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# Effect of Different Dietary Lipid Levels on Growth, Feed Utilization, and Health Status of the Pacific White Shrimp *Litopenaeus vannamei* at Two Salinity Levels

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## ABSTRACT

The present experiment was conducted to investigate the effects of five different lipid levels (6,8,10, 12 and 14%) under two different levels of salinity (15 and 30ppt) on growth performance, feed utilization, survival, and economic evaluation of the white leg shrimp *Litopenaeus vannamei* post-larvae. The experiment was carried out in 20 rectangular tanks (66x47x44cm, 50L), with water salinity of 15 and 30ppt over a period of 90 days. Each treatment had 3 replicates; each tank contained 30 post-larvae with an initial body weight of 0.02± 0.001g. The shrimp samples were fed with the experimental diets containing 35% crude protein twice daily till satiation. Moreover, the growth performance of shrimp was recorded biweekly. The best treatment, which contained 6% lipid and salinity of 30ppt, showed the best results in terms of growth performance, feed utilization, survival (%), and economic evaluation under these experimental condition.

## INTRODUCTION

The Pacific white leg shrimp, *Litopenaeus vannamei* **(Boone, 1931)** is the most important cultivated shrimp species and stands out as the highest value among all cultivated crustaceans. The world shrimp production reached a new high estimate of 9.4 million tonnes in 2022 **(FAO, 2023)**. The Pacific white shrimp, *Litopenaeus vannamei*, is a popular species for the commercial aquaculture production in the world due to its high tolerance of salinity from 1 to 50 practical salinity unit (psu) **(Cheng *et al.*, 2006)**. This species has several merits that make it more suitable for aquaculture than the other penaeid shrimp species, such as high-density tolerance, and adaptability to a wide ranges of several environmental parameters, such as salinity and temperature **(Rocha *et al.*, 2010)**.

Protein is the most expensive component in aquafeeds that not only helps in growth but also supplies energy for body function. Excess dietary protein causes environmental pollution in terms of more nitrogenous wastes that induce stress and lead to less production. Thus, to make the aquafeeds cost-effective and environment-friendly, protein level needs to be spared by a non-protein energy source, allowing dietary protein to be utilized for growth. Dietary lipid being a concentrated source of energy can spare the dietary protein. Moreover, dietary lipid acts as a source of essential fatty acids (EFA), phospholipids, steroids, especially cholesterol and carotenoids required for the proper physiological functions of the aquatic animals including shrimp (**Chuphal *et al*., 2021)**.

Lipids have the highest energy density among the three major nutritional components; they are sources of fatty acid, phospholipid, and glycolipid, and act as a source of fat-soluble vitamins **(Xu *et al*., 2018)**. Highly unsaturated fatty acids (HUFAs) have a higher nutritional value to improve the growth of *L. vannamei* at low salinity levels **(Chen *et al*., 2015)**. Phospholipid and glycolipid are both the indispensable components for the cell membrane, which can affect the osmoregulation capacity by changing their contents in the cell membrane **(Tseng & Hwang, 2008)**. Therefore, additional research should be done on the importance of dietary lipids for aquatic species osmoregulation and energy provision.

The optimal dietary lipid levels of *L. vannamei* have been evaluated at several ambient salinities, and the research shows that the optimal level of dietary lipid changes with salinity **(González-*et al.*,2002**; **Zhu *et al*., 2010)**. Some studies have shown that higher dietary lipids can improve the growth and immunity of *L. vannamei* at low salinity levels **(Zhu *et al.*, 2010; Zhang *et al*., 2013)**. However, an excessive dietary lipid may decrease the shrimp growth and increase mortality due to the effect of nutrient imbalance and lipid toxicity owing to lipid oxidation **(Xu *et al*. 2018)**.

The present study was conducted to investigate the effects of five different lipid levels (6, 8 10, 12 and 14%) of *L. vannamei* post-larvae under two different levels of salinity (15 and 30ppt) on growth performance, feed utilization, survival, health status, and economic evaluation.

## MATERIALS AND METHODS

### Experimental diets preparation and feeding regime

Five isonitrogenous practical diets (35%crude protein) were formulated with five levels of crude lipid (6, 8,10, 12 and 14%). The experimental diets were individually prepared via weighing each component and thoroughly mixing the mineral, vitamins and additives with corn. This mixture was added to the components together with oil. Until the mixture was suitable to form granules, water was added. The wet mixture ran through a 2mm diameter CBM granule machine. The produced pellets were dried at room temperature and kept frozen until the experiment started. Ingredient and proximate composition of the experimental diets are presented in Table (1). Shrimp specimens were fed with experimental diets twice a day till satiation.

## Experimental design

The white leg shrimp *L. vannamei* post-larvae were obtained from a commercial shrimp hatchery (Berket Ghalioun, Kafr Al-Sheikh, Egypt). Shrimps were transported in oxygenated double–layered polythene bags. When the shrimp reached the laboratory, they were moved into the acclimation tanks filled with seawater after being filtered by a plankton net (50µm) to prevent the entry of unwanted materials and suspended particles into the tanks, and was then diluted with fresh water to achieve a salinity of15 and 30ppt. Prior to starting the experiment, shrimps were acclimated to laboratory condition for two weeks and fed twice daily with a commercial diet containing 38% crude protein. Initial samples were taken from hatchery immediately after reaching post-larvae, and the final sample was taken from each tank at the end of study for the determination of chemical body composition.

After two weeks of acclimatization, all tanks were stocked with shrimp post-larvae in 2 replicates. Before distributing among tanks, the shrimps were weighed, and the initial body weight (g) was measured. To maintain water quality at the optimum range for shrimps, the following parameters were monitored during the experiment.

**Daily parameters:** Using a multipara meter analyzer, daily measurements of pH, salinity, and water temperature (°C) were taken at 10:00 a.m.

**Biweekly parameters:** Water samples (100ml) were collected from each tank and filtered by filter papers to analyze the total ammonium nitrogen (TAN), nitrite-N (NO2-N), nitrate-N (NO3-N) using a spectrophotometer model (JENWAY 6100).**(APHA, 1998)**.

## Whole-body composition and serum biochemical analysis

Whole-body shrimps and diets were analyzed in triplicate for a proximate composition according to the guidelines of **AOAC (2019)**. Hemolymph samples were collected from five shrimps in each replicate of a treatment (fifteen shrimp per treatment) through the ventral sinus in the first abdominal segment using a 26-gauge hypodermic needle on a 1ml syringe containing 0.3ml of anticoagulant solution. The anticoagulant used in our study was the formulation of **Söderhäll and Smith (1983**), which contains a mix of sodium chloride (0.45M), glucose (0.1M), sodium citrate tribasic dihydrate (30mM), citric acid monohydrate (26mM), and EDTA disodium salt (10mM). Triglyceride (TG), total protein, aspartate transaminase (GOT), and alanine aminotransferase (GPT) were measured with the commercial assay kits. Serum was diluted with a normal saline according to respective preliminary experiments before formal test. After different treatments according to the specification of the kits, the contents of TG, total protein,the activities of GOT and GPT in serum were measured by the methods of **Reitman and Frankel (1957)**,**Reiners *et al.*(1991)** and **Nebot *et al.*(1993)**.

**Table 1.**Formulation and proximate composition of the experimental diets (Dry matter base)

| Ingredient | Treatments | | | | |
|---|---|---|---|---|---|
| | **T1(6%)** | **T2(8%)** | **T3(10%)** | **T4(12%)** | **T5(14%)** |
| Fish meal (70%) protein | 25.00 | 25.00 | 25.00 | 25.00 | 25.00 |
| Soybean meal (44%) protein | 35.00 | 35.00 | 35.00 | 35.00 | 35.00 |
| Yellow corn | 32.50 | 30.50 | 28.50 | 26.50 | 24.50 |
| Sun flour oil | 3.00 | 5.00 | 7.00 | 9.00 | 11.00 |
| Cholesterol | 0.50 | 0.50 | 0.50 | 0.50 | 0.50 |
| Vitamin mixture[1] | 1.00 | 1.00 | 1.00 | 1.00 | 1.00 |
| Mineral mixture[2] | 2.00 | 2.00 | 2.00 | 2.00 | 2.00 |
| Binder | 1.00 | 1.00 | 1.00 | 1.00 | 1.00 |
| Total | 100 | 100 | 100 | 100 | 100 |
| Proximate analysis | | | | | |
| Mositure | 9.61 | 8.96 | 8.51 | 9.54 | 8.84 |
| Protein | 35.71 | 35.54 | 35.37 | 35.19 | 35.02 |
| Lipids | 6.67 | 8.60 | 10.53 | 12.46 | 14.39 |
| Ash | 5.23 | 5.21 | 5.18 | 5.16 | 5.13 |
| Fiber | 3.46 | 3.42 | 3.37 | 3.33 | 3.28 |
| NFE[3] | 48.93 | 47.23 | 45.55 | 43.86 | 42.18 |
| Gross energy (Kcal /100g) [4] | 465.89 | 476.19 | 486.56 | 496.84 | 507.21 |
| P/E (%)[5] | 7.66 | 7.46 | 7.27 | 7.08 | 6.90 |

[1]Each Kg mineral mixture premix contained Mn, 22g; Zn, 22g; Fe, 12g; Cu, 4g; I, 0.4 g, Selenium, 0.4g and Co, 4.8mg.
[2]Each Kg vitamin contained Vitamin A, 4.8 million IU, D3, 0.8 million IU; E, 4g; K, 0.8g; B1, 0.4g; Riboflavin, 1.6g; B6, 0.6g, B12, 4mg; Pantothenic acid, 4g; Nicotinic acid, 8g; Folic acid, 0.4g Biotin,20mg
[3]Nitrogen Free Extract = 100 – (%Protein + %Fat + %Fiber + %Ash).
[4]Gross Energy based on protein (5.65 Kcal/g), fat (9.45 Kcal/g) and carbohydrate (4.11Kcal/g),according to **NRC (2011)**.
[5]Protein energy ratio = Crude protein / Gross energy (Kcal/100g)x100

## Growth performance parameters

Shrimp post-larvae weight (g) was measured at the beginning of the experiment and biweekly by collecting shrimp from each tank, weighing the musingan analytical digital balance, and then returning them back to their tanks. Shrimp weight gain (WG), specific growth rate (SGR) and survival % (S) were calculated according to the following equations:

- **Weight gain (WG**) = Final body weight (g) - Initial body weight (g).
- **Specific growth rate (SGR, %⁄days)** = [(ln FBW - ln IBW) /day of experiment] ×100

- **Survival (%)** = (Final number of shrimp / Initial number of shrimp) ×100.

## Feed utilization parameters

Feed utilization parameters were calculated according to the following equations:

- **Feed Conversion Ratio (FCR)** = Total feed consumption (g)/ weight gain (g).
- **Feed Efficiency (FE %)** = (Final weight – initial / feed consumed ×100.
- **Protein Efficiency Ratio (PER)**= body weight gain (g)/ protein intake (g)

## Economical Evaluation

Cost/kg diet (LE) = Cost per Kg diet L.E

Consumed feed to produce 1kg fish (kg) = Feed intake per fish per period/ final weight per fish Kg/ Kg

Feed cost per kg fresh fish (LE) = Step 1x step 2

Relative % of feed cost/ kg fish= Respective figures for step3/ highest figure in this step

Feed cost/ 1Kg gain (LE) = Feed intake per Kg gain x step 1

Relative% of feed cost of Kg gain= Respective figures for step 5/ highest figure in this step**(Eid & Mohamed, 2008)**.

## Statistical analysis

The data were statistically analyzed by a two way analysis of variance following the method of **SAS (2004)** and using the following model: $Y_{ij} = \mu + T_i + E_{ij}$, where μ is the overall mean; $T_i$ is the fixed effect of $i^{th}$ treatments, and $E_{ij}$ is the random error. Difference between treatments were tested at 5% probability level using Duncan test **(Duncan, 1955)**.

# RESULTS

## Water quality

In the present study, the physico-chemical parameters of water such as temperature (°C), pH, dissolved oxygen (mg/L), nitrite-N (mg/L), and nitrate-N were found to be under the optimum ranges (Table 2). Moreover, the water salinity level was maintained within the range of 15 to 30ppt throughout the experimental period of 90 days.

## Growth performance

In the present study, the dietary lipid levels significantly affected the growth of *L. vannamei*, as shown in Table (3). The group of the white shrimp fed a diet containing 6% lipid had a significantly ($P$<0.05) higher FBW, WG and SGR compared to other experimental groups. Additionally, the group that was fed a diet containing 14% lipid had the lowest values of FBW, WG and SGR. Moreover, the survival rates were influenced by the lipid and salinity levels. Survival rates were at their highest for shrimp at a salanity of 30ppt with diets containing 6% and 8% lipid. In contrast, the lowest survival rate was observed for shrimp fed a diet containing 14% lipid at a 15ppt salinity. In the present study, *L. vannamei* at 30ppt salinity had a better growth performance than at the 15ppt salinity.

**Table 2.** Effect of different levels of lipid and salinity on tank's water quality with *Litopenaeus vannamei* fed experimental diets

| Items<br>Lipid (%)-Salinity (ppt) | Feed utilization parameters | | | | |
|---|---|---|---|---|---|
| | **Temperature°C** | **pH** | **TAN (mg/l** | **$NO_2$(mg/l)** | **$NO_3$ (mg/l)** |
| **6 -30** | 27.8±1.53 | 7.8±0.20 | 0.03±0.01 | 0.02±0.020 | 0.03±0.010 |
| **8 -30** | 28.5±1.53 | 7.8±0.16 | 0.03±0.01 | 0.02±0.010 | 0.03±0.010 |
| **10 -30** | 28.1±1.13 | 7.8±0.32 | 0.03±0.01 | 0.03±0.030 | 0.02±0.010 |
| **12-30** | 28.5±1.03 | 7.9±0.30 | 0.03±0.01 | 0.02±0.010 | 0.03±0.010 |
| **14 -30** | 27.4±0.53 | 7.8±0.30 | 0.03±0.01 | 0.03±0.001 | 0.03±0.002 |
| **6- 15** | 28.0±1.48 | 7.9±0.48 | 0.02±0.01 | 0.02±0.010 | 0.02±0.008 |
| **8 -15** | 28.1±0.47 | 7.7±0.10 | 0.02±0.01 | 0.03±0.018 | 0.02±0.005 |
| **10 -15** | 28.2±1.19 | 7.8±0.19 | 0.02±0.01 | 0.02±0.010 | 0.02±0.080 |
| **12 -15** | 28.7±2.45 | 7.9±0.33 | 0.02±0.01 | 0.03±0.010 | 0.02±0.001 |
| **14 -15** | 28.6±2.30 | 7.8±0.33 | 0.02±0.01 | 0.02±0.010 | 0.02±0.008 |

Data are presented as means ±SD. Values in the same column with different superscript letters are significantly different ($P< 0.05$).

**Table 3.** The growth performance and survival of *Litopenaeus vannamei* fed diets with different levels of lipid and salinity

| **Items**<br>**Lipid (%)-Salinity (ppt)** | **Growth parameters** | | | | |
|---|---|---|---|---|---|
| | **IBW (g)** | **FBW (g)** | **WG (g)** | **SGR (%/day)** | **SR (%)** |
| **6-30** | 0.02±0.011 | 8.63±0.91$^{a}$ | ٨.٦١±٠.٩١$^{a}$ | 6.74±0.67$^{a}$ | 94.66±6.41$^{a}$ |
| **8- 30** | 0.02±0.001 | 8.52±0.07$^{a}$ | ٨.٥٠±٠.٠٦$^{a}$ | 6.73±0.69$^{a}$ | 94.66±4.41$^{a}$ |
| **10-30** | 0.02±0.014 | 8.30±1.40$^{a}$ | ٨.٢٨±١.٣0$^{a}$ | 6.70±0.67$^{a}$ | 92.68±3.55$^{a}$ |
| **12-30** | 0.02±0.005 | 7.84±0.69$^{b}$ | ٧.٨٢±٠.٦٩$^{b}$ | 6.63±0.41$^{b}$ | 92.01±3.48$^{a}$ |
| **14-30** | 0.02±0.003 | 7.29±1.55$^{c}$ | ٧.٢٧±١.٥٤$^{c}$ | 6.55±0.65$^{c}$ | 91.67±3.60$^{b}$ |
| **6- 15** | 0.02±0.005 | 7.52±0.70$^{b}$ | ٧.٥٠±٠.٧٠$^{b}$ | 6.59±0.52$^{b}$ | 91.11±5.35$^{b}$ |
| **8- 15** | 0.02±0.000 | 7.11±1.56$^{c}$ | ٧.٠٩±١.٥٥$^{c}$ | 6.53±0.64$^{c}$ | 90.66±5.00$^{b}$ |
| **10- 15** | 0.02±0.003 | 6.64±0.82$^{c}$ | ٦.٦٢±٠.٨٣$^{c}$ | 6.45±0.61$^{c}$ | 87.19±1.95$^{c}$ |
| **12-15** | 0.02±0.003 | 6.32±0.06$^{d}$ | ٦.٣٠±٠.٠٦$^{d}$ | 6.40±0.59$^{d}$ | 80.20±3.42$^{c}$ |
| **14-15** | 0.02±0.001 | 6.14±1.50$^{d}$ | ٦.١٢±١.٣0$^{d}$ | 6.36±0.42$^{d}$ | 75.11±7.03$^{d}$ |

Data are presented as means ±SD. Values in the same column with different superscript letters are significantly different ($P< 0.05$).

## Feed utilization

Feed efficiency parameters including feed intake (FI), feed conversion ratio (FCR), feed efficiency ratio (FER), and protein efficiency ratio (PER) are illustrated in Table (4). The feed consumption of the shrimp samples decreased significantly at a 30ppt salinity and dietary lipid content of 6 and 8%,compared with the group of shrimp samples reared in a low salinity level (15ppt) and fed diets containing different levels of lipid (6, 8 10, 12, and 14%).

The group of shrimps fed a diet containing 14% lipid showed that PER and FE were significantly ($P$<0.05) the highest in the group of the white shrimp fed a diet containing 6% lipid, and the lowest value was recorded for the group of the white shrimp fed a diet containing 14% lipid, regardless of the salinity level. Feed conversion ratio (FCR) showed that the best significant enhancement for all groups of shrimp fed 6% dietary lipids level at 30ppt salinity compared with groups reared in 15ppt salinity.

**Table 4.** The feed utilization of *Litopenaeus vannamei* fed diets with different levels of lipid and salinity

| **Items Lipid (%)-Salinity (ppt)** | **Feed utilization parameters** | | | |
|---|---|---|---|---|
| | **FI (g feed / shrimp)** | **FCR** | **PER** | **FE (%)** |
| **6 -30** | 10.17±0.24$^{d}$ | 1.18±0.09$^{d}$ | 2.37±0.21$^{a}$ | 84.66±1.72$^{a}$ |
| **8 -30** | 10.23±0.32$^{d}$ | 1.20±0.14$^{d}$ | 2.34±0.35$^{a}$ | 83.09±1.63$^{a}$ |
| **10 -30** | 10.33±0.24$^{d}$ | 1.25±0.33$^{c}$ | 2.27±0.23$^{a}$ | 80.15±1.89$^{a}$ |
| **12-30** | 11.15±0.44$^{b}$ | 1.43±0.23$^{b}$ | 1.99±0.16$^{b}$ | 70.13±1.35$^{b}$ |
| **14 -30** | 10.45±0.45$^{c}$ | 1.45±0.07$^{b}$ | 1.99±0.14$^{b}$ | 69.57±2.3$^{c}$ |
| **6- 15** | 10.40±0.72$^{c}$ | 1.39±0.12$^{c}$ | 2.02±0.25$^{a}$ | 72.12±2.51$^{b}$ |
| **8 -15** | 11.25±0.66$^{a}$ | 1.59±0.06$^{b}$ | 1.77±0.34$^{c}$ | 63.02±2.56$^{c}$ |
| **10 -15** | 11.33±0.93$^{a}$ | 1.71±0.11$^{a}$ | 1.65±0.24$^{c}$ | 58.43±2.42$^{c}$ |
| **12 -15** | 11.36±0.54$^{a}$ | 1.80±0.36$^{a}$ | 1.58±0.36$^{d}$ | 55.46±2.57$^{d}$ |
| **14 -15** | 11.20±0.63$^{b}$ | 1.83±0.22$^{a}$ | 1.56±0.12$^{d}$ | 55.64±2.74$^{d}$ |

Data are presented as means ±SD. Values in the same column with different superscript letters are significantly different ($P$<0.05).

## Whole-body proximate composition and lipid metabolism-related parameters

No differences were detected in the body moisture and body crude protein content among all treatments (Table 5). The body crude lipid increased with the increasing dietary lipid content at 30ppt salinity($P$<0.05). The highest body crude lipid of shrimp was found at 30ppt fed adiet with 14% lipid. A significant difference ($P$<0.05) was found in the body ash content among five diets. The Body ash contents of shrimp at 14% lipid and 30ppt salinity was significantly higher than those of the shrimps at 10% lipid and15ppt salinity. Similar to the body crude lipid, the highest content of the serum TG was found in the shrimp fed diet with 14% lipid at 30‰ ppt salinity, while the lowest TG was found in the shrimp fed 6% lipid at 15ppt salinity. The serum TG increased upon increasing the dietary lipid content, regardless of salinity (Table 6). No differences were found in the serum protein content among all treatments. The serum GOT and GPT contents of both showed the lowest value in the shrimp fed 10% lipid at 15ppt salinity. The highest contents of both the GOT and GPT contents were found in the shrimp fed 14% lipid at 30ppt salinity.

**Table 5.** Proximate composition of *Litopenaeus vannamei*(%/wet weight) acclimated to two different water salinities and fed different lipid levels

| Lipid (%)- Salinity (psu) | Moisture (%) | Crude protein (%) | Crude lipid (%) | Ash (%) |
|---|---|---|---|---|
| **6- 30** | 75.20 ±9.9 | 17.48 ±1.8 | 26.1 ±1.6$^{c}$ | 31.4 ±4.4$^{c}$ |
| **8-30** | 74.20 ±3.9 | 17.34 ±2.4 | 25.4 ±4.5$^{d}$ | 32.9 ±1.4$^{c}$ |
| **10-30** | 74.75 ± 1.7 | 17.09 ±1.8 | 26.2 ±3.2$^{c}$ | 33.6 ±1.8$^{b}$ |
| **12-30** | 75.12 ± 3.7 | 17.18 ±4.8 | 27.9 ±2.7$^{b}$ | 34.0 ±0.5$^{b}$ |
| **14-30** | 74.17 ± 5.6 | 17.34 ±5.0 | 28.3 ±1.5$^{a}$ | 35.1 ±0.3$^{a}$ |
| **6-15** | 75.10 ± 1.9 | 17.49 ±1.5 | 18.4 ±2.6$^{f}$ | 28.8 ±1.4$^{e}$ |
| **8-15** | 76.20 ± 2.5 | 17.44 ±1.9 | 19.5 ±4.7$^{ef}$ | 29.0 ±1.2$^{d}$ |
| **10-15** | 74.20 ± 3.8 | 17.32 ±3.6 | 21.8 ±3.3$^{e}$ | 28.2 ±0.6$^{e}$ |
| **12-15** | 75.10 ± 6.9 | 17.07 ±1.4 | 24.0 ±3.8$^{d}$ | 28.6 ±0.6$^{e}$ |
| **14-15** | 74.20 ± 8.7 | 17.17 ±1.6 | 26.4 ±1.8$^{c}$ | 28.9 ±0.3$^{e}$ |

Values (mean ± SEM) are mean of 6.Values within a column without a common superscript letters are different (c: indicated the highest value); $P$<0 .05 means significant difference.

**Table 6.** Serum contents of TG, protein and activities of GOT and GPT of *L. vannamei* fed diets containing different lipid levels at two salinities

| Lipid (%)- Salinity (psu) | TG (mmol/L) | Protein (mg/ml) | GOT (U/L) | GPT (U/L) |
|---|---|---|---|---|
| **6-30** | 1.89 ± 0.49 | 45.52 ± 2.55 | 14.93 ± 1.02$^{b}$ | 33.44 ± 3.64$^{c}$ |
| **8-30** | 2.65 ± 0.21 | 44.23± 3.18 | 14.50 ± 0.89$^{b}$ | 32.91 ± 1.93$^{d}$ |
| **10-30** | 3.00 ± 0.11 | 43.40 ± 3.78 | 13.85 ± 0.89$^{b}$ | 32.71 ± 2.53$^{d}$ |
| **12-30** | 4.11 ± 0.43 | 44.31 ± 2.53 | 17.76 ± 0.15$^{a}$ | 35.79 ± 3.56$^{b}$ |
| **14-30** | 4.65 ± 0.13 | 43.00 ± 2.53 | 18.16 ± 0.45$^{a}$ | 36.19 ± 1.76$^{a}$ |
| **6-15** | 1.02 ± 0.18 | 44.55 ±7.17 | 15.30 ± 1.61$^{b}$ | 36.21 ± 1.98$^{a}$ |
| **8-15** | 1.64 ± 0.17 | 45.03 ± 1.27 | 13.83 ± 1.29$^{c}$ | 29.17 ± 1.19$^{f}$ |
| **10-15** | 1.85 ± 0.57 | 44.00± 1.30 | 11.52 ± 1.29$^{c}$ | 21.27 ± 1.12$^{g}$ |
| **12-15** | 2.06 ± 0.22 | 45.80 ± 2.61 | 12.86 ± 2.84$^{d}$ | 31.37 ± 3.55$^{e}$ |
| **14-15** | 2.89 ± 0.27 | 43.10 ± 3.61 | 12.90 ± 2.84$^{d}$ | 32.06 ± 4.25$^{d}$ |

Values are means ± SD. Values in the same row with different superscripts are significantly different ($P$< 0.05).

**Economic evaluation**

Results of the economic evaluation including feed costs of one kg gain in weight and its ratio to that of the highest group are presented in Table (7). It was noticed that, the feeding price increases proportionally with increasing the lipid levels in the diets, each two percent of lipid level increases by 0.50% for the diet price. However, using only the cost of feeding is not an adequate parameter to determine the best feeding strategy for shrimp culture since it does not relate costs to the biomass produced. The economic feed conversion rate (EFCR) represents the total feeding cost biomass produced, which is a better estimate to determine the cost-benefit of the diets. In the present study, the economic evaluation proved that the group of shrimp fed on diet containing 6% lipid at 30ppt has the lowest feed cost per kg fresh shrimp (17.64 LE), with a relative % of feed cost/ kg of the shrimp 57%, whereas the group of shrimp fed on diet containing14% lipid at 15psu has the highest feed cost per kg of the fresh shrimp (30 .85 LE).

**Table 7.** Effects of different lipid levels on economic analysis of *L. vannamei* post-larvae

| Item | Lipid levels % | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | $T_1$ (6%) | | $T_2$(8%) | | $T_3$ (10%) | | $T_4$ (12%) | | $T_5$ (14%) | |
| **Salinity ppt** | **15** | **30** | **15** | **30** | **15** | **30** | **15** | **30** | **15** | **30** |
| **Cost /kg of diet (LE)** | 14.95 | 14.95 | 15.45 | 15.45 | 15.95 | 15.95 | 16.45 | 16.45 | 16.95 | 16.95 |
| **Feed intake** | 10.40 | 10.17 | 11.25 | 10.23 | 11.33 | 10.33 | 11.36 | 11.15 | 11.20 | 10.45 |
| **Feed intake cost (LE)[1]** | 155.5 | 152.0 | 173.8 | 158.1 | 180.7 | 164.8 | 188.9 | 183.4 | 189.8 | 177.1 |
| **Relative to feed cost %[2]** | 82 | 80 | 92 | 83 | 95 | 87 | 99 | 97 | 100 | 93 |
| **Consumed feed to produce 1kg of shrimp (kg)[3]** | 1.39 | 1.18 | 1.59 | 1.20 | 1.71 | 1.25 | 1.80 | 1.43 | 1.82 | 1.45 |
| **Feed cost per kg of fresh shrimp (LE)[4]** | 20.78 | 17.64 | 24.57 | 18.54 | 27.27 | 19,94 | 29.61 | 23.52 | 30.85 | 24.58 |
| **Relative % of feed cost/ kg of shrimp[5]** | 67 | 57 | 79 | 60 | 88 | 64 | 96 | 76 | 100 | 80 |

[1]Feed cost X feed intake.
[2]Value of each treatment feed intake cost /highest value x100.
[3]Feed conversion ratio.
[٤]Feed cost/ kg gain = FCR x Feed cost/kg in EGP.
[5]Feed cost /kg fresh fish EGP value for each treatment/ highest value x100.

## DISCUSSION

The current study was carried out to optimize the dietary lipid levels of *P. vannamei* juveniles under two salinity values (15 and 30ppt). Inland saline water is a new diversified aquaculture resource where euryhaline species like *P. vannamei* can be suitably cultured. In this study, performances of the shrimp such as growth, nutrient utilization and physio-metabolic responses were evaluated in relation to the graded levels of dietary lipid under the rearing condition at 15 and 30g/L salinity.

Water quality plays a pivotal role as it directly or indirectly governs the survival of organisms in the aquatic ecosystem. In the present study, the water temperature, pH and dissolved oxygen of the culture system were reported within the favorable range of *P. vannamei* cultivation **(Bett & Vinatea, 2009)**. *P. vannamei* is capable of tolerating a wide range of salinities ranging from 0.5 to 40g/L due to its capability to maintain an osmotic regulation (**Saoud *et al*., 2003)**. However, the isosmotic point of *P. vannamei* lies within the range of 21.1 to 26.1g/L **(Jaffer *et al*., 2020)**. Moreover, salinity fluctuation can affect the growth and nutrient utilization of the shrimp **(Maicá *et***

***al*., 2014)**. The water salinity of the present study was maintained at around 15 and 30g/L. Moreover, in the present study, the total ammonium-nitrogen, nitrite-nitrogen, nitrate-nitrogen concentration were found to be within the recommended range **(Talukdar *et al*., 2020)**.

In the present study, dietary lipid levels significantly affected the growth of *L. vannamei*, as shown in Table (3). The group of the white shrimp fed a diet containing 6% lipid had significantly ($P$<0.05) recorded a higher FBW, WG and SGR compared to the other experimental groups, while the group that was fed a diet containing 14% lipid had lower values of FBW, WG and SGR. The highest WG and SGR of the post larval *L. vannamei* were observed at 6% lipid, consistent with findings reported by **González-Félix *et al.* (2002)**, where a dietary lipid level of 60g $kg^{-1}$ at 25ppt salinity was regarded as optimal for *L. vannamei*. However, at 3psu, *L. vannamei* fed 90g $kg^{-1}$ lipid had the best growth performance **(Xu *et al*., 2018)**. There is a wide variety for the dietary lipid requirement of *L. vannamei*. In some research, *L. vannamei* samples were fed diets containing 81.0– 112.0g/kg of lipids for 8 weeks **(Wouters *et al*., 2001)**, or diets containing 30.0– 90.0g/kg lipids for 6 weeks **(González-Félix *et al*., 2002)**. There is a wide variety for the dietary lipid requirement of *L. vannamei* in the study research. Another interesting study indicated that shrimp fed a diet of 120.0g/kg lipid level for 30 days and 100.0g/kg lipid level for 60 days were optimal for the growth of *L. vannamei* **(Zhang *et al*., 2013)**. The dietary lipid requirement may be complex and related to many factors, such as diet composition, temperature, and salinity **(Xie *et al*., 2019)**. Moreover, PER and FE were significantly ($P$<0.05) higher in the group of the white shrimp fed a diet containing 6% lipid and lower in value for the group of the white shrimp fed a diet containing 14% lipid, regardless of salinity. The WG and SGR of shrimp at 30ppt recorded a higher value than those fed with the same diet at 15ppt ($P$<0.05). Salinity had a significant influence on the survival of the test shrimps ($P$<0.05), as reported by **Walker *et al.* (2009)**, *L. vannamei* had a higher growth rate at 28psu than shrimps at 2psu. The distinctive results may be related to the initial shrimp size, moult frequency, or the different dietary nutrient compositions **(Ding *et al*., 2008)**, but more work should be further conducted to demonstrate the difference in these reports.

The survival rate of *P. vannamei* juveniles in the current study was significantly lower in higher lipid (100 and 120g/kg) fed groups compared with their lower lipid fed counterparts. In accordance with this finding, it was reported that excess dietary lipid probably could be detrimental to the shrimp in relation to growth and survival **(Toledo *et al*., 2016)**,although a survival percentage of more than 80% was reported to be good for crustacean culture **(Cuzon& Guillaume, 1997)**.

As exibited in Table (5), the highest value of the body crude lipid content occurred in the shrimp fed diet with 14% lipid at 15 and 30ppt. Some studies also showed that shrimp and some other crustaceans have a limited capacity to digest high dietary lipid **(Glencross *et al*., 2001)**, and excess lipid may lower shrimp growth due to the lipid peroxidation and imbalance of dietary nutrition **(Nogueira *et al.*, 2003)**. The serum triglyceride content has a close relationship with the dietary lipid level **(Jacobs & Barrett-Connor, 1982)**. In this study, the serum triglyceride in shrimp at 15 and 30ppt was positively related to the dietary lipid levels, which is consistent with the findings in other crustaceans **(Hu *et al.*, 2008)**.

Aspartate transaminase (GOT) and alanine aminotransferase (GPT) are usually used as biochemical markers to determine hepatopancreatic damage **(Wu*et al.*, 2008)**. The change in GOT and GPT activities in the serum is related to the hepatopancreatic disorder in *L. vannamei* **(Wu *et al*., 2008)**. The activities of GOT and GPT in shrimp fed 6% lipid at low salinity were both significantly higher than those of the shrimp fed 9 and 12% lipids. The damage of hepatopancreas caused by salinity stress can be alleviated through increasing the dietary lipid content **(Lightner & Redman, 1994)** since more dietary lipid can supply more energy for metabolism and can enhance the oxygen-carrying capacity in the shrimp **(Li *et al.*, 2015)**.

Furthermore, In the present study, the feeding price increases proportionally with the lipids levels on diets; each percent of lipid level increases by 0.25% on diet price. However, using only the cost of feeding is not an adequate parameter to determine the best feeding strategy for shrimp culture, since it does not relate costs to the biomass produced. The economic feed conversion rate (EFCR) represents the total feeding cost biomass produced, which is a better estimate to determine the cost-benefit of the diets. In the present study, the economic evaluation proved that the group of shrimp fed diet containing 6% lipid at 30pp has the lowest feed cost per kg of fresh shrimp (17.64 LE). Moreover, the relative% of feed cost/ kg of shrimp was 57%, which aligns with the findings of the study of **Eid *et al*. (2024)**.

## CONCLUSION

It could be concluded that the treatment containing 6% lipid at salinity 30ppt was the best in terms of growth performance, feed utilization, survival (%), health status, and economic evaluation under these experimental conditions.

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