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# ACCLIMATION OF DIFFERENT POSTLARVAL STAGES OF *PENAEUS KERATHURUS* (FRÖSKAL, 1775) TO LOWER SALINITY LEVELS

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**Key words:** Acclimation. salinity. *Penaeus kerathurus*. post-larval stages.

## ABSTRACT

Lower salinity acclimation of hatchery-reared *Penaeus kerathurus* postlarvae was investigated in short and long term experiments at PL20, PL30, PL40 and PL50. In short term experiment, the salinity tolerance was determined with/without acclimation. In tolerance test without acclimation, PLs were transferred suddenly from 36 ppt to 2, 5, 10, 20 and 30 ppt. In tolerance test with acclimation, PLs were acclimated over four days from the reared salinity (36 ppt) to 2, 5, 10 and 20 ppt. After acclimation, PLs were transferred from acclimated salinities to lower and higher salinities. In both tolerance testes, PLs were kept for 96 hr. It was found that acclimation has a significant effect in some cases at PL30 and PL40. In both tolerance tests. the highest salinity tolerance (80-90 % survival rate) was attained at PL50 in salinity range of 10-36 ppt. In long term growth experiment. PLs were reared at 5. 10, 20 and 36 ppt for 30 days using minced clam meat. The highest specific growth rate (SGR), survival and production were recorded at the salinity range of 10-20 ppt.

## INTRODUCTION

Low and meagre natural shrimp production suffer an evident shortage in filling the gap of local market demand. Thus, great attention was paid toward shrimp culture. The site unavailability problem is considered the main obstacle hindering the shrimp culture in Egypt and other parts of the world. As a result, many countries adopt low-salinity shrimp culture such as Thailand (Flaherty and Vandergeest, 1998), Indonesia (Kusmastanto *et al.*, 1998) and USA (Saoud *et al.*, 2002). In Egypt, Abdel Rahman *et al.* (2002) examined the profit of this trend with one of penaeid shrimp species in Egyptian waters (*Penaeus japonicus*). They concluded that *P. japonicus* postlarvae successfully tolerate a wide range of low salinity especially after acclimation. The available litterature in this regard was focussed on *P. vannamei* and *P. monodon* as being the main cultured species in low saline water but they are not endemic in Egyptian waters. So, there is a need to study the possibility of low saline shrimp culture with other economical and valuable species which are endemic in Egyptian waters such as *P. kerathurus*.

It has been obserued that in sea water, the total concentration of amino acids is greater in the muscle than in the hepatopancreas but the latter contains much more of the amino acids which are considered as essential in crustaceans, while the conc. off amino acid decreased in muscle more than hepatopancpeas when the medium is diluted. The role played by amino acid pool in the osmoregulation seems to be greater within the range of salinity 23-28 pt. Richard & Ceccaldi (1974).

Information regarding the salinity tolerance of *P. kerathurus* postlarvae is yet scanty. Mourente and Rodriguze (1997) reported that the total length and dry weight values tended to be greater in *P. kerathurus* postlarvae at 35 ppt than those reared at 25 ppt independent of the diet used. Besides, several studies have demonstrated that salinity tolerance can be modified by acclimation process (Venkataramiah *et al.*, 1974; Cawthorne *et al.*, 1983: Kumlu and Jones, 1995).

The postlarval quality stocked in nurseries and grow-out ponds and their tolerance to environmental conditions are of major considerations in shrimp culture. Rearing postlarvae in water that is not fully controlled can influence the development of various organs in this critical stage (Lin and Gary, 1996). So, studying the salinity tolerance on postlarval stages are of great importance.

The aim of the present study is to investigate the salinity tolerance of different postlarval ages with/without acclimation and the growth of acclimated postlarvae to different low salinities.

## MATERIALS AND METHODS

This study was conducted at the Invertebrate Aquaculture Laboratory in National Institute of Oceanography and Fisheries " NIOF" using facilities of the shrimp hatechery at Alexandria.

Wild mated females of *Penaeus kerathurus* were caught at the Gulf of Abu-Quir (northern Alexandria) during the spawning season.

Spawning and larval rearing techniques were done using a modified Japanese method (Abdel Rahman. 1993). When postlarvae (PL) reached the age for experimentation. they were transferrd from the outdoor cement tanks to indoor experimental round glass tanks. Initial measurments (carapace length, total length and total body weight) in each experimental age were obtained from samples of 10 postlarvae withdrawn from the same population.

Low salinity levels were obtained by mixing dechlorinated tap water with natural filtered seawater. Salinity was measured with an optical refractometer (Atago. 1 ppt precision). For all testes. each container was supplied with gentle aeration.

### A- Short term experiment:

#### I-Tolerance test without salinity acclimation:

PL20, PL30, PL40 and PL50 were 0.28, 0.35, 0.47, 0.59 cm mean carapace length. and 0.005, 0.007, 0,02, 0.05 gm average weight, respectively. They were directly transferred from 36 ppt to 2, 5, 10, 20, 30 and 36 ppt and kept for 96hr. Each test was performed with 20 postlarvae in a 5L round glass container. The water was exchanged daily. Animals were counted and dead ones were removed after 1, 2, 3, 6, 9, 12, 24, 48, 72 and 96hr.

#### II- Tolerance test with salinity acclimation:

PL15, PL25, PL35 and PL45 were gradually acclimated to salinity levels from 36 ppt to 2, 5, 10 and 20 ppt. The acclimation scheme was extended over four days. The acclimation period per day was 8 hours. The fraction of fresh water added was calculated as follows:

Fraction of fresh water needed= $V - V\ (S_{final} / S_{initial})$

$V$ = Volume of desired final salinity

$S_{initial}$ = Initial salinity in the tank

$S_{final}$ = Desired final salinity

After the PL20. PL30, PL40 and PL50 were acclimated to the desired salinity levels, they were kept for 16hr at each salinity. Another salinity tolerance was conducted after acclimation to evaluate the acclimation effect. In this test, twenty postlarvae were directly transferred from each acclimated group to 2, 5, 10, 20, 30 and 36 ppt and kept for 96hr. The control was 36 ppt at the ambient temperature (1± 25°C).

### B- Long term growth experiment:

The current experiment was conducted with low-salinity acclimated postlarvae PL30 at 5, 10 and 20 ppt. The control was 36 ppt. and the ambient temperature was 1± 25°C. Postlarvae were fed twice daily with chopped clam meat *ad libtum.* Unlimited feeding was maintained by daily observation. The water was exchanged every two days at a rate of 30%. Survival, specific growth rate (SGR) and production were calculated as follows:

Survival rate = Number of survived postlarvae/number of initial postlarvae ×100

Specific growth rate (SGR) = Ln (final weight)- Ln ( initial weight) / Time (day)

Production = Average gain in body weight × number of survived postlarvae.

## RESULTS

### A- Short term experiment:

#### I -Tolerance test without salinity acclimation:

The salinity tolerance is expressed as survival rate percentage. It was found that the salinity tolerance increased as either postlarval stage and transferred salinity increased. There was no salinity tolerance at all tested postlarval stages after sudden transfer from 36 ppt to 2 ppt. The salinity tolerance of PL20 was 0% after transfer to 2, 5 and 10 ppt, then increased markedly to 60%, 76% after transfer to 20 ppt, 30 ppt, respectively. At PL 30, the salinity tolerance increased from 0% after transfer to 2 and 5 ppt to be 10% and 80% after transfer to 10 ppt and 20 ppt, respectively. The trend of salinity tolerance at PL40 was similar to that at PL50, but with the highest values at PL50. The highest salinity tolerance among all tested salinities was observed at 20 ppt (Fig. 1).

#### II- Tolerance test with salinity acclimation:

The positive effect of acclimation on enhancing the salinity tolerance was clearly observed for PL20 and PL30 more than for PL40 and PL50. At PL20 and PL30, the transfer from any acclimated groups to 2 ppt has no effect in improving the salinity tolerance. However, the acclimation was found to duplicate the salinity tolerance values after transferring the acclimated postlarvae at 10 ppt and 20 ppt to lower salinities (Figs. 2& 4).

In transferring acclimated PL20 and PL30 to higher salinities, the acclimation effect was only recorded in transferring from acclimated groups at 2 ppt and 5 ppt to 5, 10 and 20 ppt (Figs. 3 & 5).

The salinity tolerance of PL40 increased at 2 ppt and 5 ppt from 0% before acclimation to 40% after acclimation. Meanwhile there was no effect of acclimation on increasing the salinity tolerance after transferring of any acclimated group to 10 ppt (Fig. 6).

The acclimation has no effect at PL40and PL50 on improving the salinity tolerance after transferring postlarvae from acclimated groups to higher salinities (Figs. 7 & 9).

The acclimation has minor effect on salinity tolerance of PL50 since salinity tolerance increased at 2 ppt from 0% to 20% and from 10% at 5 ppt to 34% (Fig. 8).

**B- Long term growth experiment:**

It was found that survival rate increased with increasing the salinity within the range of 5-20 ppt. The survival rate at 36 ppt was similar to that at 5 and 10 ppt., while the highest survival rate was attained at 20 ppt (Fig. 10).

The highest specific growth rate (SGR) was observed at salinity range of 10-20 ppt. The SGR at 36 ppt was the lowest among other tested salinities (Fig. 11).

The production trend was similar to that of survival rate. The highest production was recorded at the salinity range of 10-20 ppt. while the poorest one was at 36 ppt.

## DISCUSSION

The salinity tolerance of *Penaeus kerathurus* postlarvae was found to be directly proportional with age within the tested range (PL20-PL50) and the highest salinity tolerance was recorded at PL50. This is strongly conflicting with the findings of other investigators in salinity tolerance of penaeid species. Abdel Rahman *et al.* (2002) showed that salinity tolerance of *P. japonicus* postlarvae increased from PL10 to PL20 to reach the maximum at PL30 then decreased at PL40 and PL50. Tsuzuki and Cavalli (2000) reported that salinity tolerance of *Farfantepenaeus paulensis* postlarvae increased at PL10-PL30 to reach the highest at PL40 then decreased at PL50-PL80. Also, *P. indicus* postlarvae sustained very high survival from PL20-PL45 but it started to decline sharply after PL45 (Kumlu and Jones. 1995). The differences in salinity tolerance trend of *P. kerathurus* from other penaeid species may be due to having fully developed osmoregulatory structure in late postlarval age (PL50) as compared to other species.

Regardless of the postlarval age, the salinity tolerance of *P. kerathurus* postlarvae at 20 ppt was higher than that at 36 ppt which is considered the control. This was recorded previously in other penaeid species since their optimum salinity was not equivelant to that of natural habitat ( > 36 ppt). Abdel Rahman *et al.* (2002) observed that the salinity range of 20-30 ppt was the optimum for *P. japonicus* postlarvae. Similarly, Liu (1983) reported that 20-30 ppt is the optimal salinity range for *P. chinensis*, while the optimal salinity for culturing *P. vannamei* was found at the range of 15-25 ppt (Boyed, 1989). Besides, Roases *et al.* (1997) found that the optimum salinity for *P. setiferus* postlarvae was 7between 5-15 ppt. However, Kumlu *et al.* (2001) reported that the optimum salinity for *Metapenaeus monoceros* postlarvae appeared to be between 30-40 ppt at 28°C.

Acclimation was reported to have marked effect on salinity tolerance; Venkataramiah *et al.* (1974) stated that salinity tolerance appears to be dependent on salinity and temperature background. In addition, Raj and Raj (1982) recorded superior growth and survival for *P. indicus* at low salinities (5-25 ppt) following 7-days acclimation period. Tsuzuki and Cavalli (2000) showed that acclimation of *Farfantepenaeus paulensis* to different salinities decreased the mortality rates to 15% or less. In the present results, the effect of acclimation in reducing the mortality rate was more observed in PL20 and PL30 than PL40 and PL50. The mortality rate of PL20 at 10 ppt decreased from 100% without acclimation to 43% after transferring from acclimated groups at 20 ppt, while Abdel Rahman *et al.* (2002) found that the mortality of *P. japonicus* at PL20 at 10 ppt decreased from 23% before acclimation to 0% after acclimation. This significant difference between *P. kerathurus* and *P. japonicus* in salinity tolerance and effect of acclimation indicates that *P. kerathurus* postlarvae has no high ability to osmoregulate in low saline water as compared to *P. japonicus* postlarvae.

The transfer to higher salinites after acclimation of *P. kerathurus* postlarvae has lesser effect on reducing the mortality rate as compared to transferring to lower salinites. It was found that the transferring to higher salinites enhance the survival at PL20 and PL30, while it has no effect at PL40 and PL50. At PL20 and PL30, the effect of acclimation appeared only after transferring to 5 ppt and 10 ppt from acclimated groups at 2 ppt and 5 ppt. This may be due to deficency to osmoregulate as the difference increased between the acclimated salinity and transferred one. However, Abdel Rahman *et al.* (2002) reported that the acclimation decreased the mortality rate either after transferring of PL10-PL50 from acclimated groups to lower salinites or higher salinites. This confirms again that *P. kerathurus* postlarvae are weak osmoregulator as compared to these of *P. japonicus*.

The present results showed that the specific growth rate of *P. kerathurus* postlarvae at 10-20 ppt was higher than that of the control (36 ppt) 8-d old post larvae of *P. kerathurus* may have sufficiently developed osmoregulatory capabilities to resist 25 ppt under good conditions (Mourente and Rodriguez, 1997), they reported that the total length and individual dry weight values tended to be greater in *P. kerathurus* postlarvae at 35 ppt than those reared at 25 ppt independent of the diet used. This confliction was previously reported in other penaeid species due to strain differences. Bray *et al.* (1994) reported that *P. monodon* and *P. semisulcatus* strains cultured in the Middle East seem to grow well at high salinity, while Taiwanese strains do not. Besides, Kumlu and Jones (1995) concluded that Red Sea *P. indicus* postlarvae survive and grow better in high saline conditions whereas Indian *P. indicus* prefer lower salinities.

Similar to *P. kerathurus* postlarvae, the optimum salinity range for growth of *P. japonicus* was 20 ppt (Rizk *et al.*, 2002). In addition, Huang (1983) reported that *P. vannamei* grew better at 20 ppt than at 5 ppt and 45 ppt. Lumare and Villami (1974) observed that *P. kerathurus* grew quickly in brackish water as long as the temperature was high.

The optimum salinity range that yielded the highest survival rate after short term exposure (4 days) to different low salinities was confirmed after long term exposure (30days) being 10-20 ppt. In *P. japonicus* postlarvae, Rizk *et al.* (2002) found that the highest survival ( > 90%) after 30days was obtained at combination of salinity and temperature range of 25-35 ppt and 20-27°C, respectively.

The production trend of *P. kerathurus* postlarvae was similar to that of the survival rate. The highest production was found at 10-20 ppt, while the poorest one was at 36 ppt. Low production at 36 ppt could be direct result of low growth rate and moderate survival rate. This further confirms that the natural salinity may not be equivelant to the optimum salinity. Contrarily, Rizk *et al.* (2002) reported that the highest biomass of *P. japonicus* after 30 days was attained at higher salinities (20-36 ppt).

It can be concluded that *P. kerathurus* postlarvae can be reared at low saline water within the range of 10-20 ppt. so it recommended to release acclimated PL20 and PL30 in ponds at the salinity range of 10-20 ppt. while PL50 is the best age to release without acclimation in ponds with 10-36 ppt.

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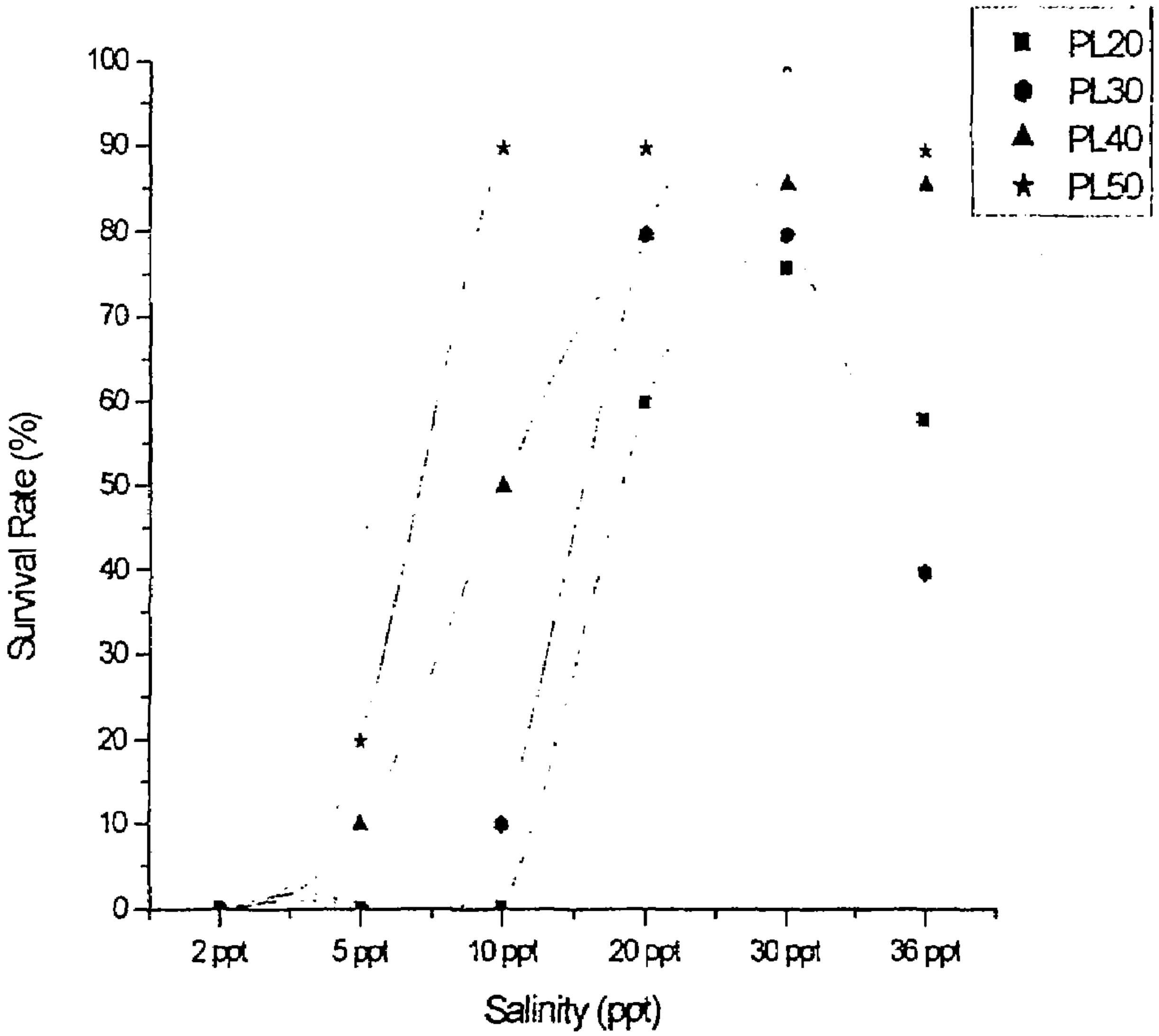

Fig. 1. Survival rate of *P. kerathurus* postlarvae of PL20, PL30, PL40 and PL50 after abrupt change to different salinities.

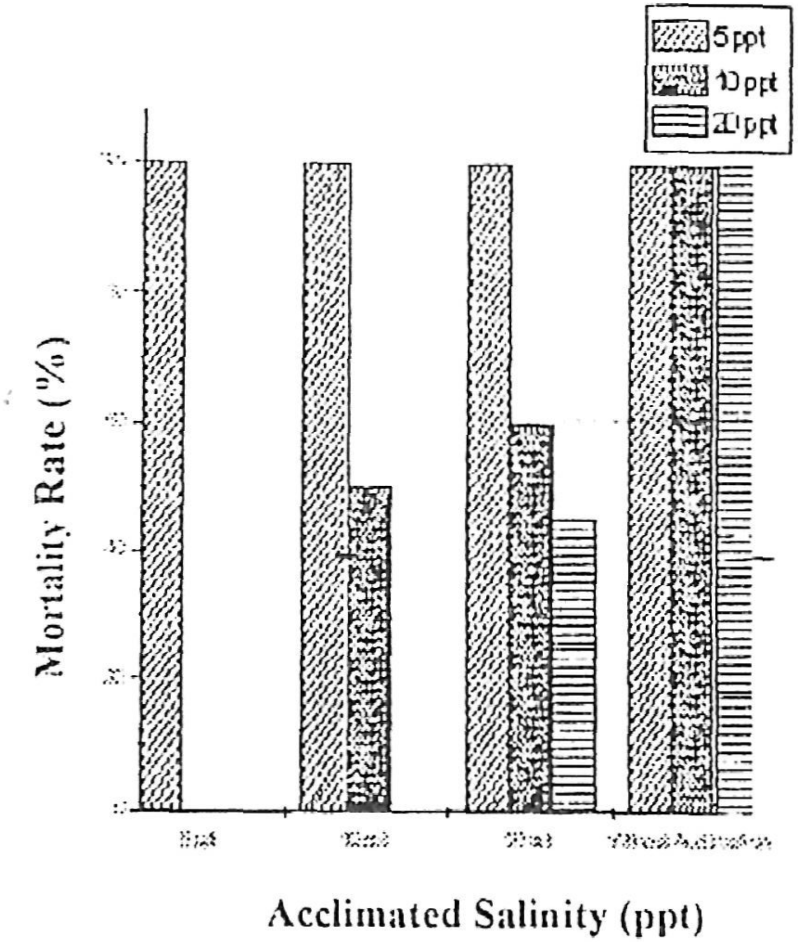

Fig. 2. Mortality rate of *P. kerathurus* PL20 acclimated to 2, 5, 10 and 20 ppt and transferred to lower salinity.

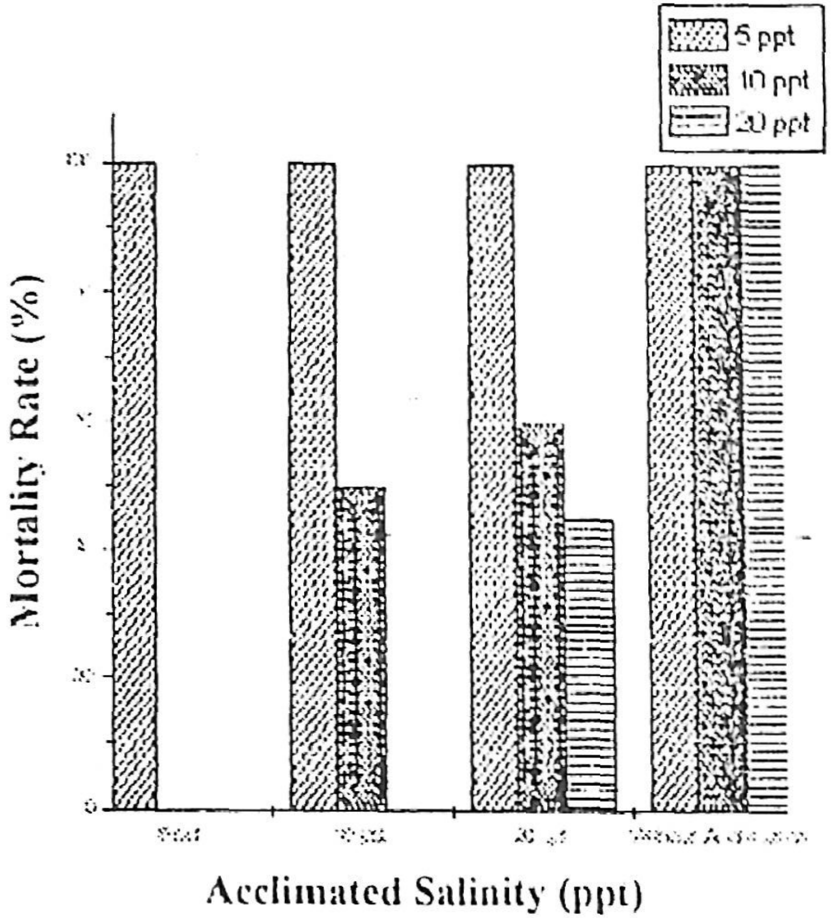

Fig. 3. Mortality rate of *P. kerathurus* PL20 acclimated to 2, 5, 10 and 20 ppt and transferred to higher salinity.

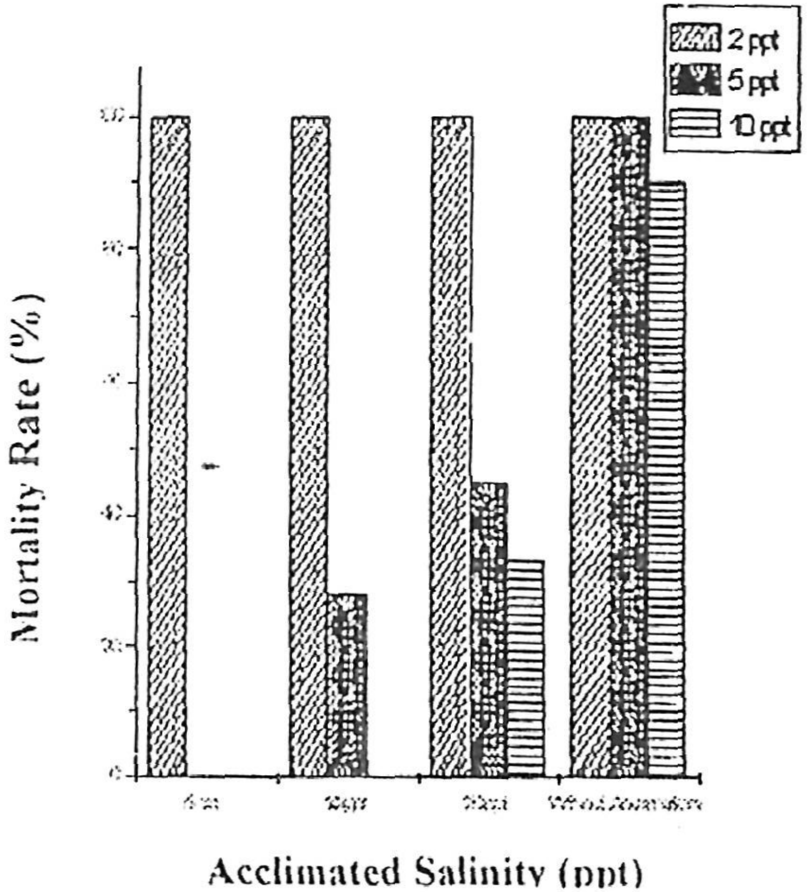

Fig. 4. Mortality rate of *P. kerathurus* PL30 acclimated to 2, 5, 10 and 20 ppt and transeferred to lower salinity.

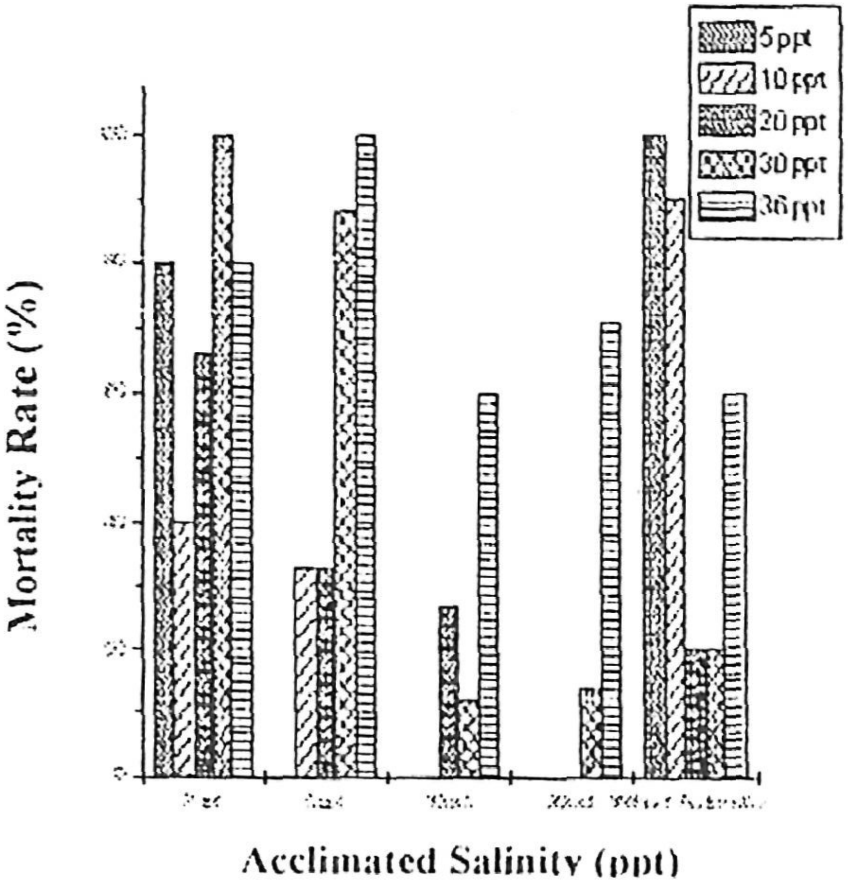

Fig. 5. Mortality rate of *P. kerathurus* PL30 acclimated to 2, 5, 10 and 20 ppt and transeferred to higher salinity.

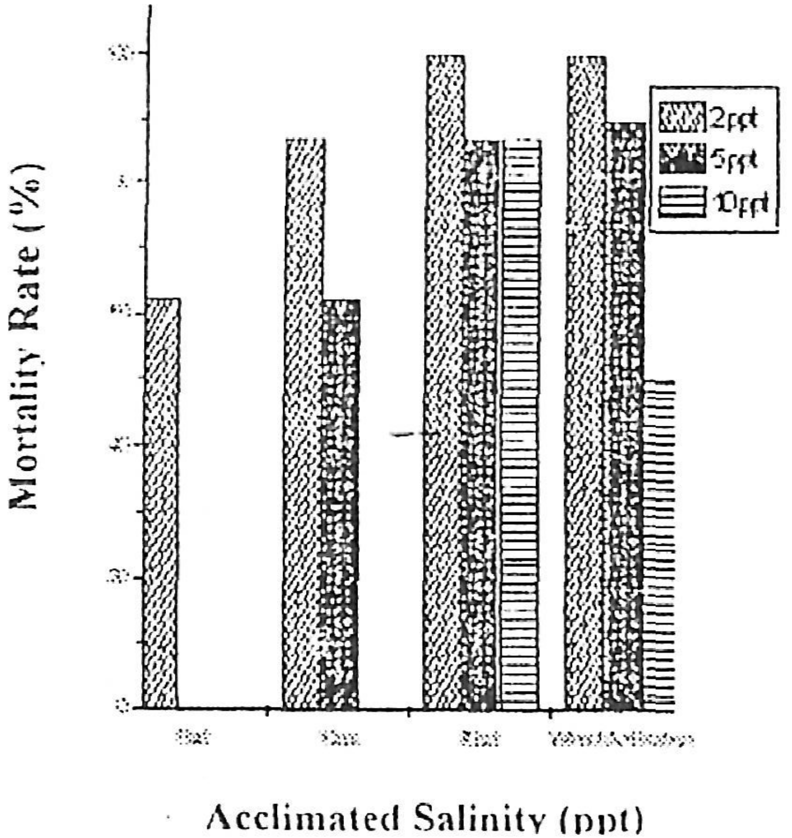

Fig. 6. Mortality rate of *P. kerathurus* PL40 acclimated to 2, 5, 10 and 20 ppt and transeferred to lower salinity.

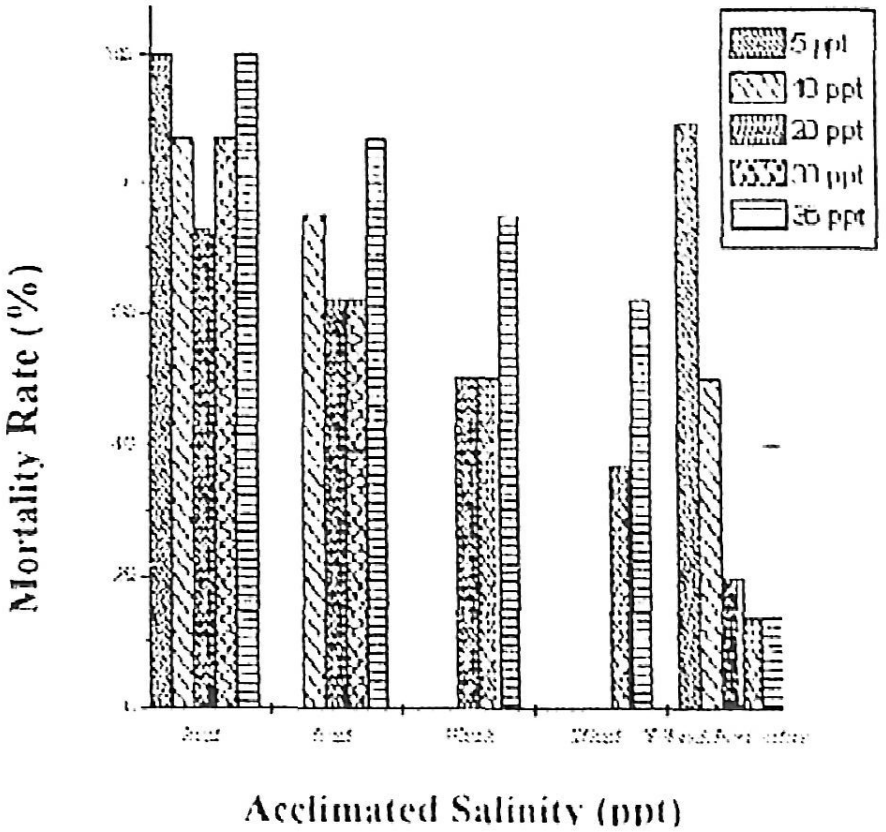

Fig. 7. Mortality rate of *P. kerathurus* PL40 acclimated to 2, 5, 10 and 20 ppt and transeferred to higher salinity.

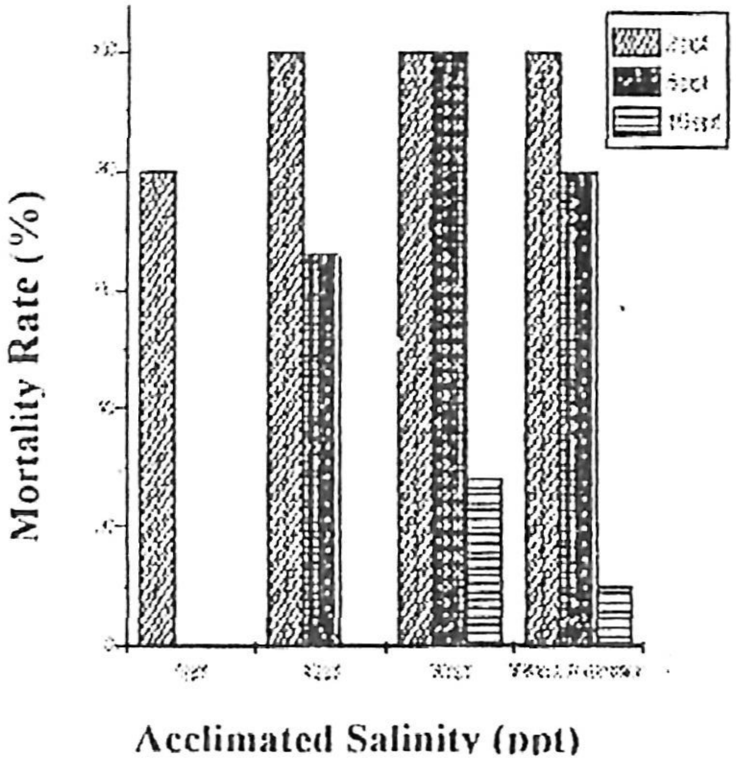

Fig. 8. Mortality rate of *P. kerathurus* PL50 acclimated to 2, 5, 10 and 20 ppt and transeferred to lower salinity.

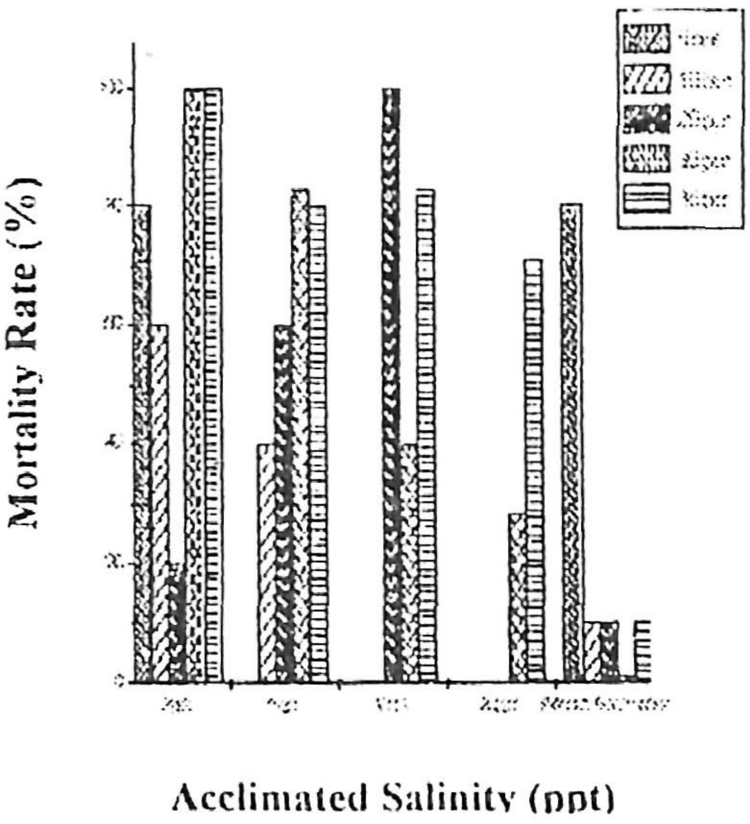

Fig. 9. Mortality rate of *P. kerathurus* PL50 acclimated to 2, 5, 10 and 20 ppt and transeferred to higher salinity.

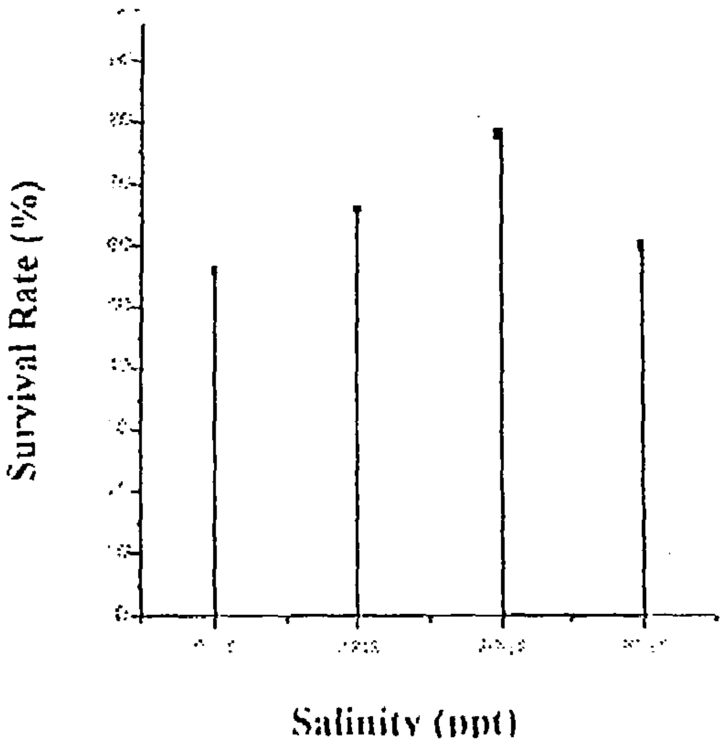

Fig. 10. Survival rate of *P. kerathurus* postlarvae at different salinities after 30 days.

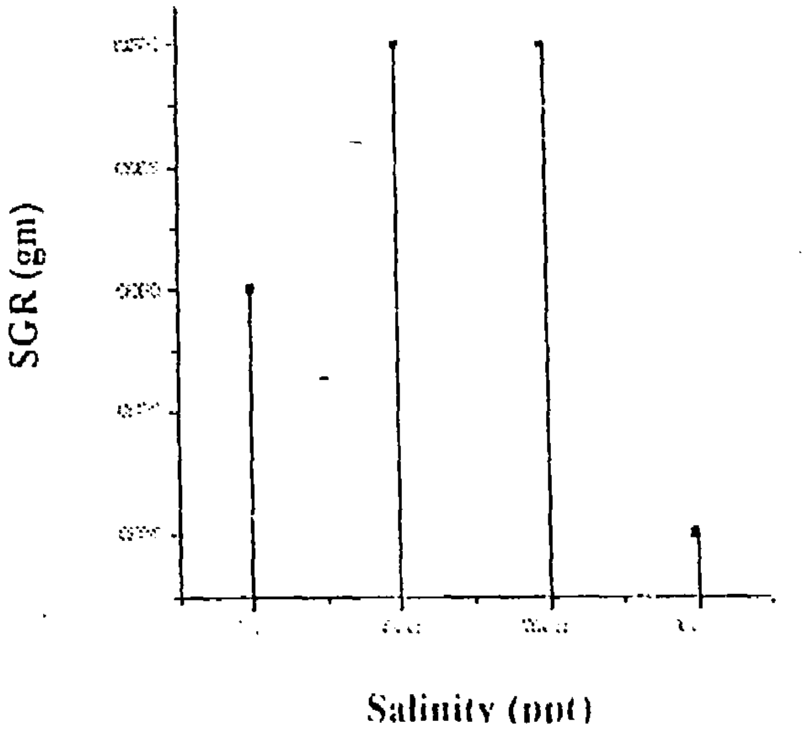

Fig. 11. Specific growth rate (SGR) of *P. kerathurus* postlarvae at different salinities after 30 days.

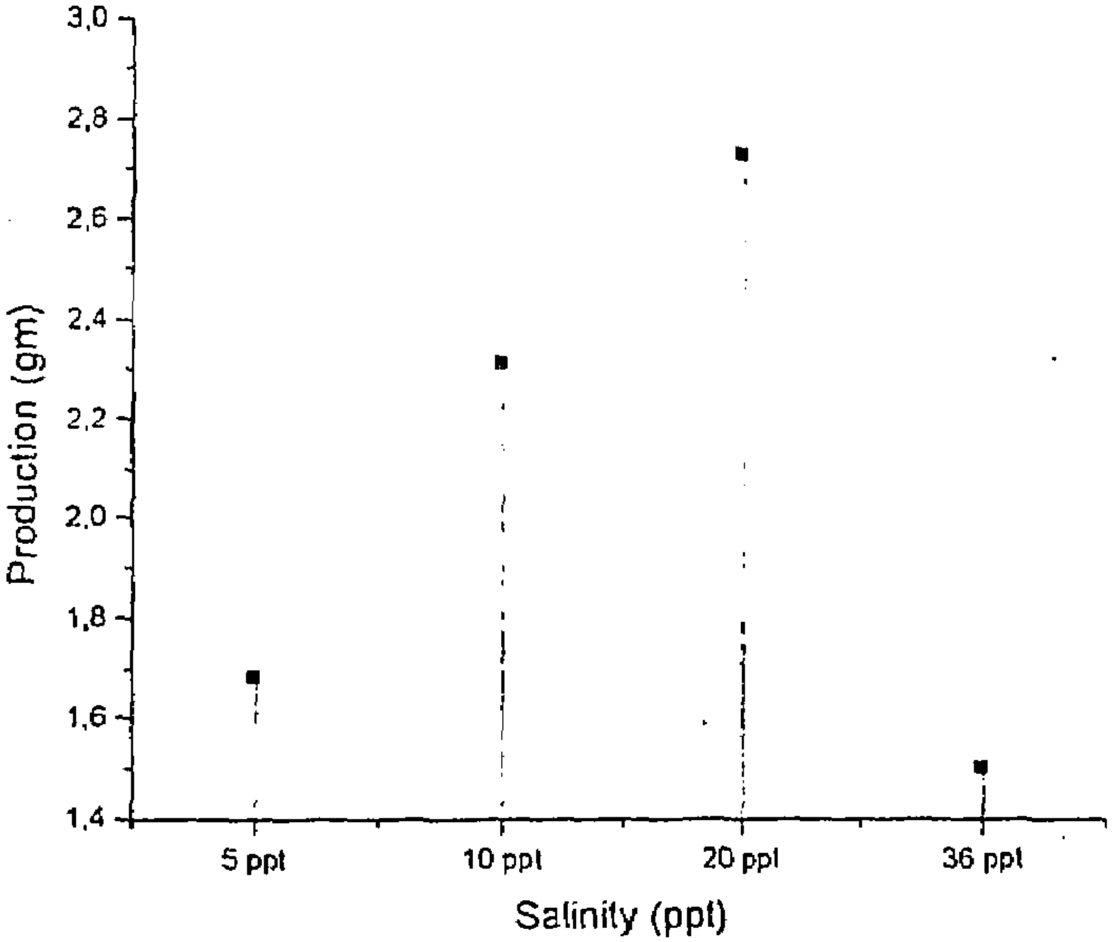

Fig. 12. Production of *P. kerathurus* postlarvae at different salinities after 30 days.