

# Photoperiodic influence on larval release and mortality in *Leptomysis lingvura* (Mysidacea: Crustacea)

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## ABSTRACT

The possible influence of light on *Leptomysis lingvura* larval production and mortality was examined in this study. Considering light as a potential factor affecting physiology, organisms were subjected to three different photoperiods: 24 hours at dark, 24 hours at light and 10:14 light:dark. Number of juveniles and mortality were daily measured during the fifteen-day experiment. Statistical results showed no significant differences for total breeding or mortality among the three photoperiods. Nevertheless, significant differences among 24 hours light and 10:14 photoperiods were established after the tenth day.

**Keywords:** *Leptomysis lingvura*, photoperiod, light, larval production, mortality.

## RESUMEN

Se estudia la posible influencia de la luz en la producción y mortalidad de las larvas de *Leptomysis lingvura*. Considerando la luz como un factor potencial que puede afectar a la fisiología, los organismos fueron sometidos a tres diferentes fotoperíodos: 24 horas en oscuridad, 24 horas a la luz y 10:14 de luz:oscuridad. Se contabilizó diariamente el número de larvas y la mortalidad de las mismas, durante quince días. Los resultados estadísticos no muestran diferencias para la producción o mortalidad larvaria entre las tres condiciones de fotoperíodo. No obstante, se establecieron diferencias significativas entre las condiciones de 24 horas de luz y 10:14 después del décimo día.

**Palabras clave:** *Leptomysis lingvura*, fotoperíodo, luz, producción larval, mortalidad.

## INTRODUCTION

Photoperiod is known to be a factor synchronizing reproduction of terrestrial plants (Vince-Prue, 1975) and animals (Farner & Follett, 1979), generally by inducing gonadal development (Farner & Follett, 1966). Indeed, most marine animals have seasonal reproductive cycles (Pearse & Eernisse, 1982) and some species present larval release coincident with lunar, diel and tidal cycles (DeCoursey, 1983; Forward, 1987; Saigusa & Kawagoye, 1997). But, the possible influence of light on offspring production remains unclear.

In mysids, larval development within a single marsupium takes on the average of 14.9 days at 16 °C (Wittman, 1981) and spawning seems to occur over one or maximum two nights (Green, 1970; Mauchline, 1980). As Johnston & Ritz (2001) confirmed if females do not play a role in synchronizing the development of their broods, some unknown factor(s) such as temperature, salinity or photoperiod must be involved. Nevertheless, how light exposure or total light depletion may affect mysid physiology or behavior and, even more, how it may determine the number of eggs produced and the time of spawning has not established yet.

Mysids are shrimp-like with a length that varies among 2-30 mm, but some species as *Gnathophausia* can reach 35 cm long (Ruppert & Barnes, 1996). In the oceans, mysids are rather common in shallow coastal waters, being more frequent sandy or muddy bottom (Murano, 1999) than in the water

column, although they can be found in different environments.

*Leptomysis lingvura* is easily identified from the other mysid species because they present specific features. This species is characterized by a short carapace with a very short face and a triangular tip. Antennal scale is long and segmented into two with the terminal segment containing between nine and twelve setae. Telson is short and wide, tongue-shaped and the apex has two long spines which might present from two to four smaller ones (Herrera-Ulibarri, 2009)

Females have a marsupium in which they carry the embryos until they become juveniles and are released. Marsupium is absent in males, but a penis is found instead. As cannibalism is common among mysids and juveniles are more likely to be preyed, *Leptomysis lingvura* spawn during the evening to minimize predation by visual predators and adults. This phenomenon suggests that photo periods of 24 hours of light will be the most suitable for mysids to release juveniles. Based on this hypothesis, production is expected to be higher in this treatment rather than the photoperiods of 24 hours light and 10 hr light - 14 hr darkness. As mysids contain high content of PUFA (Polyunsaturated fatty acid), mainly EPA (eicosapentaenoic acid), AA (arachidonic acid) and DHA (docosahexaenoic acid), they constitute a valuable natural prey. Those fatty acids play an important role by maintaining cellular membranes structures and functions and also they favor the eicosanoids

formation, a group of hormones highly active (Herrera-Ulibarri, 2009). Indeed, laboratory studies have proved that mysids grown in present higher PUFA values as those found in mysids collected from natural conditions (Herrera-Ulibarri, 2009; Domínguez *et al.*, 2001; Molina *et al.*, 2007; Otero-Ferrer *et al.*, 2010). By increasing mysid production, a new alternative for aquaculture feeding is available. Thus, mysids are proved to be more adequate principally in the first life stages than the current options, such as *Artemia*, in the culture of cephalopods s (Domínguez *et al.*, 2000).

On the other hand, production is not just the factor to take into account in order to maintain a laboratory culture. Mysid survival has to be considered too as breeding, and consequently production, is directly related to the number of females alive. Based on the natural environmental conditions, photoperiod 10:14 is expected to show the best rate of survival. As this photoperiod has greater similitude with the natural one, individuals are believed to achieve better acclimation and suffer less stress which increase the probability of survival. In this experiment, survival was obtained in relation to mortality, by subtracting the number of dead individuals measured daily.

The main aim of this study was to study three different photoperiods and its possible influence on mysids physiology and consequently on larval production in order to improve juveniles' production.

## MATERIAL AND METHODS

Organisms for this study were collected at Risco Verde, East coast of Gran Canaria. Sampling was performed with SCUBA equipment using a 500µm hand- net. Depth of capture was 5-15 m. about 200 mysids were collected and carried to the laboratory. After two days of acclimation, species were identified by binocular lens and stereoscopic microscope and separated into groups. Only *Leptomysis lingeura* organisms were chosen for this experiment.

Culture system was installed at the University of Las Palmas de Gran Canaria (ULPGC). Filtered seawater was recirculated among three plastic tanks of 35 liters which were connected to a mechanic and biologic filter. Seawater passes through it and afterwards reaches a 70 liter tank containing biologically active rock. About one third of water from each tank was replaced daily in order to avoid the accumulation of feeding waste. Ammonium, nitrates and nitrites were measured daily to assure their maintenance at low level. A thermostat kept temperature of seawater constant at 21.3 °C. White light was provided by two lamps of 16 W. Three different photoperiods (FPs) were chosen: 24 hours at dark, 24 hours at light and 10:14 light:-dark.

Mysids were housed in three plastic tanks, subdivided in three sections, and exposed to specific regimes of light (there were three tanks, one per light's regimen described previously). Tanks were isolated from the rest and from the room by black-plastic walls in order to minimize external disturbances. Each tank contained 75 individuals, with 20 females and 5 males per

subdivision. Replicates were placed inside plastic hatcheries with a 500 µm net to avoid the nauplii mix. When released, nauplii were capable of passing through the hatchery- longitudinal small gaps while adults were not. Consequently, nauplii left it and were kept inside the net. This fact tries to reduce cannibalism and keep juvenile away from predation. Furthermore, the net prevents juveniles from dispersion around the tank, making easier the daily counting.

As mysids seem to prefer animal-matter as food (Murano, 1999), organisms were fed twice a day with 100 48-hours artemia nauplii per mysid. Cysts of artemia were kept in filtered seawater during 24 hours with high aeration. After eclosion, artemia nauplii were filtered and enriched with Easy-DHA Selco® (INVE, Belgium) and kept 24 hours more before being used as food.

From October 30th to November 13th 2010, tanks were inspected daily. Breeding was determined by counting the number of juveniles, retained in the net, from each hatchery. Mortality was established by the number of dead individuals each day.

Number of juveniles and mortality data were statistically analyzed in order to determine whether significant differences exist among variables related to FP. Firstly, test for homoscedasticity (Bartlett test of homogeneity of variances) were carried out for the total number of newborns (Bartlett's K-squared=1.5818, df=2, p-value= 0.45) and total number of adults dead (Bartlett's K-squared= 12.3357, df=2, p-value=0.002). Also,

Shapiro-Wilk normality tests were achieved. Data for mortality and breeding was analyzed separately showing no normal distribution, consequently normality may not be accepted because of data are not homoscedastic. Then, both variables were analyzed by a non parametric Kruskal-Wallis test. No significant differences for the total mortality or production are found among the three photoperiods at the end of the experiment.

In addition, Kruskal-Wallis tests were done at the 5<sup>th</sup>, 10<sup>th</sup> and 15<sup>th</sup> day. Differences among replicates for each FP and among FPs were considered. No differences were found at the end of the 5<sup>th</sup> day, but significant differences among replicates of the FP 24-D appeared at day 10<sup>th</sup> (Fig 3), so this FP was not considered for subsequent analysis

## RESULTS

The total number of juveniles released shows no significant differences at the end of the experiment for the three different FPs (Fig. 1).

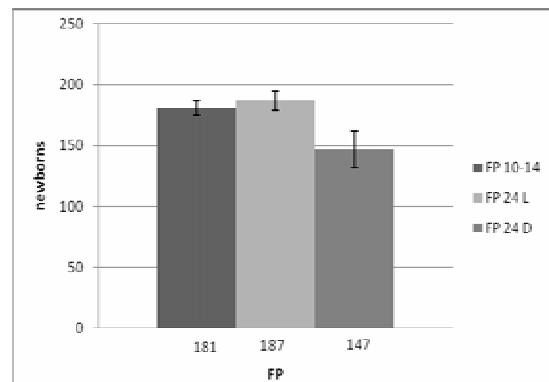


Figure 1. Total number of newborns of each photoperiod. 14:10 (hours dark/hours light), 24-D (24 hours of dark), 24-L (24 hours of light ).

According to the total mortality, no significant differences appear at the end of the experiment (Fig 2).

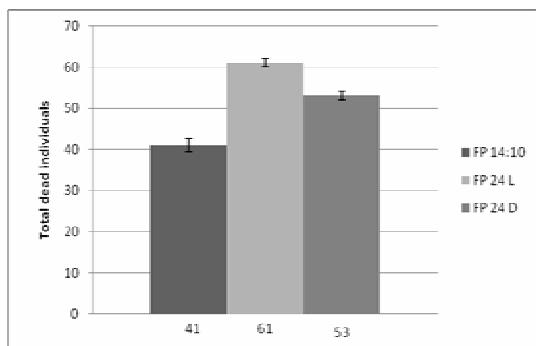


Figure 2: Total number of dead individuals of each photoperiod. 14:10 (hours dark/hours light), 24-D (24 hours of dark), 24-L (24 hours of light).

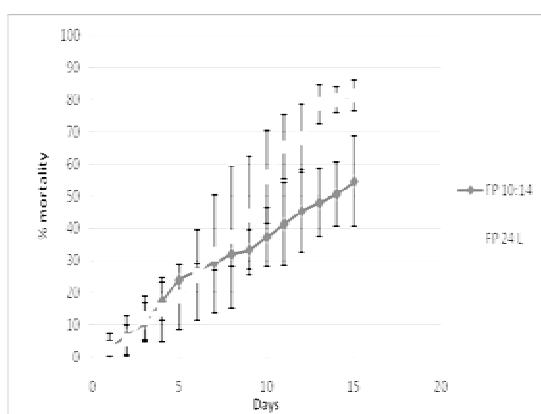


Figure 3: Percentage mortality for 24-L and 10:14 photoperiods.

Mortality was also considered in relation to time. Periods of five days were established in order to examine whether significant differences appear over time. As stated before, the FP 24-D was not considered due to differences among replicates exist. This fact means that disturbances more than FP are affected the replicates and data should not be considered as valid.

## DISCUSSION

Statistical analysis confirmed that variables follow no normal distribution. Furthermore, neither number of juveniles nor mortality show significant differences as a result of the exposition to the three different photoperiods and also the 24 hours of darkness had to be dismissed. Consequently no conclusion is able to establish related to this treatment. Regardless to this photoperiod, significant differences among 24 hr of light and 10:14 appear after the 10<sup>th</sup> day. Thus, the photoperiod 10:14 shows the lowest mortality so it may be accepted as the most suitable for laboratory cultures.

Nevertheless, it is important to point out that this was a fifteen-day experiment with three replicates of 25 individuals, with a total number of 75 individual for each photoperiod. Even accepting that photoperiod may affect in some way the reproduction and/or mortality of *Leptomysis lingvura*, more replicates are needed to affirm it. Furthermore, as Wittman (1981) states marsupial development takes on the average 14.9 days in which the young pass through three main stages (embryonic stage, nauplioid stage and postnauplioid stage). As individuals do not belongs to the same cohort, the sample was not obtained from a controlled culture; individuals might be in a different life stage, so aging is a possible factor affecting mortality. Aged individuals are more likely to present high rate of mortality independently of the photoperiod used.

On the other hand, results for production also show no significant difference. The range for the total number of newborns is quite similar

among photoperiod treatments, consequently no one is assumed to be the best for maximizing productivity. Nor influence of photoperiod on larval development and breeding may be inferred. Nevertheless, as in the case of mortality, the life stage of collected mysids was not initially considered. *Leptomysis lingvura* has been proved to have a frequency of spawning with an average of 7.3 (SD=0.65) days between each release, with an average of 4.8 (SD=2.2) nauplii (Herrera-Ulibarri, 2009). Accepting that and considering that the experiment duration was shorter than the embryonic development, it might have occurred that selected females even those carrying eggs had not enough time of development and no egg-laying took place. Consequently, the potential of photoperiod to induce more breeding could be masked due to the lack of time for eggs to develop. Research involving longer periods of time and cohorts obtained from lab culture should be considered in order to assure the same initial physiological conditions.

On the other hand, cannibalism might represent a factor decreasing breeding (Lussier et al., 1988). Mysid cannibalism appears both among adults and adults and breeding. As females are predated by other mysids (Herrera-Ulibarri, 2009), the chances of spawning are reduced. However, cannibalism is difficult to observe as it requires identification techniques for each individual so, data has not been recorded for this experiment.

In view of these results, photoperiod does not seem to influence directly mysid physiology to induce more larval development

and breeding. Consequently, nor preferred photoperiod pattern may be chosen in order to increase productivity in cultures. In relation to survival, the 10:14 one presented the lowest mortality, as it was expected. However, significant differences for 24 hr of light and 10:14 light/dark patterns appear after the 10<sup>th</sup> day, so this last could be accepted as the most suitable in order to assure the highest survival in culture.

Despite these conclusions, further research and experiments must be done in order to achieve a better understanding on the physiology and biology of this sp.

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