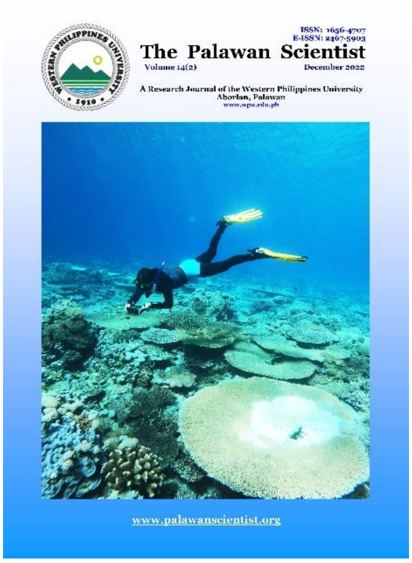
Original Article





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# Growth and survival of juvenile gold-lip pearl oyster *Pinctada maxima* (Jameson, 1901) at different depths with and without regular cleaning

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## ABSTRACT

The lucrative commercial culture of gold-lip pearl oyster *Pinctada maxima* (Jameson, 1901) for pearl production has been in existence in the Philippines for several decades, however, no growth studies for this species has been published in the country as of this writing. To fill this gap, the results of two consecutive 60-day growth trials were conducted in the island province of Palawan. The first experiment (E1) examined the growth and survival of 4-month-old hatchery-produced pearl oysters in net trays (200 individuals per tray or 583 individuals $m^{-2}$) hung in a long line at three different depths (2, 4, and 6 m) below the water surface subjected to cleaning and without cleaning regimes. The second experiment (E2) was a continuation of E1, except that the 6-month-old pearl oysters were raised in 30-individual pocket net baskets. Average shell length increments (SLI) and survival rates (SR) in E1 did not significantly vary among depths ($P > 0.05$) and between cleaning conditions ($P > 0.05$). In E2, the SLI did not significantly vary among depths ($P > 0.05$) and between cleaning conditions ($P > 0.05$), while the SR was statistically similar among depths ($P < 0.05$) but not between cleaning conditions ($P > 0.05$). The results suggest that instead of the usual single row, the three rows of net baskets at different depths and the absence of cleaning could be considered in the early stage of gold-lip pearl oyster farming.

**Keywords:** biofouling, intermediate culture, long line method, Palawan, Philippines

## INTRODUCTION

Among the different pearl oyster species used in the pearl farming industry (Southgate et al. 2008; Tisdell and Poirine 2008; Nagai 2013; Cartier and Carpenter 2014; Zhu et al. 2019; Johnston et al. 2022) the gold-lip pearl oyster *Pinctada maxima* (Jameson, 1901) is popularly cultured in Australia and many other countries (Taylor 1999; Yukihira et al. 2006; Tisdell and Poirine 2008) including the Philippines (Bondad-Reantaso et al. 2007). Of the 30 registered pearl farms in the Philippines (Bondad-Reantaso et al. 2007), 11 farms are found in Palawan (Baltazar and Dalusung-Rodrigues 2016) engaging in the rearing of *P. maxima*.

The pearl oysters are cultured using a long line method, where a series of spherical or cylindrical floats are attached to long lines moored with anchors to keep the distances between the rows of ropes and floats. Long lines in some pearl farms in Palawan are installed at depths ranging between 25 and 55 m, while only single rows of net baskets are hung 3-12 m below the water surface, thus creating plenty of available spaces below the grow-out farms. Pearl oysters inhabit clear waters under the influence of currents at depths up to 60 m but are most common from 5 to 30 m (Poutiers 1998). They can grow fast within 5 m deep (Lee 2010). The large spaces under the rows of long lines could be maximized by having two or three rows of suspended net baskets at different depths to potentially help reduce the leased area without affecting the production of pearls.

The growth out culture of pearl oyster requires regular removal of biofouling organisms to improve the growth and survival rates (SR). However, this activity constitute a major cost in pearl farm operations (Taylor et al. 1997b). Several studies however, have found that the effects of biofouling vary between localities and sizes of cultured species (Southgate and Beer 2000; Milione and Southgate 2011; Cueba et al. 2022). Hence, the economic costs associated with biofouling control and prevention could be minimized by understanding its site-specific influence on the cultured pearl oysters.

While many growth studies have been published about *P. maxima* (Taylor et al. 1997a, b; Lee 2010; Deng et al. 2013; Hao et al. 2018), no growth studies for the species has been done in the Philippines, in spite of the country's significant contribution to the global pearl production (Zhu et al. 2019). This study determined the growth and survival of hatchery-produced *P. maxima* hanged at 2, 4, and 6 m below the water surface, subjected to cleaning and without cleaning conditions.

## METHODS

### Study Site

The two 60-day experiments were conducted in the grow-out facility of a small-scale pearl farm in Honda Bay, Puerto Princesa City (Figure 1). The grow-out area is about 8-10 m deep with moderate wave action, having an annual average water temperature (31.11 ± 1.26°C) and salinity (34.58 ± 0.37 ppt) falling within the optimum requirements for pearl oyster farming (Lucas 2008; Deng et al. 2013).

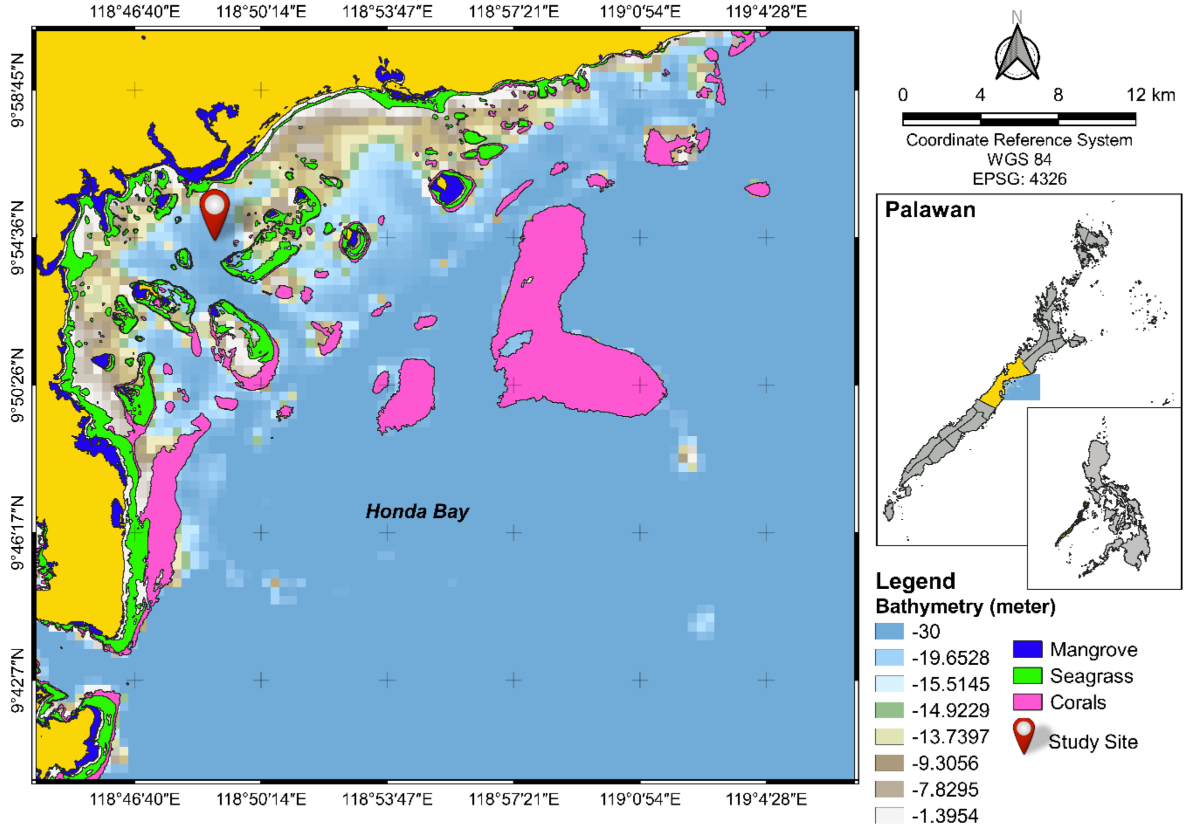


**Figure 1.** Map of Honda bay in Puerto Princesa City, Palawan showing the site ( ) of the study, the nearby coastal ecosystems and the elevation of the coast.

**Experimental Design**

The first (E1) and second (E2) experiments used a 3 x 2 factorial design. For E1, 4-month-old hatchery-produced *P. maxima* measuring 30.22 ± 3.73-32.59 ± 3.92 mm, average shell lengths (SL) were raised in net trays (48.26 cm x 71.12 cm) hanged at three different depths (2, 4, and 6 m) in a long line, subjected to two management options (with and without monthly cleaning). Each net tray served as a replicate contained 200 individuals (583 ind. $m^{-2}$) pearl oysters (Table 1). Each tray was wrapped with a 5 mm meshed-size black net to keep the pearl oysters inside the tray. The trays subjected to cleaning and without cleaning conditions were alternately suspended at 1-meter interval in a 20-m vacant longline of the pearl farm. The net trays were triple-hung vertically at 2-m intervals. The experiment was terminated after 60 days. During monthly sampling, pearl oysters that received monthly cleaning underwent manual removal of marine growth and other unwanted species. Net used for wrapping the trays was replaced every 30 days to ensure adequate water exchange essential for the growth and survival of the cultured species.

The second experiment (E2) was also conducted for another 60 days (Table 2), involving a total of 540 individuals taken from E1. The density was 30 individuals (ind.) per net basket (48.26 cm x 71.12 cm) or 87 ind. $m^{-2}$. The net baskets were triple-hung at 2 m interval with three replications. Net baskets subjected to cleaning and without cleaning conditions were alternately suspended at 1-meter interval within a 20-m longline. Monthly cleaning was carried out using a pressurized water sprayer without removing the pearl oysters on the net baskets. The net baskets without regular cleaning were inspected monthly to dislodge any predatory species. The study was terminated after 60 days.

**Sampling**

Due to the delicate nature of the young oyster shell, the initial measurement of shell lengths (Hwang et al. 2007) in E1 only involved 10 samples from each replicate. Succeeding monthly sampling for E1 and all samplings for E2 involved 20 individuals per replicate. The SL were measured using a caliper to a precision of 0.01 mm. Monthly survival was determined by counting the number of live pearl oysters.

**Data Analysis**

The average monthly SL from each replicate of both E1 and E2 were computed. After which, the SL increment was obtained by subtracting the averages of the preceding month to the next month's data. Shell length increments (SLI) and SR were transformed as needed to satisfy the test for normality before conducting a 2-way ANOVA. The survival in E2 did not meet the test for normality even after data transformations; hence comparison was carried out using the non-parametric Kruskal Wallis test. All computations were carried out using the SPSS trial version.

**Table 1**. The first experimental (E1) set-up used for the 4-month-old gold-lipped pearl oyster *Pinctada maxima* raised in net trays from August to October 2017.

| Particulars | With cleaning | | | Without cleaning | | |
|---|---|---|---|---|---|---|
| Depth (m) | 2 | 4 | 6 | 2 | 4 | 6 |
| Replication | 3 | 3 | 3 | 3 | 3 | 3 |
| Initial stock per replicate or net bag | 200 | 200 | 200 | 200 | 200 | 200 |
| Density (ind. m-$^2$) | 583 | 583 | 583 | 583 | 583 | 583 |
| Initial average (±sd) shell length (mm); n = 10 | 31.43 (±4.24) | 31.47 (±4.23) | 31.62 (±4.68) | 30.22 (±3.73) | 32.59 (±3.92) | 30.39 (±4.30) |

**Table 2**. The second experimental (E2) set-up used for the 6-month-old gold-lip pearl oyster *Pinctada maxima* raised in 30-pocket net baskets from October to December 2017.

| Particulars | With cleaning | | | Without cleaning | | |
|---|---|---|---|---|---|---|
| Depth (m) | 2 | 4 | 6 | 2 | 4 | 6 |
| Replication | 3 | 3 | 3 | 3 | 3 | 3 |
| Initial stock per replicate or net basket | 30 | 30 | 30 | 30 | 30 | 30 |
| Density (ind. $m^{-2}$) | 87 | 87 | 87 | 87 | 87 | 87 |
| Initial average (±sd) shell length (mm); n = 20 | 43.80 (±5.58) | 42.20 (±5.47) | 44.85 (±5.47) | 43.50 (±4.81) | 44.30 (±4.65) | 44.18 (±5.34) |

## RESULTS

### Growth and Survival in Net Trays for Experiment 1

The *P. maxima* grew at a similar rate from 30.22-32.59 mm initial SL into 42.20-44.85 mm after 60 days of culture (Figure 2). The average monthly SLI (5.36-6.89 mm; Figure 3) did not significantly vary among depths ($P < 0.05$), between cleaning conditions ($P > 0.05$) and depth x cleaning conditions ($P > 0.05$).

The SR largely dropped (32.67-51.00%) during the first 30 days, followed by a minimal decline (24.67-45.67%) on the 60th day (Figure 4). The SR did not significantly vary among depths ($P > 0.05$), between cleaning conditions ($P > 0.05$) and depths x cleaning conditions ($P > 0.05$).

### Growth and Survival in 30-pocket Net Baskets for Experiment 2

The growth was relatively increasing within the culture period. From an average initial SL ranging between 42.20 and 44.85 mm, the pearl oyster reached 60.01-66.54 mm after 60 days of culture (Figure 5). The average monthly SLI ranged between 7.86 and 10.99 mm (Figure 6). The SLI did not significantly vary among depths ($P > 0.05$), between cleaning conditions ($P > 0.05$) and depth x cleaning conditions ($P > 0.05$).

The average SR in E2 was relatively high (67.78-98.89%; Figure 7) and did not significantly vary among depths ($P > 0.05$) but differed between cleaning conditions ($P < 0.05$).

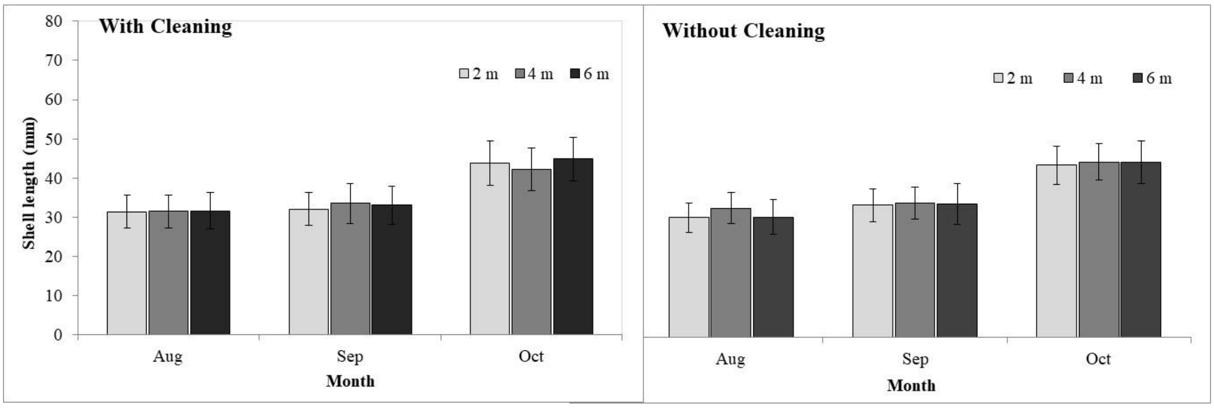


**Figure 2**. Monthly average (±sd) shell lengths of 4-month-old *Pinctada maxima* kept in net trays hung in a long line at three different depths subjected to cleaning and without cleaning conditions.

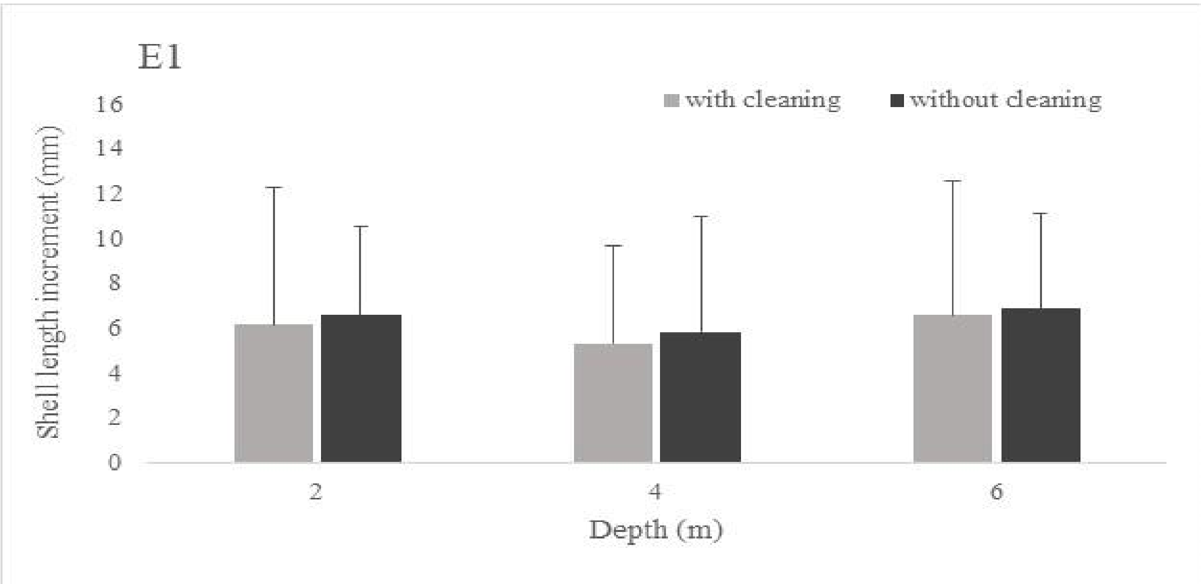


**Figure 3.** Average (±sd) shell length increments of 4-month-old *Pinctada maxima* kept in net trays for 60 days, subjected to cleaning and without cleaning conditions.

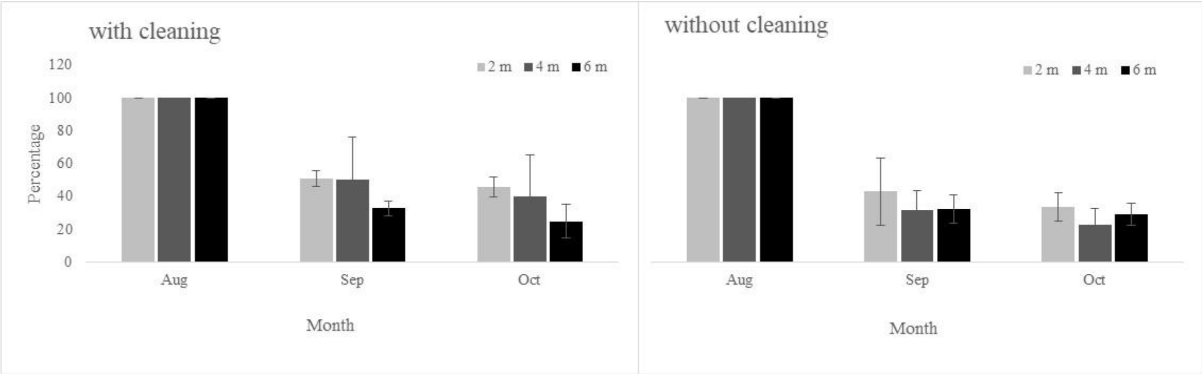


Figure 4. Average (±sd) survival rate of 4-month-old *Pinctada maxima* kept in net trays hung in a long line at three different depths, subjected to cleaning and without cleaning conditions.

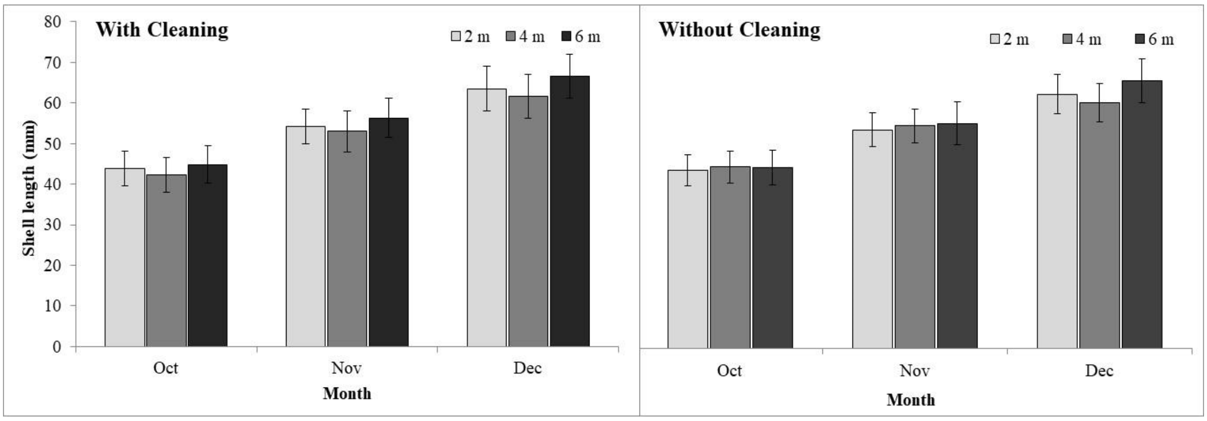


**Figure 5.** Monthly average (±sd) shell lengths of 6-month-old *Pinctada maxima* kept in 30-pocket net baskets hung in a long line at three different depths subjected to cleaning and without cleaning conditions.

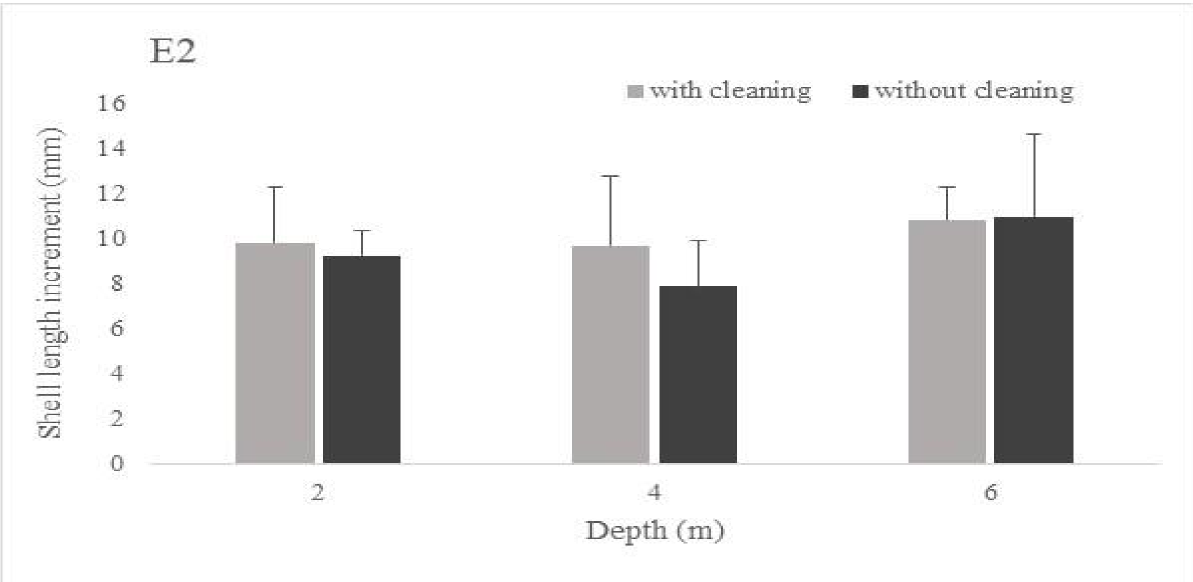


**Figure 6.** Average (±sd) shell length increments of 6-month-old *Pinctada maxima* held in 30-pocket net baskets for 60 days, hung in a long line at three different depths subjected to cleaning and without cleaning conditions.

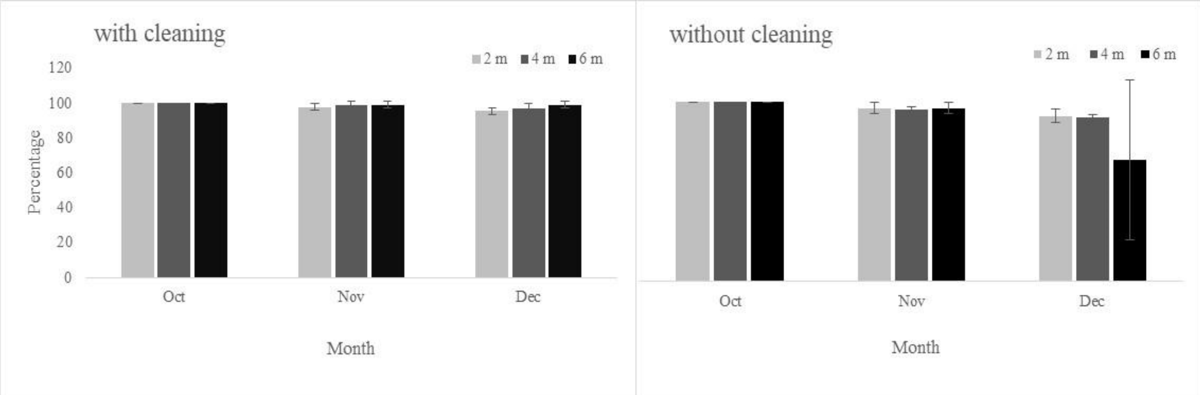


**Figure 7.** Survival rates of 6-month-old *Pinctada maxima* raised in 30-pocket net baskets at three different depths in a long line from October to December 2017.

## DISCUSSION

### Growth

The growth rates among *P. maxima* were highly variable. In E1, the pearl oyster having initial SL of 30.21-32.59 mm, reached 43.50-44.18 mm in two months, representing a monthly increment ranging between 5.36 and 6.89 mm. While in the study of Yukihira et al. (2006) pearl oyster measuring about 29.2 mm shell height, had slower growth. It only reached 40 mm shell height after 7 or 8 months (~1.54 mm increment per month). As for the case of E2, the pearl oyster reached 60.01-66.53 mm after 60 days, representing 7.50-8.31 mm monthly SLI. The final size we obtained in E2 or when the pearl oysters were 8-month-old, were comparable to the initial size (59.2 ± 0.5 mm shell height) of 1-year-old *P. maxima* placed in 10-pockets panel nets suspended at 3 m from the long line (Taylor et al. 1997a). If the monthly SLI increment in E2 is maintained in the succeeding four months, the pearl oyster would measure 90.00-99.72 mm after reaching 1-year of age. This estimate is about 10 mm shorter compared to the observed sizes of fast-growing 1-year old *P. maxima* (100-110 mm) in some commercial farms in Palawan.

The fast growth and absence of significant variation among growths at three depths for both E1 and E2 suggest the abundance of food in the study area. While no plankton monitoring was conducted to validate this claim, the presence of thick mangrove forest and estuaries within Honda Bay is favorable to the growth of natural food for oyster (Saifullah et al. 2015; Lan et al. 2021). The sea surface waters contain abundant and diverse phytoplankton species than deeper areas (Taylor 1999), thus promoting faster growth for pearl oysters (Haws 2002; Lee 2010) and of other bivalve species (Ogilvie et al. 2000; Joubert et al. 2014). Similarly, Yukihira et al. (2006) attributed the better growth of *P. maxima* to the high suspended particulate matter.

Biofouling can cause adverse effect on the growth of pearl oyster (Taylor et al. 1997b; Pit and Southgate 2003). However, there was no significant variation in the growth of *P. maxima* subjected to cleaning and without cleaning conditions. Several studies also reported that the absence of regular cleaning does not affect the growth of black-lip pearl oyster *Pinctada margaritifera* (Lacoste et al. 2014; Hulot et al. 2019; Cueba et al. 2022).

In Pioneer Bay, Orpheus Island, north Queensland, Australia, depth had no significant effect on growth, survival or fouling of *Pteria penguin* cultured for six months in three types of culture units deployed at 3 and 6 m deep (Milione and Southgate 2011). In the study site, the biofouling organisms were mostly hydroids, macro-algae and sponges which did not adversely affect the oyster's feeding activity and growth in general. The barnacles and other oyster species which can reduce the growth and cause mortalities on pearl oysters (Fitridge et al. 2012) were not noted during the study.

### Survival

The low survival in E1 (24.67-45.67%) is comparable to the study of Yukihira et al. (2006) which obtained 30-50% survival after 14 months for *P. maxima* (29.2 and 28 mm shell heights) raised in two dissimilar environments in the Great Barrier Reef lagoon. However, these mortalities were attributed to low temperature during the winter season. The net trays of E1 was covered with net, hence it is unlikely that the mortalities were caused by predatory crab such as the *Acanthocyclus albatrossis.* Predatory crab feeds on the recruits and seeds of other bivalve such as the mussel *Mytilus chilensis* (Uzkiaga et al. 2022). It is speculated that the mortality could have been associated with density and suffocation from accumulated dirt on the trays' net cover as also been observed in several studies (see Southgate 2008). The reduction in density as an effect of 32.67-51.00% SR during the first 30 days could have helped reduce the stress brought about by overcrowding and suffocation, the reason for improved SRs (24.67-45.67%) on the 60th day. In this instance, the survival could be improved by reducing the density and regular change of net cover. Grading and density reduction also promoted higher survival among small groups of *Pinctada martensii* (Fan et al. 2021), groupers (Villanueva et al. 2021a) and siganids (Villanueva et al. 2021b).

As expected, the survival in E2 was higher (Figure 6) than in E1. Survival rates for pearl oysters tend to increase with age or size as also been observed for the pearl oyster *Pteria hirundo* (Albuquerque et al. 2012) and many other organisms (Pauly 1998; Ridgway et al. 2011). The significant variation in the SR between cleaned and uncleaned trays could have been influenced by the low survival in one of the replicates of uncleaned net baskets at 6 m deep. Only 5 ind. (16.67% survival) remained in one of the replicates while the other two replicates had 28 ind. (93.33% survival). The hairy triton (*Cymatium* spp.), considered as serious pest in pearl farms (see Humphrey et al. 1998), occurred in each net basket conditions. However, its low number (3-5 ind. per cleaned net basket and 3-6 ind. per uncleaned net basket) may not be associated with the observed high mortality. The crab *Charybdis* sp. also considered as serious pest (see Humphrey et al. 1998), but these were not noted during the study. Another pest in pearl oyster is the polycad flatworm *Stylochus* sp. which had caused the isolated 100% mortality in one of the replicates for the cultured rainbow pearl oyster *Pteria sterna* (Monteforte et al. 2005), but these was also not observed in the net basket. Inventory of predatory species in a pearl farm could aid in deciding specific farm management strategies.

The absence of significant variation among SR at three different depths suggests that during the early stage of pearl oyster culture, the net baskets could be tripled hung in a long line at 2-6 m deep. Similar studies involving larger or older individuals is suggested to maximize the use of space occupied by the pearl farms. An image analysis method such as the use of Coral Point Count with Excel extension (CPCe) software is highly recommended to increase the number of measurements on oysters without manipulating the samples one by one.

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