**Journal of Environmental Biology**
p-ISSN: 0254-8704 • e-ISSN: 2394-0379 • CODEN: JEBIDP

JEB®

Original Research Paper
Volume 47 Issue 6 November 2026 pp. 1242-1249
*DOI:* http://doi.org/10.22438/jeb/47/6/MRN-6015

*J. Environ. Biol.*, 2026
© Triveni Enterprises, Lucknow (India)
Journal website : www.jeb.co.in ★ E-mail : editor@jeb.co.in

# Density optimization of pearlspot, *Etroplus suratensis* (Bloch, 1790), fry and fingerlings for live transportation

**P.A. Patil[1]*, K.P.K. Vasagam[2], S. Khara[1], S. Sundi[1], M. Kailasam[2], A. Panigrahi[2] and P. Mahalakshmi[2]**

[1]*Navsari Gujarat Research Centre of ICAR-CIBA, Navsari Agricultural University, Navsari-396 450, India*
[2]*ICAR-Central Institute of Brackishwater Aquaculture, Chennai-600 028, India*

**Received:** 11 March 2026 **Revised:** 18 June 2026 **Accepted:** 21 August 2026

***Corresponding Author Email:** pankaj.patil@icar.org.in ***ORCiD:** https://orcid.org/0000-0003-2514-6957

## Abstract

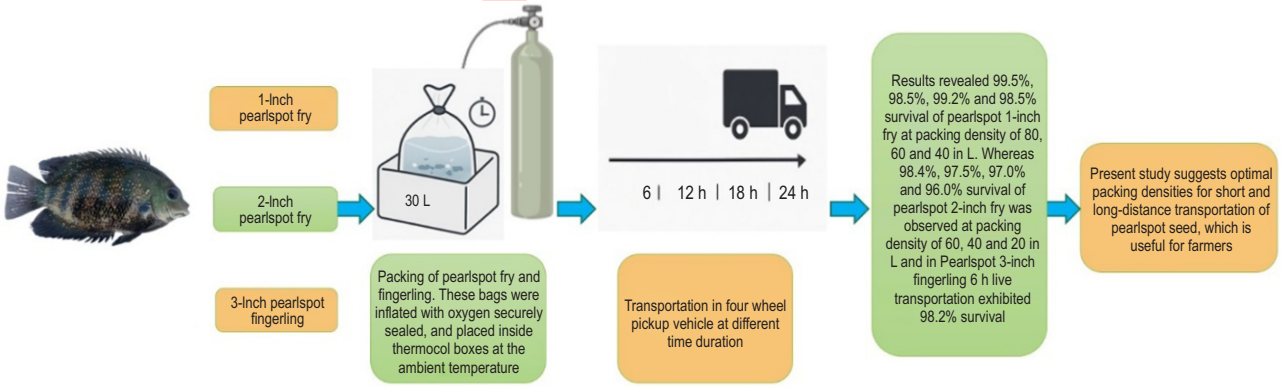
**Aim:** The study aimed to identifying optimal stocking densities and transport durations of Pearlspot (Etroplus suratensis) of varying sizes (1–3 inches) in closed, oxygenated systems.

**Methodology:** Pearlspot fry and fingerlings were transported for 6, 12, 18, and 24 hr in 30 l polythene bags containing 5 l of oxygenated 15 ppt brackishwater. In Trial one, 1-inch fry (2.5-2.7 cm) were packed at stocking densities of 20, 40, 60, 80 and 100 fish $l^{-1}$. In Trial two, 2-inch fry (5.0-5.5 cm) were packed at 20, 40 and 60 fish $l^{-1}$, while in Trial three, 3 inch-fingerlings (7.3-7.9 cm) were packed at 10, 20 and 30 fish $l^{-1}$. An oxygenated-water-only treatment was maintained as the control in all trials. Bags held in thermocol boxes, and survival and water quality (temperature, salinity, pH, dissolved oxygen, ammonia) were assessed pre-packing and post-transport.

**Results:** A 1-inch fry, survived above 99% up to 18 hr at of 20–60 Nos. $l^{-1}$, with the 20 Nos. $l^{-1}$ supporting 99.0% survival at 24 hr. A 2-inch fry, survived 97% up to 12 hr at 20 and 40 Nos. $l^{-1}$ but dropped sharply at higher densities. A 3-inch fingerlings were most sensitive, tolerating only 10 nos. $l^{-1}$ for 6 hr before survival declined to 0% by 24 hr.

**Interpretation:** Optimal stocking densities for transporting fry and fingerlings over short and long distances, benefit seed producers and farmers.

**Key words:** *Etroplus suratensis*, Live transport, Pearlspot fry, Transit time, Water quality




online copy

**How to cite:** Patil, P.A., K.P.K. Vasagam, S. Khara, S. Sundi, M. Kailasam, A. Panigrahi and P. Mahalakshmi: Density optimization of pearlspot, *Etroplus suratensis* (Bloch, 1790), fry and fingerlings for live transportation. *J. Environ. Biol.*, **47**, 1242-1249 (2026).

## Introduction

Transportation of fish seeds, including spawn, fry and fingerlings, from hatcheries to rearing ponds require application of scientifically validated techniques. This process involves careful management of appropriate temperature, oxygen levels and water quality. Live fish transportation requires suitable materials such as oxygen-filled polythene bags, insulated containers, FRP tanks and aerated transport vehicles to ensure safe movement of seed from hatcheries to grow-out systems (Shah, 2021). Since hatcheries and nursery facilities are often located far from grow-out ponds, cages, and net pens, fish seed must be transported over varying distances through road, rail, air, or water transport systems (Harmon, 2009). Determining suitable loading densities for during transportation remains a challenge due to lack of species-specific information. As a result, fish transport operations frequently rely on traditional or conservative stocking densities as a precautionary approach to minimize stress and reduce mortality.

Loading and transportation of fish, particularly under high-density conditions, may lead to mechanical injuries, deterioration of water quality, and elevated physiological stress, reducing survival rates (Urbinati *et al.,* 2004; Carneiro *et al.,* 2009). Therefore, optimizing the number of animals per unit volume or area is essential to achieve a satisfactory cost–benefit ratio while mitigating stress-related physiological responses (Carneiro and Urbinati, 2002). Recent studies on live fish transportation have demonstrated that optimization of transport density and duration is critical for maintaining survival, minimizing physiological stress, and preserving water quality in species such as *Pampus argenteus* (Peng *et al.,* 2012), *Rhamdia quelen* (Bittencourt *et al.,* 2018), *Labeo rohita* (Das *et al.,* 2025) highlighting the need for species-specific transport protocols in aquaculture. The goal of effective fish transportation is to use the least amount of water, while maximizing stocking density without raising mortality, degrading water quality or causing undue stress (Grøttum *et al.,* 1997). Plastic bags have emerged as the most popular global way of transporting fish fry globally, valued for their logistical effectiveness and functional simplicity (Mirzargar *et al.,* 2022; Mousavi *et al.,* 2023). Previous research has identified key factors affecting survival during transitions, including stocking density (Braun and Nuñer, 2014; Hong *et al.,* 2019; Hoseini *et al.,* 2019), transit period length (Sampaio and Freire, 2016), and containment strategies (Gomes *et al.,* 2006; Harmon, 2009).

This method is widely adopted for long-duration live fish transport (>24 hr), as it reduces the total volume and weight of transport water and improves economic feasibility for prolonged travel (Berka, 1986). Pearlspot, *Etroplus suratensis* (Bloch, 1790), is a native brackishwater fish that is widely found along Peninsular India's eastern and south-western coasts. It is a member of the Cichlidae family. It is an omnivorous euryhaline species that can be raised in both freshwater and brackishwater ponds and can withstand a broad range of salinity levels (Padmakumar *et al.,* 2012). However, the overall distribution of this species remains largely skewed toward the southern western coastline because most of the hatchery is in the southern region (Vikas *et al.,* 2020; Smina *et al.,* 2025), resulting in a substantial reliance on long-distance seedstock shipping to serve aquaculture operators and farmers in other regions of the nation. The best density duration combinations for the live transit of Pearlspot fry and fingerlings are still unknown despite advancements in hatching techniques and commercial success in India. This suggests a critical knowledge gap in transport management practices. As a result, this study identifies the best densities and transit times for live transportation of Pearlspot fry (1-2 inch) and fingerlings (3-inch) in 30 l oxygen-filled polyethylene bags.

## Materials and Methods

**Study site:** The study was conducted from July to August 2023 at the Brackishwater Aquaculture Research and Development (BARD) Farm, Matwad, Navsari Gujarat Research Center (NGRC), of Central Institute of Brackishwater Aquaculture (CIBA), in Navsari, Gujarat (20°55' 13.89"N and 72°49' 18.28"E).

**Experimental animals:** Hatchery-reared Pearlspot (*Etroplus suratensis*) fry produced were acclimatized in the tank to achieve the desired salinity (15 ppt). The fry and fingerling of Pearlspot were used in the experiment for transportation in sizes ranging from 1-3 inches at different densities in a polythene bag. The fry was reared in hapa to achieve the required size classes for 30–35 days post-hatch for 1-inch, 2 inches within 50-60 days, and 3 inches within 75–80 days. Brackishwater used for acclimatization, transportation, and all experimental treatments was disinfected by bleaching and conditioned to a salinity of 15 ppt prior to use.

**Experimental design:** The study consisted of three independent trials based on fish size classes. In experiment-I, 1-inch pearlspot fry were packed at stocking densities of 20, 40, 60, 80, and 100 Nos. $l^{-1}$; in experiment-II, 2-inch fry were packed at 20, 40, and 60 Nos. $l^{-1}$; and experiment-III, 3-inch fingerlings were packed at 10, 20, and 30 Nos. $l^{-1}$. Each treatment was evaluated at four transportation durations (6, 12, 18, and 24 hr) with suitable replicates, while oxygenated 15 ppt brackishwater without fish served as the control. The experimental units (30 l polythene bags containing 5 l water) were randomly allocated and placed inside thermocool boxes at ambient temperature for transportation in four-wheel pickup vehicle. Each treatment was replicated thrice. Survival rate and water quality parameters were assessed before packing and after each transportation duration.

**Water quality analysis:** The bags were opened once the allotted transportation time had passed, and Eutech CyberScan Series 600 portable meters were used to measure the water temperature (°C), salinity (ppt), pH and dissolved oxygen (mg $l^{-1}$). Standard meters were used to measure the amounts of ammonium-N (mg $l^{-1}$) (Hanna).

**Statistical analysis:** The completely randomized design was used for all experimental trials. Treatment Means were compared

Online Copy

**Table 1:** Survival rate of Pearlspot one-inch fry loaded at densities of 20, 40, 60, 80 and 100 Nos. $l^{-1}$ and transported for 6, 12, 18 or 24 hrs

| Density/hour | 20 Nos. $l^{-1}$ | 40 Nos. $l^{-1}$ | 60 Nos. $l^{-1}$ | 80 Nos. $l^{-1}$ | 100 Nos. $l^{-1}$ |
|---|---|---|---|---|---|
| 6 hr | $100 \pm 0.0^{a}$ | $100 \pm 0.0^{a}$ | $99.8 \pm 0.1^{a}$ | $99.5 + 0.6^{a}$ | $92.5 \pm 2.9^{b}$ |
| 12 hr | $99.3 \pm 0.0^{a}$ | $99.2 \pm 0.0^{a}$ | $99.0 \pm 0.5^{a}$ | $98.5 \pm 0.6^{a}$ | $89.9 \pm 3.3^{b}$ |
| 18 hr | $99.3 \pm 0.1^{a}$ | $99.2 \pm 0.3^{a}$ | $99.2 \pm 0.3^{a}$ | $90.2 \pm 4.3^{b}$ | $79.3 \pm 7.8^{c}$ |
| 24 hr | $99.0 \pm 0.3^{a}$ | $98.5 \pm 0.0^{a}$ | $93.7 \pm 0.1^{b}$ | $79.6 \pm 1.0^{c}$ | $66.7 \pm 1.1^{d}$ |

Values are presented as mean ± SD (n = 3). Mean values within the same column bearing different superscript letters differ significantly at $p < 0.05$ according to Tukey's post-hoc test, whereas values sharing the same superscript letter are not significantly different

**Table 2:** Survival rate of Pearlspot 2-inch fry loaded at densities of 20, 40 and 60 Nos. $l^{-1}$ and transported for 6, 12, 18 and 24 hrs

| Density/hour | 20 Nos. $l^{-1}$ | 40 Nos. $l^{-1}$ | 60 Nos. $l^{-1}$ |
|---|---|---|---|
| 6 hr | $99.3 \pm 0.3^{a}$ | $99.0 \pm 0.2^{a}$ | $98.4 \pm 0.7^{b}$ |
| 12 hr | $98.0 \pm 0.6^{a}$ | $97.5 \pm 0.3^{a}$ | $90.33 \pm 0.5^{b}$ |
| 18 hr | $97.0 \pm 0.6^{a}$ | $76.2 \pm 1.6^{b}$ | $60.6 \pm 2.4^{c}$ |
| 24 hr | $96.0 \pm 0.6^{a}$ | $47.6 \pm 2.3^{b}$ | $28.1 \pm 1.9^{c}$ |

Values are presented as mean ± SD (n = 3). Mean values within the same column bearing different superscript letters differ significantly at $p < 0.05$ according to Tukey's post-hoc test, whereas values sharing the same superscript letter are not significantly different.

using One-way ANOVA (Statistical package for the Social Sciences SPSS 16 version).

## Results and Discussion

The results on the survival rate of one-inch Pearlspot fry packed in different densities at different transportation durations are presented in Table 1. Among different densities, the maximum survival rate (99.0%) was observed when the seed was packed up to 60 Nos. $l^{-1}$, and the transportation duration was up to 18 hr. The differences observed between the treatments were also statistically insignificant ($p>0.05$), thereby indicated maximum survival rate. However, the packing densities of 80 Nos. $l^{-1}$ and 100 Nos. $l^{-1}$ showed reduction in the survival rate (90.2 ± 4.3% and 79.3 ± 7.8%), and the differences were statistically significant ($p<0.05$) according to Tukey's post hoc test. However, higher packing densities of 80 and 100 $l^{-1}$ resulted in reduced survival rates, particularly during prolonged transportation. At 24 hr, the highest survival rates were recorded at 20 and 40 $l^{-1}$, whereas the lowest survival was observed at 100 $l^{-1}$

The present experiment demonstrated that one-inch size Etroplus suratensis fry exhibited better survival at packing densities of 20–40 Nos. $l^{-1}$ during transportation up to 24 hr, with maximum survival ranging from 98.0 to 99.0%, whereas survival declined significantly at higher densities and prolonged transport duration. The lowest stocking density of 20 Nos. $l^{-1}$ was selected as a baseline treatment based on preliminary trials and earlier reports indicating that lower packing densities reduce metabolic waste accumulation, maintain dissolved oxygen levels, and improve survival during live fish transportation (Berka, 1986). These findings indicate clear size-dependent differences in the capacity of fry and fingerlings to tolerate transport stress.

Similar observations have been reported in other hardy euryhaline fishes. High tolerance of Oreochromis mossambicus to crowding stress and reduced dissolved oxygen has been attributed to its strong osmoregulatory ability and adaptability to fluctuating salinity conditions (Lim and Webster, 2006). Likewise, *Chanos chanos* fry have been reported to tolerate relatively high transport densities with minimal mortality when water quality is maintained, reflecting strong physiological resistance to handling stress (Bagarinao, 1991). Similarly, *Mugil cephalus* showed considerable resilience during confined transport because of its euryhaline nature and metabolic adaptability (Sampaio *et al.,* 2002). These findings suggest that the high survival of Pearlspot fry under moderate packing densities may be associated with similar physiological hardiness.

However, at higher packing densities (80 and 100 Nos. $l^{-1}$), survival decreased markedly, indicating that prolonged confinement increases stress and deteriorates transport condition. Comparable reductions in survival at elevated stocking densities have also been reported for Pacama (Navarro *et al.,* 2017), Rohu fry (Chatterjee *et al.,* 2010), and juvenile Tambaqui (Gomes *et al.,* 2006). Therefore, optimizing both packing density and transport duration is essential for ensuring maximum survival during live transportation of Pearlspot seed. The result on the survival of 2-inch Pearlspot fry packed at different densities at different transport durations is presented in Table 2. Among three different densities, the lowest density 20 Nos. $l^{-1}$ showed above 96% survival rate up to 24 hr transportation time. The differences observed between the treatments were also statistically insignificant ($p>0.05$), thereby indicated maximum survival rate. While, when the seeds were transported at 40 Nos. $l^{-1}$ up to 12 hr better survival indicated (up to 97.5 ± 0.3) and no statistical difference ($p>0.05$) was found between 6 and 12 hr transport

Online copy

**Table 3:** Survival rate of Pearlspot 3-inch fingerlings loaded at densities of 10, 20 and 30 Nos. $l^{-1}$ and transported for 6, 12, 18 and 24 hrs, respectively.

| Density/hour | 10 Nos. $l^{-1}$ | 20 Nos. $l^{-1}$ | 30 Nos. $l^{-1}$ |
|---|---|---|---|
| 6 hr | 98.0± 1.1[a] | 68.3 ± 2.1[b] | 23.6 ± 3.3[c] |
| 12 hr | 64.0 ± 4.6[a] | 27.7 ± 4.3[b] | 6.2 ± 0.9[c] |
| 18 hr | 9.3 ± 2.9[a] | 0 | 0 |
| 24 hr | 0 | 0 | 0 |

Values are presented as mean ± SD (n = 3). Mean values within the same column bearing different superscript letters differ significantly at $p < 0.05$ according to Tukey's post hoc test, whereas values sharing the same superscript letter are not significantly different.

duration. However, at the highest stocking density (60 fish $l^{-1}$), high survival (98.4 ± 0.7%) was recorded only during the shortest transportation duration of 6 hr, indicating that this stocking density was suitable for short-duration transport, but may not be optimal for longer transportation periods. In the current study, 2-inch fry exhibited optimal survival when stocked at a density of 20 Nos. $l^{-1}$. For shorter transport durations ranging from 6 to 12 hr, the packing density was successfully increased to 40–60 Nos. $l^{-1}$, while maintaining a survival rate exceeding 90.0%. Transport stress is driven primarily by rapid deterioration of water quality including declining dissolved oxygen, increasing Ammonium-N, and pH reduction which collectively impair physiological homeostasis. The findings of Bittencourt *et al.* (2018) support the current study by demonstrating that stocking density, temperature, and length of transport during early life stages have a significant impact on survival during live fish transportation because these variables control metabolic stress and declining water quality.

The results on the survival rate of 3-inch Pearlspot fingerlings packed at different densities over various transportation durations are presented in Table 3. Among the treatments, a high survival rate was observed only at the lowest density of 10 Nos. $l^{-1}$ during first 6 hr of transportation (98.0 ± 1.1%). At this same duration, increasing the density led to a sharp and significant decline in survival, dropping to 68.3 ± 2.1% at 20 Nos. $l^{-1}$ and further to 23.6 ± 3.3% at 30 Nos. $l^{-1}$. As the transportation duration increased to 12 hr, survival rates plummeted across all groups. By 24 hr mark, total mortality (0% survival) was observed across all packing densities.

Pearlspot 3-inch fingerlings, transportation is only viable for short durations. High survival (above 97%) could be maintained only at a low density of 10 Nos. $l^{-1}$ up to 6 hr. The data clearly indicates that three-inch fingerlings are not suitable for long-distance transportation (beyond 6 hr) at these densities, as higher packing rates and extended durations lead to total mortality. Similar patterns were observed in Rohu (*Labeo rohita*) fry, where near-complete survival occurred at moderate densities for up to 12–18 hr, but mortality increased significantly once crowding thresholds exceeded (Honnananda and Kumar, 2021; Das *et al.,* 2025). Water quality parameters showed significant changes over time and across different stocking densities. Salinity remained constant in all treatments throughout the experiment showing no significant variation ($p > 0.05$). In contrast, during one-inch fry transportation, temperature increased significantly ($p < 0.05$) with both time and fry density, with the 80 and 100 Nos. $l^{-1}$ treatments exhibiting higher temperatures compared to the 20, 40, and 60 Nos. $l^{-1}$ treatments (Table 4).

In 2-inch fry transportation, temperature increased over time in all treatments, with 60 Nos. $l^{-1}$ treatment showing significantly higher temperatures compared to 20 and 40 Nos. $l^{-1}$ at most time points. At 6 and 12 hr, temperatures recorded at 20 and 40 Nos. $l^{-1}$ did not differ significantly ($p>0.05$), while at 60 Nos. $l^{-1}$ showed a significant increase. By 24 hr, the 60 Nos. $l^{-1}$ treatment attained the highest temperature, significantly exceeding 20 Nos. $l^{-1}$ group, while the 40 Nos. $l^{-1}$ treatment was intermediate (Table 5). In 3-inch fingerlings transportation temperature increased significantly over time, with 30 Nos. $l^{-1}$ treatment consistently showing higher temperatures compared to 10 and 20 Nos. $l^{-1}$ treatments at each time point. The 20 Nos. $l^{-1}$ treatment exhibited intermediate temperatures, while the 10 Nos. $l^{-1}$ treatment maintained the lowest temperatures throughout the study period (Table 6). A gradual increase in temperature observed with prolonged transport duration and higher stocking densities likely accelerated the metabolic activity, thereby increasing oxygen consumption and nitrogenous waste accumulation, collectively intensifying transport stress (Berka, 1986).

In one-inch fry transportation, the pH declined over time in all treatments, with 80 and 100 Nos. $l^{-1}$ treatments showing significantly lower pH compared to 20, 40, and 60 Nos. $l^{-1}$, indicating greater acidification at higher densities (Table 5). In 2-inch fry transportation the pH decreased gradually over time in all treatments, but no significant differences ($p>0.05$) were observed among the 20, 40, and 60 Nos. $l^{-1}$ treatments at any time point (Table 5). In 3-inch fingerlings transportation the pH declined progressively over time in all treatments, with the highest stocking density (30 Nos. $l^{-1}$) showing the most pronounced and significant decrease, followed by 20 Nos. $l^{-1}$, while the 10 Nos. $l^{-1}$ treatment retained relatively higher pH values (Table 6).

At the conclusion of the experiment, acidic pH values were observed, which is consistent with the findings of Colt (2002), who discovered acidic pH levels in young silver catfish (23–28 g) transported for 5 hr at 24.6°C at different loading densities (75–350 g $l^{-1}$). When rohu seeds were transported traditionally, the pH levels ranging from 8.4 to 6.5 were recorded

Online Copy

**Table 4:** Water quality parameter of Pearlspot 1-inch fry loaded at densities of 20, 40, 60, 80 and 100 Nos. $l^{-1}$ and transported for 6, 12, 18 and 24 hrs

| | | 6 hr | | | | | 12 hr | | | | | 18 hr | | | | | 24 hr | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Water Parameters** | **Initial** | **20 Nos. $l^{-1}$** | **40 Nos. $l^{-1}$** | **60 Nos. $l^{-1}$** | **80 Nos. $l^{-1}$** | **100 fry $l^{-1}$** | **20 Nos. $l^{-1}$** | **40 Nos. $l^{-1}$** | **60 Nos. $l^{-1}$** | **80 Nos. $l^{-1}$** | **100 Nos. $l^{-1}$** | **20 Nos. $l^{-1}$** | **40 Nos. $l^{-1}$** | **60 Nos. $l^{-1}$** | **80 Nos. $l^{-1}$** | **100 Nos. $l^{-1}$** | **20 Nos. $l^{-1}$** | **40 Nos. $l^{-1}$** | **60 Nos. $l^{-1}$** | **80 Nos. $l^{-1}$** | **100 fry $l^{-1}$** |
| Temperature ($^0$C) | 28.2 ± 0.00 | 28.4 ± 0.06$^a$ | 28.4 ± 0.06$^a$ | 28.4 ± 0.21$^a$ | 28.5 ± 0.06$^a$ | 28.5 ± 0.10$^a$ | 28.6 ± 0.15$^a$ | 28.7 ± 0.12+ | 29.0 ± 0.15$^a$ | 28.9 ± 0.17$^a$ | 29.3 ± 0.26$^b$ | 29.0 ± 0.15$^a$ | 29.2 ± 0.21$^a$ | 29.4 ± 0.12$^b$ | 29.5 ± 0.06$^b$ | 29.7 ± 0.15$^b$ | 29.4 ± 0.21$^a$ | 29.6 ± 0.15$^a$ | 30 ± 0.06$^b$ | 30 ± 0.10$^b$ | 30.1 ± 0.10$^b$ |
| Salinity (ppt) | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 |
| pH | 7.9 ± 0.10 | 7.7 ± 0.12$^a$ | 7.6 ± 0.15$^a$ | 7.5 ± 0.51$^a$ | 7.5 ± 0.41$^a$ | 7.2 ± 0.58$^a$ | 7.4 ± 0.35$^a$ | 7.4 ± 0.13$^a$ | 7.4 ± 0.12$^a$ | 7.0 ± 0.11$^a$ | 6.8 ± 0.30$^b$ | 7.5 ± 0.65$^a$ | 7.3 ± 0.37$^a$ | 7.3 ± 0.32$^a$ | 7.2 ± 0.30$^a$ | 6.8 ± 0.23$^b$ | 7.5 ± 0.06$^a$ | 7.0 ± 0.15$^b$ | 7.1 ± 0.06$^b$ | 7.1 ± 0.06$^b$ | 6.0 ± 0.21$^c$ |
| Dissolved oxygen (mg $l^{-1}$) | 13.3 ± 0.06 | 9.1 ± 0.10$^a$ | 8.8 ± 0.06$^a$ | 8.4 ± 0.30$^b$ | 8.4 ± 0.20$^b$ | 7.9 ± 0.25$^b$ | 7.5 ± 0.12$^a$ | 7.3 ± 0.25$^a$ | 7.0 ± 0.42$^a$ | 6.1 ± 0.15$^b$ | 5.0 ± 0.17+ | 6.1 ± 0.15$^a$ | 5.8 ± 0.15$^a$ | 5.4 ± 0.25$^b$ | 5.0 ± 0.21$^b$ | 4.2 ± 0.17$^c$ | 4.9 ± 0.15$^a$ | 4.9 ± 0.17$^a$ | 4.3 ± 0.21$^b$ | 4.1 ± 0.15$^b$ | 3.8 ± 0.15$^c$ |
| Ammonia (mg $l^{-1}$) | 0.02 ± 0.00 | 0.08 ± 0.01$^a$ | 0.10 ± 0.01$^a$ | 0.14 ± 0.04$^b$ | 0.16 ± 0.01$^b$ | 0.19 ± 0.01$^c$ | 0.12 ± 0.01$^a$ | 0.17 ± 0.02$^{ab}$ | 0.21 ± 0.01$^{bc}$ | 0.22 ± 0.02$^c$ | 0.37 ± 0.04$^d$ | 0.16 ± 0.02$^a$ | 0.22 ± 0.02$^b$ | 0.25 ± 0.01$^b$ | 0.25 ± 0.02$^c$ | 0.49 ± 0.02$^d$ | 0.22 ± 0.01$^a$ | 0.28 ± 0.02$^{ab}$ | 0.34 ± 0.04$^b$ | 0.4 ± 0.05$^c$ | 0.64 ± 0.04$^d$ |

Values are expressed as mean ± SE. Means bearing different superscript letters within the same row differ significantly (P<0.05)

Online Copy

**Table 5:** Water quality parameter of Pearlspot 2-inch fry loaded at densities of 20, 40 and 60 Nos. $l^{-1}$ and transported for 6, 12, 18 and 24 hrs

| | | 6 hr | | | 12 hr | | | 18 hr | | | 24 hr | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Water Parameters** | **Initial** | **20 Nos. $l^{-1}$** | **40 Nos. $l^{-1}$** | **60 Nos. $l^{-1}$** | **20 Nos. $l^{-1}$** | **40 Nos. $l^{-1}$** | **60 Nos. $l^{-1}$** | **20 Nos. $l^{-1}$** | **40 Nos. $l^{-1}$** | **60 Nos. $l^{-1}$** | **20 Nos. $l^{-1}$** | **40 Nos. $l^{-1}$** | **60 Nos. $l^{-1}$** |
| Temperature ($^0$C) | 28.2 ± 0.00 | 28.40 ± 0.14$^a$ | 28.4 ± 0.21$^a$ | 28.4 ± 0.25$^a$ | 28.70 ± 0.00$^a$ | 28.7 ± 0.21$^{ab}$ | 29.1 ± 0.12$^b$ | 29.37 ± 0.46$^a$ | 29.1 ± 0.06$^a$ | 29.7 ± 0.32$^a$ | 29.73 ± 0.15$^{ab}$ | 29.3 ± 0.38$^a$ | 30.1 ± 0.10$^b$ |
| Salinity (ppt) | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 |
| pH | 7.8 ± 0.10 | 7.4 ± 0.04$^a$ | 7.3 ± 0.15$^a$ | 7.2 ± 0.19$^a$ | 7.3 ± 0.28$^a$ | 7.2 ± 0.26$^a$ | 7.0 ± 0.11$^a$ | 7.0 ± 0.38$^a$ | 6.9 ± 0.05$^a$ | 6.7 ± 0.49$^a$ | 6.9 ± 0.57$^a$ | 6.6 ± 0.17$^a$ | 6.5 ± 0.11$^a$ |
| Dissolved oxygen (mg $l^{-1}$) | 13.5 ± 0.26 | 8.3 ± 0.23$^a$ | 7.6 ± 0.25$^b$ | 7.1 ± 0.12$^b$ | 7.3 ± 0.26$^a$ | 6.6 ± 0.38$^a$ | 5.9 ± 0.06$^b$ | 6.6 ± 0.25$^a$ | 5.3 ± 0.15$^b$ | 4.4 ± 0.06$^c$ | 5.4 ± 0.21$^a$ | 4.2 ± 0.20$^b$ | 3.8 ± 0.06$^b$ |
| Ammonia (mg $l^{-1}$) | 0.0 ± 0.01 | 0.07 ± 0.02$^a$ | 0.06 ± 0.04$^a$ | 0.13 ± 0.01$^b$ | 0.15 ± 0.04$^a$ | 0.17 ± 0.01$^a$ | 0.21 ± 0.03$^a$ | 0.17 ± 0.01$^a$ | 0.20 ± 0.01$^a$ | 0.24 ± 0.05$^a$ | 0.24 ± 0.03$^a$ | 0.28 ± 0.01$^a$ | 0.34 ± 0.02$^b$ |

Values are expressed as mean ± SE. Means bearing different superscript letters within the same row differ significantly (P<0.05)

**Table 6:** Water quality parameter of Pearlspot 3-inch fingerlings loaded at densities of 10, 20 and 30 Nos. $l^{-1}$ and transported for 6, 12, 18 and 24 hrs

| | | 6 hr | | | 12 hr | | | 18 hr | | | 24 hr | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Water Parameters** | **Initial** | **10 Nos. $l^{-1}$** | **20 Nos. $l^{-1}$** | **30 Nos. $l^{-1}$** | **10 Nos. $l^{-1}$** | **20 Nos. $l^{-1}$** | **30 Nos. $l^{-1}$** | **10 Nos. $l^{-1}$** | **20 Nos. $l^{-1}$** | **30 Nos. $l^{-1}$** | **10 Nos. $l^{-1}$** | **20 Nos. $l^{-1}$** | **30 Nos. $l^{-1}$** |
| Temperature ($^0$C) | 28.2 ± 0 | 28.7 ± 0.06$^a$ | 28.8 ± 0.10$^a$ | 29.1 ± 0.06$^b$ | 29.2 ± 0.12$^a$ | 29.6 ± 0.06$^b$ | 30.0 ± 0.0$^c$ | 29.8 ± 0.10$^a$ | 30.2 ± 0.12$^b$ | 30.6 ± 0.10$^c$ | 30.3 ± 0.12$^a$ | 30.7 ± 0.06$^b$ | 31.1 ± 0.06$^c$ |
| Salinity (ppt) | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 | 15 |
| pH | 7.83 ± 0.06 | 7.49 ± 0.09$^a$ | 6.69 ± 0.04$^b$ | 6.0 ± 0.10$^c$ | 6.85 ± 0.06$^a$ | 6.02 ± 0.08+ | 5.5 ± 0.15$^c$ | 6.16 ± 0.16$^a$ | 5.62 ± 0.13$^b$ | 5.1 ± 0.14$^c$ | 5.57 ± 0.41$^a$ | 5.35 ± 0.39$^b$ | 4.7 ± 0.12$^b$ |
| Dissolved Oxygen (mg $l^{-1}$) | 13.3 ± 0.06 | 6.9 ± 0.06$^a$ | 4.3 ± 0.10$^b$ | 3.3 ± 0.21$^c$ | 4.5 ± 0.10+ | 3.5 ± 0.17$^b$ | 2.7 ± 0.10$^c$ | 4.1 ± 0.06$^a$ | 2.7 ± 0.03$^b$ | 2.5 ± 0.06$^c$ | 2.4 ± 0.12$^a$ | 2.8 ± 0.12$^b$ | 1.1 ± 0.12$^c$ |
| Ammonia (mg $l^{-1}$) | 0.02 ± 0.00 | 0.30 ± 0.05$^a$ | 1.5 ± 0.15$^b$ | 2.1 ± 0.15$^c$ | 0.7 ± 0.06$^a$ | 1.7 ± 0.15$^b$ | 2.9 ± 0.15$^c$ | 1 ± 0.21$^a$ | 3 ± 0.15$^b$ | 3.0 ± 0.12$^b$ | 3 ± 0.10$^a$ | 3 ± 0.12$^a$ | 3.1 ± 0.0$^a$ |

Values are expressed as mean ± SE. Means bearing different superscript letters within the same row differ significantly (P<0.05)

Online copy

(Islam and Hossain, 2013). Increased $CO_2$ production during confinement reduces pH, which in turn inhibits nitrifying bacteria responsible for converting ammonia to less harmful forms (Kim *et al.,* 2006; Moran *et al.,* 2008; Pan *et al.,* 2019). This contributed to the elevated Ammonium-N levels observed during transport.

In one-inch fry transportation, the dissolved oxygen level decreased progressively with increasing fry density, with the 100 Nos. $l^{-1}$ treatment showing the lowest oxygen levels, followed by 80 Nos. $l^{-1}$, while 20, 40 and 60 Nos. $l^{-1}$ maintained comparatively higher DO concentrations (Table 4). In 2-inch fry transportation, DO level decreased significantly with increasing fry density and exposure time. At 6 hr, the 20 Nos. $l^{-1}$ treatment maintained significantly higher DO compared to 40 and 60 Nos. $l^{-1}$, and by 24 hr, the lowest DO was observed in the 60 Nos. $l^{-1}$ treatment (Table 5). In 3-inch fingerling transportation, dissolved oxygen decreased significantly with increasing stocking density and exposure duration, with the lowest oxygen levels recorded in the 30 Nos. $l^{-1}$ treatment and the highest in the 10 Nos. $l^{-1}$ treatment (Table 6). Dissolved oxygen is another essential parameter influencing survival. While seawater DO levels above 5 mg $l^{-1}$ are adequate for supporting both fish and nitrifying bacteria (Heriyati *et al.,* 2022). Importantly, end-point DO readings may not fully reflect real-time fluctuations during transport, and short-term oxygen deficits earlier in the process may contribute to mortality even when final measurements appear sufficient. The time of transit and the rate of oxygen consumption were found to be significantly correlated negatively at various densities. According to Peng *et al.* (2012), *Pampus argenteus* stocking density rose as DO levels decreased.

In one-inch fry transportation Ammonium-N accumulation was strongly density-dependent, with 80 and 100 Nos. $l^{-1}$ treatments showing significantly higher Ammonium-N concentrations than 20, 40, and 60 Nos. $l^{-1}$ treatments during the experiment (Table 4). In 2-inch fry transportation Ammonium-N concentrations increased over time in all treatments, with the 60 Nos. $l^{-1}$ treatment showing significantly higher Ammonium-N at 24 h compared to the 20 Nos. $l^{-1}$ treatment, while 40 Nos. $l^{-1}$ was intermediate (Table 5). In 3-inch fingerling transportation, ammonia concentrations increased markedly over time and were significantly higher in the 30 Nos. $l^{-1}$ treatment compared to 10 and

20 Nos. $l^{-1}$ (Table 6). Ammonium-N was measured as the principal nitrogenous waste indicator, because under prevailing experimental pH conditions, it represents the dominant dissolved nitrogen fraction and serves as a reliable indicator of metabolic waste accumulation during live transportation (Boyd and Tucker, 1998). Ammonium-N accumulation is a critical stressor during transport, originating from metabolic excretion and decomposition of any unnoticed dead fish (Fivelstad *et al.,* 1993). Un-ionized ammonia ($Nh_3$) is particularly toxic, and elevated total ammonia levels impair normal excretion processes, disrupt physiological function, and can lead to mortality (da Silva *et al.,* 2013). A progressive increase in Ammonium-N with longer durations and higher densities shows similar patterns reported in species such as large yellow croaker (Zhang *et al.,* 2014), silver catfish (Golombieski *et al.,* 2003) and topmouth culter (Hu *et al.,* 2014), confirming that higher transport intensity amplifies metabolic burdens.

A consistent pattern across studies is that smaller fish tolerate transportation better than larger juveniles. Their lower metabolic rate and reduced ammonia excretion delay water quality deterioration, extending their safe transport window. In contrast, fingerlings accumulate toxic nitrogen and experience dissolved oxygen depletion more rapidly, elevating physiological stress and lowering survival thresholds. A reduced tolerance level with increasing fish size observed in the present study aligns with these established relationships, reinforcing the need to tailor transport protocols to fish size requiring shorter durations and lower densities as fish grow larger. Water quality dynamics during transport are widely recognized as a major determinant of fish stress and survival (Portz *et al.,* 2006; Harmon, 2009). High densities and prolonged confinement accelerate the deterioration of key parameters, with dissolved oxygen and pH declining and Ammonium-N increasing over time (Bui *et al.,* 2013; Zeppenfeld *et al.,* 2014; Salbego *et al.,* 2015). Consistent with this trend, the present study showed a steady decrease in pH and a rise in Ammonium-N regardless of fish density.

The findings of this study establish scientifically validated transport density and duration guidelines for pearlspot seed hatcheries and suppliers, enabling reduced mortality, improved fish welfare, and cost-effective seed transportation.

## Acknowledgments

The authors are grateful to the ICAR and The Director, ICAR–Central Institute of Brackishwater Aquaculture (CIBA), Chennai, for providing necessary facilities for conductance of the experiment at NGRC-CIBA, Navsari, Gujarat.

**Authors' contribution: P.A. Patil:** Conceptualization, investigation, validation, methodology, visualization, Writing–original draft, Writing – revision and editing; **K.P.K. Vasagam:** Methodology, Data curation; **S. Khara** and **S. Sundi:** Data analysis, Writing-revision and editing manuscript; **M. Kailasam,** Supervision and revision and Editing of manuscript; **A. Panigrahi:** Supervision and revision of MS; **P. Mahalakshmi:** Supervision.

**Funding:** The work was supported by the Indian Council of Agricultural Research [FISHCIBACIL201500900126].

**Research content:** The content of this study is original and has not been published elsewhere.

**Ethical approval:** The study was undertaken with the approval of the statutory authorities of the Central Institute of Brackishwater Aquaculture, Chennai, India. *Etroplus suratensis* (Pearlspot) is not listed as an endangered or protected species under the Government of India's Wildlife (Protection) Act, 1972.

**Conflict of interest:** The authors declare that is there is no conflict of interest.

**Data availability:** The data that support the findings of this study are available from the lead author.

**Consent to publish:** All authors agree to publish the paper in *Journal of Environmental Biology.*

## References

Bagarinao, T.U.: Biology of milkfish (*Chanos chanos* Forsskal). SEAFDEC Aquaculture Department, Tigbauan, Iloilo, Philippines, 94 pages (1991).

Berka, R.: The Transport of Live Fish: A Review. EIFAC Technical Paper No. 48, Food and Agriculture Organization of the United Nations, Rome, Italy, 52 pages (1986).

Bittencourt, F., D.Z. Damasceno, T.A. Lui, A. Signor, E.A. Sanches and D.H. Neu: Waterquality and survival rate of *Rhamdia quelen* fry subjected to simulated transportation atdifferent stock densities and temperatures. *Acta Scientiarum. Anim. Sci.,* **40**, e37285 (2018).

Bui, T.M., N.T. Phuong, G.H. Nguyen and S.S. De Silva: Fry and fingerling transportation inthe striped catfish, *Pangasianodon hypophthalmus*, farming sector, Mekong Delta, Vietnam: A pivotal link in the production chain. *Aquaculture,* **388**, 70–75 (2013).

Boyd, C.E. and C.S. Tucker: Pond aquaculture water quality management. Kluwer Academic Publishers, 698 pages (1998).

Braun, N., and A. P. D. O. Nuner: Stress in *Pimelodus* maculatus (Siluriformes: Pimelodidae) at different densities and times in a simulated transport. *Zoologia (Curitiba)*, **31**, 101–104 (2014).

Carneiro, P. and E.C. Urbinati: Transport stress in matrinxã, *Brycon cephalus* (Teleostei:Characidae) at different densities. *Aquac. Int.,* **10**, 221–229 (2002).

Carneiro, P.C.F., P.H. d.S. Kaiseler, E.d.A.C. Swarofsky and B. Baldisserotto: Transport ofjundiá *Rhamdia quelen* juveniles at different loading densities: Water quality and blood parameters. *Neotrop. Ichthyol.,* **7**, 283–288 (2009).

Chatterjee, N., A.K. Pal, T. Das, R. Dalvi, M.S. Mohammad, K. Sarma and K. Baruah: Effectof stocking density and journey length on the welfare of rohu (*Labeo rohita* Hamilton)fry. *Aquac. Int.,* **18**, 859–868 (2010).

Colt, J.: List of spreadsheets prepared as a complement. In: Fish Hatchery Management (Ed.:G. Wedemeyer). American Fisheries Society, Bethesda, USA, 751 pages (2002).

Online copy

da Silva, F.J.R., F.R.S. Lima, D.A. do Vale and M.V. do Carmo: High levels of total ammonia nitrogen as $NH_4^+$ are stressful and harmful to the growth of Nile tilapia juveniles. *Acta. Sci. Biol. Sci.,* **35**, 475–481 (2013).

Das, R., M.S. Uddin, D. Pandit, I.A. Fagun, M.A. Kawsar, N. Begum and M.T. Alam: Investigation of transportation carrying capacity of rohu (*Labeo rohita*) fry: Insights into water quality parameters. *Discover Animals,* **2**, 40 (2025).

Fivelstad, S., H. Kallevik, H.M. Iversen, T. Møretrø, K. Våge and M. Binde: Sublethal effectsof ammonia in soft water on Atlantic salmon smolts at a low temperature. *Aquac. Int.,* **1**, 157–169 (1993).

Golombieski, J., L. Silva, B. Baldisserotto and J. Da Silva: Transport of silver catfish (*Rhamdia quelen*) fingerlings at different times, load densities and temperatures. *Aquaculture,* **216**, 95–102 (2003).

Gomes, L.C., C.A.R.M. Araújo-Lima, A.R. Chippari-Gomes and R. Roubach: Transportation of juvenile tambaqui (*Colossoma macropomum*) in a closed system. *Braz. J. Biol.,* **66**, 493–502 (2006).

Grøttum, J., M. Staurnes and T. Sigholt: Effect of oxygenation, aeration and pH control on water quality and survival of turbot, *Scophthalmus maximus* (L.), kept at high densities during transport. *Aquac. Res.,* **28**, 159–164 (1997).

Harmon, T.S.: Methods for reducing stressors and maintaining water quality associated withlive fish transport in tanks: A review of the basics. *Rev. Aquac.,* **1**, 58–66 (2009).

Heriyati, E., R. Rustadi, A. Isnansetyo, B. Triyatmo, I. Istiqomah, D. Deendarlianto and W. Budhijanto: Microbubble aeration in a recirculating aquaculture system increased dissolved oxygen,fish culture performance and stress resistance of red tilapia (*Oreochromis* sp.). *Trends Sci.,* **19**, 6251 (2022).

Honnananda, B.R., M. Kumar, B.N. Sanap, K.K. Choudhary, A. Verma, A. Verma and Jyoti: Effect of packaging density during different transportation periods on water quality and survival of *Labeo rohita* (Hamilton, 1822) fingerlings. *J. Exp. Zool. India,* **24**, 255–259 (2021).

Hong, J., X. U. Chen, S. Liu, Z. Fu, M. Han, Y. Wang, Z. Gu and Z. Ma: Impact of fish density on water quality and physiological response of golden pompano (*Trachinotus ovatus*) fingerlings during transportation. *Aquaculture*, **507**, 260–265 (2019).

Hu, P., R. Liu, Z. Zhao, H. He, Q. Fan and K. Zong: Effects of transportation time and densityon whole-body cortisol concentrations, oxygen consumption rate and water quality of juvenile topmouth culter, Culter alburnus. *Acta Hydrobiol. Sin.,* **38**, 1190–1194 (2014).

Hoseini, S. M., M. Yousefi, S.H. Hoseinifar and H. Van Doan: Cytokines' gene expression, humoral immune and biochemical responses of common carp (*Cyprinus carpio*, Linnaeus, 1758) to transportation density and recovery in brackish water. *Aquaculture*, **504**, 13–21 (2019).

Islam, M. and M. Hossain: Mortality rate of fish seeds (*Labeo rohita*) during traditional transportation system in northwest Bangladesh. *J. Sci. Res.,* **5**, 1–7 (2013).

Kim, D.-J., D.-I. Lee and J. Keller: Effect of temperature and free ammonia on nitrification and nitrite accumulation in landfill leachate and analysis of its nitrifying bacterial community by FISH. *Bioresour. Technol.,* **97**, 459–468 (2006).

Lim, C. and C.D. Webster: Tilapia: Biology, Culture and Nutrition. New York: Food Products Press, 678 pages (2006).

Mirzargar, S. S., A. T. Mirghaed, S.M. Hoseini, M. Ghelichpour, M. Shahbazi and M. Yousefi: Biochemical responses of common carp, *Cyprinus carpio*, to transportation in plastic bags using thymol as a sedative agent. *Aquacul. Res.*, **53**, 191–198 (2022).

Moran, D., R.M. Wells and S.J. Pether: Low stress response exhibited by juvenile yellow tailking fish (*Seriola lalandi* Valenciennes) exposed to hypercapnic conditions associated with transportation. *Aquac. Res.,* **39**, 1399–1407 (2008).

Mousavi, H. E., A. T. Mirghaed, S. M. Hoseini, M. Ghelichpour, A. A. Moghaddam, B. Gharavi and B. Aydın: Myrcene as water conditioner, stress-reducing and antioxidant agent in transportation of common carp, *Cyprinus carpio*, with plastic bags. *Aquacul. Rep.*, **28**, 101458 (2023).

Navarro, R.D., D.C. Costa, W. de Souza, B.C. da Silva and R.K. Luz: Long-term transportation of juvenile pacamãs (*Lophiosilurus alexandri*) at different densities. *Acta Scienti. Technol.,* **39**, 211–214 (2017).

Padmakumar, K.G., L. Bindu and P.S. Manu: *Etroplus suratensis* (Bloch), the state fish of Kerala. *J. Biosci.,* **37**, 925–931 (2012).

Pan, Y., H. Chen, X. Huang and X. Lu: Requirement and control for water quality in marinefish culture (continued). *Sci. Fish Farm,* **1**, 62–63 (2019).

Peng, S., X. Chen, Z. Shi, F. Yin and P. Sun: Survival of juvenile silver pomfret, *Pampus argenteus,* kept in transport conditions at different densities and temperatures. *Isr. J. Aquacult. Bamidgeh,* **64**, 1–6 (2012).

Portz, D.E., C.M. Woodley and J.J. Cech Jr.: Stress-associated impacts of short-term holdingon fishes. *Rev. Fish Biol. Fish.,* **16**, 125–170 (2006).

Salbego, J., A.G. Becker, T.V. Parodi, C.C. Zeppenfeld, J.F. Goncalves, V.L. Loro, V.M. Morsch, M.R.C. Schetinger, G. Maldaner and A.F. Morel: Methanolic extract of *Condalia buxifolia* added to transport water alters biochemical parameters of the silvercatfish *Rhamdia quelen*. *Aquaculture*, **437**, 46–50 (2015).

Sampaio, L.A., W. Wasielesky and K.C. Miranda-Filho: Effect of salinity on acute toxicity of ammonia and nitrite to juvenile mullet (*Mugil platanus*). *Bull. Environ. Contam. Toxicol.,* **68**, 668–674 (2002).

Shah, M.I.: Transportation of live fish seed. *Fish. Aquac. J.,* **12**, 1–4 (2021).

Smina, M.S., S. Joseph and P.B. Ajith Kumar: Backyard seed production of pearl spot (*Etroplus suratensis*) in FRP/cement tanks using river water: A case study from Chengannur, Kerala. *J. Entomol. Zool. Stud.,* **13**, 52–55 (2025).

Urbinati, E.C., J.S. de Abreu, A.C. da Silva Camargo and M.A.L. Parra: Loading and transport stress of juvenile matrinxã (*Brycon cephalus*, Characidae) at various densities. *Aquaculture*, **229**, 389–400 (2004).

Vikas, P.A. and S. Subramannian: Accelerated seed production of pearl spot using modified hatchery method. In: Aquaculture Worker. ICAR–Central Marine Fisheries Research Institute, Kochi, pp. 57–67. ISBN 978 93-82263-32-6 (2020).

Zeppenfeld, C.C., C. Toni, A.G. Becker, D. dos Santos Miron, T.V. Parodi, B.M. Heinzmann, L.J.G. Barcellos, G. Koakoski, J.G.S. da Rosa and V.L. Loro: Physiological and biochemical responses of silver catfish, *Rhamdia quelen,* after transport in water with essential oil of *Aloysia triphylla. Aquaculture*, **48**, 101107 (2014).

Zhang, W., Y. Wang and M. Li: Effects of transportation density and salinity on cortisol, glycogen and lactate of large yellow croaker (*Larimichthys crocea*) juveniles. *J. Fish. China,* **38**, 973–981 (2014).

Online Copy