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Research Article

Life Cycle of *Cymadusa filosa* (Amphipod: Gammarids) Under Experimental Laboratory Conditions in the Red Sea-Egypt

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Abstract

Objective: The life cycle of *Cymadusa filosa* under experimental laboratory conditions in the Red Sea-Egypt. **Methodology:** Five of the ovigerous females collected from the coastal zone in front of the National Institute of Oceanography and Fisheries (NIOF) (27°17'09 N, 33°46'07 E) in Hurghada-Red Sea between summer, 2010 and middle of winter, 2011 were of a length that ranged between 8.0-9.0 mm. Each female was separated in a water tank with a capacity of 3 L and was fed different types of micro-algae. The water was changed daily (the rate of water changed was about 1/2 tank) and the follow-up process stages were from the hatching to the juvenile. **Results:** There are two identical periods throughout this experiment. The first period: The developing part after releasing juveniles; the aim of the experiment is to take care of juveniles to reach sexual maturity (ovigerous females) and then hatch directly into juveniles. The second period: The days taken between hatching (G0, 1) to (G0, 2) were 12-15 days, (G0, 2) to (G0, 3) were 7-10 days and (G0, 3) to (G0, 4) were 8-10 days of the ovigerous females. **Conclusion:** The total experimental period of the life cycle of *C. filosa* was (77-83 days). The fecundity of *Cymadusa filosa* was estimated by the number of juveniles released by a female. Females were able to carry eggs at a size of 6.50 mm. During the study, a curve linear represented the relationship between the number of juveniles and the size of ovigerous females *C. filosa*, it was a significant positive correlation.

Key words: *Cymadusa filosa*, adult, juvenile's, field, generation, fertilization

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Amphipod crustaceans of the genus *Gammarus* are common throughout many places in the world and are almost universally characteristics of the surface waters of Northern Eurasia and America. In fresh waters, particularly running waters, they have a central position in food webs, being responsible (along with some cased-caddisfly larvae) for shredding and comminuting all kinds of decomposing leaf litter; hence, they are classified as detritivores in the trophic economy of lowland rivers. Talitrid amphipods are one of the most abundant invertebrates of the upper shore levels of temperate sandy beaches throughout the world[1]. These amphipods are an ecologically important group that plays a key role in the biological processing of macroalgal inputs and facilitates the transfer of nutrients from the ocean to the coastline[2]. According to Orav-Kotta *et al*.[3], gammarid amphipods are very common and important species in the rocky sub littoral of the Northern Baltic Sea. Bakir and Katagan[4] stated that a total of 143 species belonging to amphipoda were found at the depths between the supralittoral zone and 200 m along the Levantine coast of Turkey[5]. In 1981 and 1982, Lyons and Myers made the report on the Red Sea amphipoda by sampling some amphipods specimens from the Gulf of Aqaba coral rubble on the Jordan coastal line. El-Sharouny *et al*.[6] indicated that the Red Sea is characterized by the presence of more than one ecosystem within its coastal areas. These ecosystems include the most famous coral reef and seagrass. When the macroalgae habitats along the Red Sea coast in the vicinity of Hurghada and Safaga were surveyed to illustrate the distribution of macroalgal communities, species composition was recorded (Phaeophyta, Rhodophyta and Chlorophyta). According to Cunha *et al*.[7], low-latitude, warm-water amphipods show iteroparous, multivoltine life history patterns. Multivoltinism and continuous reproduction have been reported in many other species occurring in the tropical Indian Ocean[8]. Steele and Steele[9] described also short brood intervals and multivoltine life cycle. On the other hand, low-latitude species are characterized by semi-annual or annual life histories, small body sizes and high reproductive potentials[10]. Wildish[11] recognized six basic life histories within the Gammaridea: Multivoltine (more than one generation per year) semiannual, univoltine (one generation per year) or multivoltine annual, semelparous (single-brooded) biannual and semelparous or iteroparous (multiple-brooded) perennial. Females of semi annual species or populations grow rapidly, mature early and are very fecund; this set of traits is presumably associated with warm and tropical habitats. Wildish[12] confirmed about talitroids.

The aim of the study is to investigate the biological studies including the life history and reproduction traits of *C. filosa* in terms of: Life span, maturation, spawning and productivity (juveniles released).

MATERIALS AND METHODS

Collection of sample: Five of the ovigerous females were collected from the coastal zone in front of the National Institute of Oceanography and Fisheries (NIOF) (27°17'09 N, 33°46'07 E) in Hurghada the Red Sea, between summer 2010 and middle of winter 2011. They were of a length that ranged between 8.0-9.0 mm. Each female was separated in a water tank with a capacity of 3 L and was fed different types of micro-algae. The water was changed daily (the rate of water changed was about 1/2 tank) and the follow-up process was from the hatching to the juvenile. After one day, juveniles were released from the ovigerous female with about 67 individuals. The 1st generation (G0) of juveniles was separated into another tank in order to assess the time required for the maturation of females and males. The water temperature ranged between 28-30°C and water salinity was 39-40‰.

When males and females of the 1st generation reached their maturity, the time taken was assessed. Then, 5 mature individuals of males and females were separated into 5 separate tanks, after measuring their sizes.

The time taken for the ovigerous female (brood pouch filled with eggs) was recorded.

Both sexes were identified. The males were identified according to the presence of an enlarged second gnathopod, while the female's recognition was based on the presence of brood pouch/lamellae and the start of the formation of eggs[13].

The time period of releasing juveniles from the ovigerous female and the number of juveniles separated in other tanks were recorded, it was called generation (G0, 1). Once again, there was a follow-up fertilization process between the adult male and female in each tank to determine the start time of the formation of eggs and the release of juveniles, respectively; this was called (G0, 2). After that, there were separations of juveniles in other tanks and once again, there was a follow-up fertilization process between the adult male and female to reach (G0, 3) and (G0, 4) in the same way.

The mortality rate in each hatching time from (G0, 1), (G0, 2), (G0, 3) and (G0, 4) was recorded.

The fecundity was estimated by the number of juveniles released by each female.

RESULTS

For the complete understanding of the various stages of the life cycle of *C. filosa*, several parameters had to be determined experimentally. These parameters included, stages of female development, the time between the oviposition and hatching of juveniles (incubation period), the number of hatched juveniles in each generation, the survival rate of the juveniles produced and the fecundity.

Beginning of the experiment: The start of the life cycle of *C. filosa* was the collection of 5 ovigerous females from the field. After one day, these females hatched directly into juveniles that resembled adults. The produced number was about 67 individuals and the size was between 2.0-2.5 mm; these stages are known as generation (G0) as shown in Fig. 1. The juveniles were separated (non-ovigerous) in a small tank and were fed a mixture of microalgae (*Nannochloropsis* spp., *Tetraselmis* spp., *Isochrysis* spp., *Chlorella* spp. and *Cylindrotheca* spp.) in order to follow the increase growth rate until they reach their sexual maturity in about (30-37 days). This allowed them to release the eggs into the brood pouch through the genital pores. The sizes of males and females at that stage were (5.0-5.5 mm). After about 37-44 days with continuous feeding, the individuals reached a size of (5.5-6.0 mm) in length and the brood pouch was complete with eggs and was known as (ovigerous female's stages). After 45-53 days from the start of the experiment, the females' sizes went up to 6.0-6.5 mm in length and released juveniles from the ovigerous females which are known as (G0, 1). The total number of juveniles in this generation was (41) individuals.

Second stage: The incubation period was between (G0, 1) and (G0, 2) for about 12-15 days. Females and males ovigerous were separated after hatching, only for a short time of about 53-60 days. From the start of the experiment, the females sizes went up to 6.5-7.0 mm. Mature females were immediately available for fertilization after a short time; then, they released the eggs into the brood pouch after 60-65 days, retained them until they passed the developmental stages and finally, the mature eggs hatched directly into juveniles that resembled adults. This is known as (G0, 2). The total number of juveniles was (47) individuals.

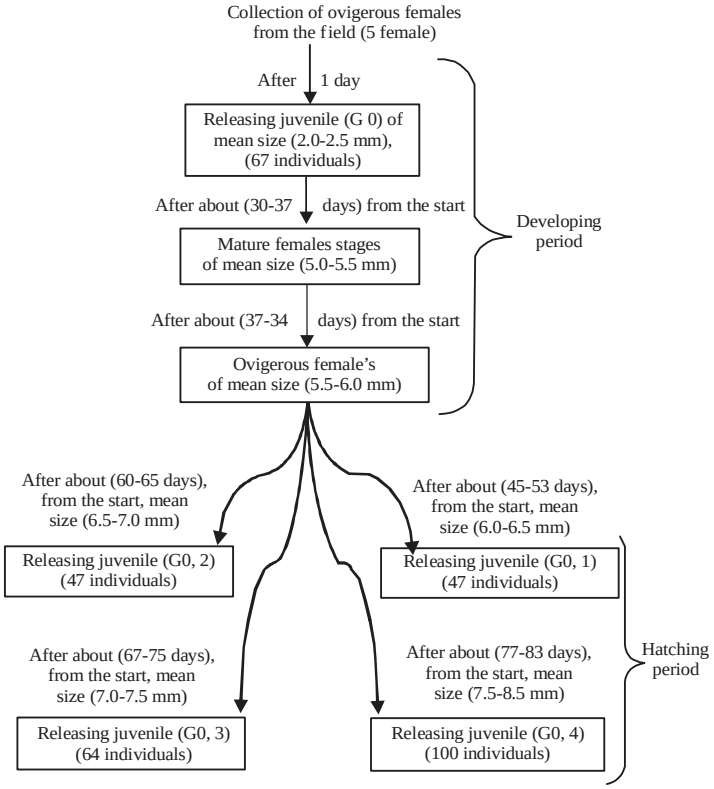

Fig. 1: Experimental life cycle diagram for the sequence of releasing juveniles and the time taken in the stages of *C. filosa* under laboratory conditions

Table 1: Survival rate of juveniles (at different time) of *C. filosa* reared from hatching at 30°C

		Survival (%)			
No. of hatched broods	Total No. of juveniles	Week 1	Week 2	Week 3	Week 4
(G0, 1)	41	100	100	100	86
(G0, 2)	48	100	100	100	100
(G0, 3)	64	100	100	100	100
(G0, 4)	100	100	100	100	100

Third stage: The incubation period was between (G0, 2) and (G0, 3) for about 7-10 days. Ovigerous females and males were separated once more after hatching, for a short time of about 64-69 days. From the start of the experiment, the females sizes went up to 7.0-7.5 mm. The mature females were immediately available for fertilization only after a short time. They then released the eggs into the brood pouch after 67-75 days, retained the eggs until they passed the developmental stages and finally, the mature eggs hatched directly into juveniles that resembled adults. This is known as (G0, 3). The total number of juvenile was (64) individuals.

Fourth stage: The incubation period was between (G0, 3) and (G0, 4) for about 8-10 days. Females and males ovigerous were separated once more after hatching, for a short time of about 70-78 days. From the start of the experiment, females sizes went up to 7.5-8.0 mm. Mature females were immediately available for fertilization only after a short time; then, they released the eggs into the brood pouch after 77-83 days, retained the eggs until they passed the developmental stages and finally, the mature eggs hatched directly into juveniles that resembled adults. This is known as (G0, 4). The total number of juveniles was (100) individuals.

Two identical periods throughout this experiment

First period: The developing part, which is after releasing juveniles. The aim of the experiment is to take care of juveniles to get them to sexual maturity (females ovigerous) and then hatch directly into juveniles. This is a long period of time in a life cycle which was 45-53 days from the start of the experiment.

Second period: The days that were taken between hatching (G0, 1) to (G0, 2) were 12-15 days, (G0, 2) to (G0, 3) were 7-10 days and (G0, 3) to (G0, 4) were 8-10 days of the ovigerous females.

From the previous results: The total experimental period of time for these fixed generations of the life cycle of *C. filosa* was 77-83 days.

Survival rate of the juveniles produced under laboratory conditions during the life cycle: Through the study of the life cycle of *C. filosa*, the survival rate of the total number of juveniles was shown in Table 1. For the determination of the survival rate of juveniles *C. filosa*, four hatched broods, each included a total of 41 juveniles released from a brood (G0, 1) and a total of 48, 64 and 100, respectively, in (G0, 2), (G0, 3) and (G0, 4), were observed throughout their life span under the laboratory conditions. Data of the changes in the survival rate are presented in Table 1. Three hatched broods (G0, 2), (G0, 3) and (G0, 4) released survived with 100% survival rates until week 4. Moreover, only in a single brood (G0, 1), two juveniles died at week 4 (14% mortality), which was the last period of the experiment.

Fecundity of natural and reared females of *Cymadusa filosa* under laboratory conditions during their life cycle: The fecundity of *C. filosa* was estimated by the number of juveniles released by a female. Newly released juveniles were transferred immediately to a new tank. The relationship between the number of broods and the number of eggs/juveniles of the life cycle of *C. filosa* is presented in Fig. 2. Figure 2 indicates that the type of relationship between the number of broods and the number of eggs/juveniles is symbolized by a curve linear and represents a significant positive correlation and is embodied by a curve exponential represented by the equation:

$$Y = 5.6811e^{2.8388x} \text{ with } R = 0.9546$$

Where:
Y = No. of juveniles
X = No. of Brood

The number of broods produced during the life cycle ranged from (G0, 1) to (G0, 4). Females were able to carry eggs at a size of 6.50 mm. During the study, only one set was observed to produce 4 broods. The total number of juveniles produced in a brood (G0, 1) was an average of (8.20±0.84) and in (G0, 2), (G0, 3) and (G0, 4) were (9.60±0.89), (12.80±3.11) and (20±2.92), respectively. The total number of juveniles over the lifespan of a single female varied

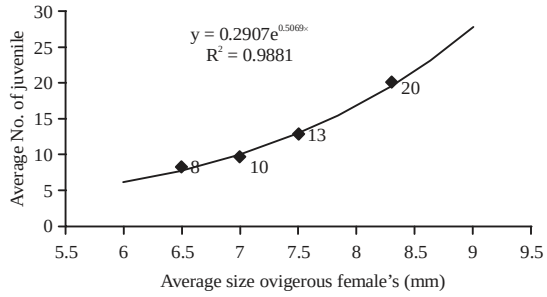

Fig. 2: Relationship between the number of broods and the number of eggs/juveniles of the life cycle of *C. filosa*

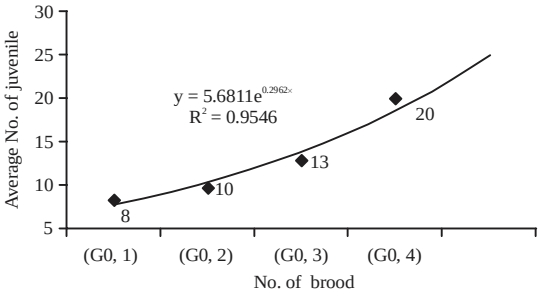

Fig. 3: Relationship between the number of juveniles and size ovigerous females of the life cycle of *C. filosa*

from 46-60 individuals and the total number of juveniles from (G0, 1) to (G0, 4) are 41, 48, 64 and 100, respectively.

The relationship between the number of juveniles and size of ovigerous females *C. filosa* is presented in Fig. 3. Figure 3 indicated the type of the relationship which was a curve linear and represented a significant positive correlation represented by the equation:

$$Y = 0.2907e^{0.5069x} \text{ with } R = 0.9881$$

Where:

Y = Number of juveniles

X = Size ovigerous females (mm)

The average size of ovigerous females at brood (G0, 1) was 6.50 mm. The corresponding average number of juveniles was (8.20±0.84) individuals. In brood (G0, 2), the size reached 7.0 mm for females and the number of juveniles were (9.60±0.89) individuals. The continued growth size of ovigerous females was 7.5 mm in brood (G0, 3). In brood (G0, 4) they were 8.3 mm. There was an increase in the number of juveniles released with an increase of ovigerous females.

DISCUSSION

The present study shows two stages for breeding activity. The first stage consists of the time period of releasing the juveniles to get to ovigerous females and that took about 30-44 days. Then, there was the release of juvenile after 45-53 days, that is called (G0, 1). The second stage shows the short brood intervals between hatching from (G0, 1) to (G0, 2) as 12-15 days and from (G0, 2) to (G0, 3) as 7-10 days and from (G0, 3) to (G0, 4) as 10 days; there are breeding activities and multivoltinism in *C. filosa* under laboratory conditions in the (Red Sea tropical region). According to Cunha *et al.*[7], low-latitude, warm-water amphipods show iteroparous, multivoltine life history patterns. Multivoltinism and continuous reproduction have been reported in many other species found in the tropical Indian Ocean, as stated by Appadoo and Myers[8]. Steele and Steele[9] pointed out the short brood intervals and multivoltine life cycle. On the other hand, low-latitude species are characterized by semi-annual or annual life histories, small body sizes and high reproductive potentials, Sainte-Marie[10]. Wildish[11] recognized six basic life histories within the Gammaridea: Multivoltine (more than one generation per year) semiannual, univoltine (one generation per year), multivoltine annual, semelparous (single-brooded) biannual and semelparous or iteroparous (multiple-brooded) perennial. Females of semiannual species or populations grow rapidly; they mature early and are very fecund. This set of traits is presumably associated with warm and tropical habitats[12].

In this present study, the time period between brood pouch and generations to the release of juveniles of *C. filosa* is about (7-15 days). In *Eriopisa chilkensis* estuarine amphipod, the time between oviposition and the hatching of juveniles, (incubation period), varied from 8-14 days, as reported by Aravind *et al.*[14].

The present study shows that the type of the relationship between the No. of juveniles and the size of the ovigerous females is a curve linear represented by a significant positive correlation. The positive correlation between the number of broods and the number of juveniles is consistent with Appadoo and Myers[8]. A linear correlation between the mean brood size and the female size is a common feature in amphipods; e.g., *Pontocrates arenarius* and *P. altamarinus* by Beare and Moore[15]. Larger females generally carry more eggs than smaller ones because of their greater body length by Sheader[16]. Brood size increases with body size in female amphipods by Steele and Steele[9]. In general, amphipods show a positive correlation between the body length and the number of embryos per brood[10].

This study finds that the lengths of ovigerous females of a length (6.5-8.5 mm) of *C. filosa* carry 7-25 eggs. Incubating females (4-13 mm) of *C. filosa* in the Mediterranean water carry 2-55 eggs; similar trends are observed for *C. filosa* in

Mauritius where females of size range 7.1-14.8 mm carry 8-56 eggs by Gilat[17]. Larger females generally carry more eggs than smaller ones because of their greater body length, Sheader[16]. The number of eggs depends on the size of females with Appadoo and Myers[8].

CONCLUSION

The results of the present study showed that the total experimental period of *Cymadusa filosa's* life cycle was between 77-83 days. Furthermore, females were able to carry eggs at a size of 6.50 mm. Finally, the study showed a positive significant relationship between the number of juveniles and the size of ovigerous females of *C. filosa*.

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