this species to thrive well in salinities below 5 ppt and encountered the postlarvae in all gradients of salinity in the Godavari and Konade estuaries along the east coast of India. Thus, the ecological distribution of juveniles and postlarvae of P. monodon correlates well with its ability to osmoregulate in very low salinity levels as obtained in this study.

Temperature influences salinity tolerance in penaeids either independently (Williams, 1960) or in combination with salinity (Zein-Eldin and Aldrich, 1965). Usually salinity tolerance tends to decrease as temperature becomes sub - or supra - optimal (Kinne, 1971). The temperature variation noticed in the present study was 27 to 29°C which is the normal temperature range found in most of the brackishwater nursery grounds. This temperature range is characteristic of tropical regions and is relatively stable in the inshore and estuarine region. This makes it possible for the postlarvae to withstand wide range of salinities in nature.

5.2. Growth Studies

The objective of this study was to determine the range of low salinities (below 20 ppt) for favourable growth of the prawns, which might give an insight into the growth rate under natural conditions. In order to analyse and understand growth phenomenon, it is convenient to consider short growth periods for arbitrarily defined time periods (Webb, 1978).

The result of the growth experiment conducted in the present study indicated that low salinities had a highly significant ($P < 0.001$) influence on the growth of juveniles of P. monodon (ANOVA - Table 8). Juveniles recorded maximum growth rate of 6.63 mg/day in 4.5 ± 0.5 ppt while between 9 and 20 ppt growth rate varied from 12.74 to 13.41 mg/day (Fig. 6). Mean values of growth rates of juveniles between 4 and 15 ppt did not differ significantly ($P > 0.05$) from the

control (19.5 ± 0.5 ppt).

In brackishwater ponds in the Philippines, juveniles of P. monodon grew to about 50 - 100 g under 10 to 20 ppt (Hideo, 1979) while in Indonesia Manik et al. (1979) recorded better growth and production of juveniles of this species in salinity ranging between 15 and 20 ppt. In West Bengal, Verghese et al. (1975) observed a growth rate of 4.36 g/month for this species when salinity of the pond water varied from 5.44 to 36.5 ppt. Reports on the optimum growth of P. monodon during culture in grow - out ponds when exposed in salinities ranging between 5 and 20 ppt (Sundararajan et al., 1979; Sebastian et al., 1980) correlate well with results of this study that optimum growth may be maintained by juveniles of P. monodon over a range of 4 to 20 ppt salinity.

Manik et al. (1979) reported that within the salinity range from 15 to 20 ppt, there was no discernible effect on growth rate of juveniles of P. monodon when food was sufficient. In the present study, mean values of growth rate of juveniles between 4 and 15 ppt did not differ significantly ($P \geq 0.05$) with the control (19.5 ± 0.5 ppt).

Observations of better growth rate in lower salinities had been reported in other penaeids too (Nair and Kutty, 1975; Venkataramaiah et al., 1975).

The superior growth rate and biomass increase obtained in this study for juveniles in salinities between 4 and 20 ppt comparing with those in 1.5 ± 0.5 ppt may be due to their better efficiency of consumption and utilization. Both consumption and utilization of food for growth depend to a lesser extent on salinity concentrations (Paloheimo and Dickie, 1966 a, b; Warren and Davis, 1967). Based on comprehensive studies on P. aztecus Venkataramaiah et al. (1975) observed better food conversion efficiency in 8.5 and 17 ppt salinities compared to higher

salinities. They recorded faster metamorphosis of postlarvae into juveniles in 18.5 ppt than in higher salinities. Better growth of juveniles may also be due to the consistent growth rate associated with higher moulting frequency in salinities between 4 and 20 ppt (Mintardjo et al., 1979 as given in Manik et al., 1979). However, Hag (1984) had noticed that higher salinity (40 to 45 ppt) favoured moulting in P. monodon under laboratory conditions while decreasing salinity did not induce moulting. Salinity also influences the burying behaviour of prawns (Lakshmi et al., 1976). Greater number of shrimps, buried in low (8.5 and 17 ppt) than in higher salinity (Venkataramaiah et al., 1975). The implication is that burying is a means of conserving energy, that can be diverted for growth. Reports of low respiratory rates in low salinities and high rates in high salinities indicated the corresponding low or high energy demands in the respective media (Equasa, 1961; Venkataramalah et al., 1975).

The poor growth rate of juveniles in 1.5 ± 0.5 ppt (Fig. 6) indicated that although the juveniles could be grown in a wide range of low salinities, weight increment was poor in very low salinities. This poor growth rate at very low salinities may be due to the stress undergone and the increase in energy cost due to osmoregulation (Kutty et al., 1971). In culture systems with very low salinity, growth rate of juveniles of P. monodon was very poor because of lesser moulting frequency and smaller growth increment with each moult (Manik et al., 1979). Similar reports of adverse effect of low salinity on growth of juveniles of this species in the laboratory (Subrahmanyam, 1973) and in ponds with saline soil and freshwater are available (Venkatesan and Joshua, 1984).

There are few reports of faster growth rate of P. monodon in culture systems when salinity of pond water was gradually increasing. (Verghese et al., 1982; Chakraborti et al., 1985b). The animals referred to in these cases were larger-sized individuals and belonged to the size-group 13-17 cm. Animals of this size-

groups are on the verge of their spawning migration to the sea (Rao, 1967; Jhingran, 1975). Hence a gradual change to stenohalinity is to be expected alongwith other physiological changes. Faster growth rate observed by Verghese *et al.* (1982) and Chakraborti *et al.* (1985b) at the increasing phase of salinity of pond-water seems to suggest such a gradual change in this species. Gunter *et al.* (1964) pointed out that the adaptation to low salinity is highly developed in younger stages while Panikkar (1968) had indicated that osmoregulation in dilute media is less effective in full-sized individuals, especially in their reproductive phase.

The decrease in survival rate of juveniles held at 19.5 ± 0.5 ppt (Fig. 4) is unexplainable. Mortality was recorded during the latter days of the experiment. If this decreased survival represented a longterm effect of the stress of low salinity it is strange that such a decrease did not occur among groups held at even lower salinities. Besides the preliminary tolerance studies had indicated that the animal survive well at 19.5 ± 0.5 ppt (Table 6).

Based on comprehensive studies on *P. aztecus*, Venkataramaiah *et al.* (1975) stated that although the juveniles of *P. aztecus* survived a wide salinity range, the best growth and survival were observed in optimum salinity of 8.5 and 17 ppt. Kinne (1971) also stated that in most of the euryhaline invertebrates, growth is restricted to a significantly narrower range of salinity when compared to survival. The present study shows that though juveniles of *P. monodon* could tolerate very low salinity upto 1 ppt (Fig. 4) their growth rate was optimal beyond 4 ppt.

5.3. Sensory Evaluations

Flavour of fish and shellfish is derived from extractable compounds which may be divided into two broad categories. The first category contains the nitrogenous compounds which include free amino acids, nucleotides and related compounds. The

second category is comprised of non-nitrogenous compounds such as organic acids, sugars and inorganic constituents. Of the nitrogenous compounds free amino acids predominate the majority of crustaceans. Together with quaternary ammonium bases, free amino acids comprise nearly 90% of the total extractable nitrogen in Molluscan and Crustacean tissues (Konosu and Yamaguchi, 1982).

Free amino acids have been implicated as being responsible for the characteristic flavour of fish and shellfish (Simudu and Hujita, 1954; Hashimoto, 1965). In addition to free amino acids, inorganic ions such as Na^+ and Cl^- have also been found to contribute significantly to the flavour of marine products (Hayashi *et al.*, 1981). In penaeid shrimp free amino acids (Mc Coid *et al.*, 1984) as well as Na^+ and Cl^- (Castille and Lawrence, 1981) have been shown to influence osmoregulation. Since these same compounds impart flavour characteristic of marine food products, manipulation of the flavour intensity in penaeid shrimp may be possible through changes in environmental salinity.

In the present study a significant difference ($P \leq 0.05$) in flavour was observed in the shrimp meat with salinity variations (ANOVA Table 10). The overall taste panel score for flavour of cooked meat of shrimps held at 9.5 ± 0.5 ppt was better than those grown at lower salinity levels (Table 16). The significant difference in this flavour intensity can possibly be associated with variations in the tissue free amino acid concentration.

Free amino acids were reported to vary with salinity in the muscle and hepatopancreas of *Penaeus kerathurus* (Richard and Ceccaldi, 1974). Muscle and blood of crabs and lobsters became enriched with free amino acids when the animals transferred from brackishwater to full marine water (Duchateau and Florkin, 1961). Larserre and Gilles (1971) observed an immense decrease in muscle free amino acid content of euryhaline intertidal teleosts *Crenimugil labrosus* and

Paralichthyes lethostisma upon transfer from seawater to freshwater. A higher amount of free amino acids was observed in P. vannamei held at higher salinities than those held at relatively lower salinities (Papadopoulos and Finne, 1986).

Free amino acids especially glycine have been shown to be a major contributing factor to the sweet taste of shrimp. In taste panel observations in Metapenaeus dobsoni Rangaswamy et al. (1970) found that glycine contributed to the sweet flavour, where as glutamic acid, leucine and proline conferred a general desirable flavour. Konosu et al. (1960) reported a flavour enhancing effect when free amino acids other than histidine in combination with inosinic acid were added to dried bonito tuna preparation. Glycine is responsible for the sweet flavour of fresh shrimp (Simudu and Hujita, 1954; Hashimoto, 1965) where as Histidine contributed to a 'meaty' characteristic flavour (Simudu et al., 1953). However, in the present trial no attempt was made to analyse the variation in individual amino acid composition in the different samples. The general preference stated by the taste panel for samples at 9.5 ± 0.5 ppt can possibly be due to the increase in their concentration of flavour enhancing amino acids. However, a low score was described for flavour of cooked meat of shrimps held at 14.5 ± 0.5 ppt (Table 16). It has been observed that although the total free amino acid content increased with increasing salinities, the relative contribution of individual free amino acid to the total amino acid pool was not constant. Molar percentage of glycine and serine/threonine varied randomly with salinity, with the concentration of arginine and proline inversely related to salinity (Mc Coid et al., 1984). Hence the lower preference by the taste panel for samples at 14.5 ± 0.5 ppt can be justified on the basis of inference stated by Mc Coid et al. (1984).

Similar observations of variation in flavour intensity in P. vannamei with variation in salinity have been reported by Papadopoulos and Finne (1986). They

observed a significant flavour difference between shrimps held at 10 and 50 ppt and not at 10 and 30 ppt. However, in the present trial, a significant flavour difference was observed between 1.5 ± 0.5 and 9.5 ± 0.5 ppt: 9.5 ± 0.5 and 14.5 ± 0.5 ppt and 14.5 ± 0.5 and 19.5 ± 0.5 ppt (ANOVA Table 10).

The present study throws light on the possibility of the influence of environmental salinity on the flavour of cultured penaeid shrimp. Since free amino acids are major osmoeffectors in shrimps and also primary flavour producers in marine products, "more flavourable" shrimp can be produced by manipulating environmental salinity. Although priority factor in prawn culture is promoting growth and adding to total production, salinity manipulations in order to improve flavour may be done only within permissible limits.

SUMMARY

SUMMARY

The effect of low salinity upon growth and survival of postlarvae/ juveniles of P. monodon (Fabricius) was studied under controlled conditions. Test salinities ranged from 1 to 20 ppt.

6.1. Tolerance Studies

1. Large fluctuations in salinities ranging from 4 to 20 ppt did not influence the survival of postlarvae significantly ($P \geq 0.05$).
2. Postlarvae showed excellent survival for 24 h in salinities ranging from 1 to 20 ppt suggesting a short-term tolerance to a wide range of salinity.
3. Complete mortality of postlarvae at salinity less than 1 ppt within 24 hours regardless of acclimation at 5, 10, 15, 20 or 25 ppt suggested a strong salinity effect.
4. Step-wise acclimation extended the range of postlarval tolerances. Such a gradual change permitted better survival at 'critical levels' of salinity (less than 4 ppt).
5. A probit regression line for the relation between the logarithmic salinity level (dose = X) and mortality (probit response = Y) was determined as $Y = 5.982023 + 1.332194 X$. The lowest lethal salinity (LC 50) at normal conditions of temperature for the postlarvae was 0.5479 ppt with its upper and lower limit as 0.377 and 0.790 ppt respectively.
6. Postlarvae exhibited abnormal behaviour at salinities less than 1 ppt. Feeding and excretion of faecal matter was low at this salinity. Abnormal swimming behaviour such as jumping out, swimming side-wise, spiral and circular movements was also exhibited
7. A higher rate of cannibalism was observed at suboptimal salinities.

6.2 Growth Studies

8. Within the range of 4 to 20 ppt, salinity had no significant effect ($P \geq 0.05$) on the growth rate of the juveniles. Below 4 ppt salinity, growth rate of the juveniles was retarded ($P < 0.001$).
9. Eventhough postlarvae survived 0.5 to 20 ppt salinities growth was restricted to 4 to 20 ppt range.

6.3 Sensory Evaluation

10. Flavour of cooked meat of shrimps differed significantly ($P < 0.05$) with variation in salinity.
11. Overall taste panel score for flavour of cooked meat of shrimps held at 9.5 ± 0.5 ppt was better than those grown at lower salinity levels.
12. The possibility of the influence of salinity on flavour of cultured shrimps is suggested.

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ABSTRACT

Growth and survival of P. monodon are influenced by a number of ecological factors (Chakraborti et al., 1986) salinity, ^{being} ~~is~~ one of the most important of them. Knowledge on the tolerance of this species to low salinity conditions has great implications in its culture especially in extending its culture to low saline fields.

In the present study, the postlarvae were subjected to wide fluctuations in salinity ranging from 1 to 20 ppt. The influence of gradual acclimation in improving ranges of tolerance of postlarvae was studied. The lowest lethal salinity (LC 50) was determined by step-wise reduction of salinity. Salinity in which more than 50% mortality occurred [✓]_^ at the end of 120 h was considered lethal. Growth of juveniles in different test salinities ranging from 1 to 20 ppt for a period of 56 days was undertaken.

Preliminary tolerance studies indicated that fluctuations in salinity ranging from 4 to 20 ppt did not influence the survival of postlarvae significantly ($P \geq 0.05$). Gradual acclimation extended the range of tolerance. A probit regression line for the relation between the logarithmic salinity level (dose=X) and mortality (probit response=Y) was found to be $Y = 5.982023 + 1.332194X$. The lowest lethal salinity (LC 50) was 0.5479 ppt with its upper and lower limits being 0.377 and 0.790 ppt respectively. Postlarvae exhibited abnormal behaviour and higher rate of cannibalism at suboptimal salinities.

Low salinities had a highly significant influence on the growth of juveniles ($P < 0.001$). However, mean values of growth rate between 4 and 15 ppt did not differ significantly ($P \geq 0.05$) from the control (19.5 ± 0.5 ppt).

Shrimps reared in different test salinities were subjected to sensory evaluation. Flavour of cooked meat of shrimps differed significantly with variation in salinities ($P \leq 0.05$). The overall taste panel score was highest at 9.5 ± 0.5 ppt. The possibility of the influence of salinity on flavour of cultured shrimps is suggested.

A simple handwritten checkmark or tick mark, consisting of a single continuous line that starts at the bottom left, curves upwards and to the right, and then curves back down to the right.