




## Article

# Population biology of *Cymadusa filosa* (Crustacea: Amphipoda) associated with *Sargassum furcatum* (Phaeophyceae) beds in a coastal area of Brazil with petrogenic hydrocarbon pollution

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**ABSTRACT.** This study aimed to assess population and reproductive aspects of the amphitoid amphipod *Cymadusa filosa* Savigny, 1816 in a region with pollution by petrogenic hydrocarbons. Sampling of *Sargassum furcatum* Kützinger, 1843 fronds and the associated fauna was conducted in March, September and December 2007 and February 2008, on four rocky shores located on São Sebastião Island, southeastern Brazil. A total of 1372 specimens of *C. filosa* were recorded, including 934 juveniles, 274 females (60 ovigerous) and 164 males. Population mean density ranged from 1.27 to 6.40 ind/g with higher mean values in March 2007 and lower in December 2007 and February 2008. The total body length of males varied from 4.11 to 20.75 mm and of females from 3.58 to 20.22 mm. No significant difference was detected between male and female body length. The overall sex ratio significantly differed from 1:1 proportion and was skewed toward females (0.60:1). Ovigerous females occurred in higher proportion in February 2008 and presented mean fecundity of  $26.01 \pm 1.84$  eggs/female (mean  $\pm$  se), with great variation in the number of eggs found in the marsupium (between 1 and 56). Fecundity increased significantly with increasing female body length. The *C. filosa* population structure and reproduction were similar to those of populations in other locations, in Brazil and abroad indicating that pollution by petrogenic hydrocarbons in the São Sebastião Channel did not affect the population parameters of *C. filosa* in this region.

**KEYWORDS.** Peracarida, amphipod, Amphithoidae, reproduction, petroleum.

**RESUMO.** **Biologia populacional de *Cymadusa filosa* (Crustacea: Amphipoda) associada a bancos de *Sargassum furcatum* (Phaeophyceae) em uma área costeira do Brasil com poluição por hidrocarbonetos petrogênicos.** Este estudo teve como objetivo avaliar aspectos populacionais e reprodutivos do anfípode amfipitoídeo *Cymadusa filosa* Savigny, 1816 em uma região com poluição por hidrocarbonetos petrogênicos. Amostragens de frondes de *Sargassum furcatum* e da fauna associada foram realizadas em março, setembro e dezembro de 2007 e fevereiro de 2008, em quatro costões rochosos localizados na Ilha de São Sebastião, sudeste do Brasil. Um total de 1372 espécimes de *C. filosa* foi registrado, incluindo 934 juvenis, 274 fêmeas (60 ovígeras) e 164 machos. A densidade populacional média variou de 1,27 a 6,40 ind/g, com valores médios maiores em março de 2007 e menores em dezembro de 2007 e fevereiro de 2008. O comprimento corporal total dos machos variou de 4,11 a 20,75 mm e das fêmeas de 3,58 a 20,22 mm. Nenhuma diferença significativa foi detectada entre o tamanho do corpo de machos e fêmeas. A razão sexual geral diferiu significativamente da proporção de 1:1 e foi desviada para as fêmeas (0,60:1). Fêmeas ovígeras ocorreram em maior proporção em fevereiro de 2008 e apresentaram fecundidade média de  $26,01 \pm 1,84$  ovos/fêmea (média  $\pm$  erro padrão), com grande variação no número de ovos encontrados no marsúpio (entre 1 e 56). A fecundidade aumentou significativamente com o aumento do comprimento corporal das fêmeas. A estrutura populacional e a reprodução de *C. filosa* foram semelhantes às de populações de outras localidades no Brasil e no exterior, indicando que a poluição por hidrocarbonetos petrogênicos no Canal de São Sebastião não afetou os parâmetros populacionais de *C. filosa* nesta região.

**PALAVRAS-CHAVE.** Peracarida, anfípode, Amphithoidae, reprodução, petróleo.

Amphipods are the most representative group of peracarids among the invertebrates that are found associated with different species of macrophytes including macroalgae and seagrasses (MUKAI, 1971; EDGAR, 1983a; NORTON & BENSON, 1983; GUNNILL, 1985) in all the coastal regions of the world. Associated with brown algae of the genus *Sargassum* C. Agardh, 1820, which occur in both temperate and tropical regions, amphipods and molluscs generally

represent the group with the highest species richness and abundance (EDGAR, 1983a; LEITE & TURRA, 2003; JACOBUCCI *et al.*, 2009; LEITE, 2011; LONGO *et al.*, 2019). Amphipods associated with *Sargassum* and other marine macroalgae represent an important link between these producers and higher trophic levels. They are an important food source for many coastal fishes including species of economic importance (NAKAMURA *et al.*, 2003).

In Brazil, especially on the north coast of state of São Paulo in southeast Brazil, there are extensive beds of different species of *Sargassum* (SZÉCHY & PAULA, 2000) where about 30 species of associated amphipods have already been found, with high abundances and different life and feeding habits (WAKABARA *et al.*, 1983; LEITE *et al.*, 2000, 2007; TANAKA & LEITE, 2003; JACOBUCCI *et al.*, 2009).

Among the amphipods associated with *Sargassum*, representatives of the Ampithoidae family are generally represented by larger species that use algae as food and to build their shelters for reproduction (APPADOO & MYERS, 2003; JACOBUCCI & LEITE, 2008; TAVARES *et al.*, 2013; MACHADO *et al.*, 2017). Due to their predominantly herbivorous diet, these organisms can exert a strong influence on the structuring of *Sargassum* bed communities, as they feed on this macroalgae and associated epiphytes (CRUZ-RIVERA & HAY, 2001; JACOBUCCI *et al.*, 2009).

In Brazil, 15 species of amphipods have already been recorded (ANDRADE & SENNA, 2017a; IWASA-ARAI *et al.*, 2021) associated with different species of algae (*Ampithoe ramondi* Audouin, 1816, *A. robustimana* Andrade & Senna, 2017, *A. seticoxae* Serejo & Licínio, 2002, *A. divisura* Shoemaker, 1938, *A. suapensis* Correia *et al.*, 2016, *A. thaix* Siqueira & Iwasa-Arai, 2021, *Peramphithoe conlanae* Souza-Filho *et al.*, 2016, *Sunampithoe pelagica* (Milne Edwards, 1830), *Cymadusa filosa* Savigny, 1816, *C. peartae* Andrade & Senna, 2013, *C. icapui* Andrade & Senna, 2017; *C. trindadensis* Andrade & Senna, 2017; *C. rasae* Andrade & Senna, 2017; *C. ygara* Andrade & Senna, 2017 and *C. tartarugae* Andrade & Senna, 2017). *Cymadusa filosa* is a widely distributed species, occurring in shallow coastal regions of North and South America, northwest and southeast Africa, the Mediterranean Sea, Indian Ocean (Madagascar) and Pacific Ocean (New Caledonia) (ANDRADE & SENNA, 2017a). In Brazil, *C. filosa* was recorded on the coast of the states of Pernambuco to Santa Catarina (ANDRADE & SENNA, 2017b) associated with different macroalgae (JACOBUCCI *et al.* 2009; TAVARES *et al.*, 2013)

Characteristics related to the algal morphology, hydrodynamics and presence of natural enemies, in addition to the occurrence of environmental contaminants, can influence the population and reproductive parameters of amphipods in different ways (BOROWSKY, 1983; LEITE, 1996; FORD *et al.*, 2003). On the north coast of the state of São Paulo, the São Sebastião channel region has an intense flow of ships that transport oil and its derivatives, as this is the largest oil terminal in the country. Historically, the São Sebastião channel has been affected by oil spills (POFFO *et al.*, 2001; ZANARDI-LAMARDO *et al.*, 2013). Apparently, these pollutants are incorporated by different marine organisms that occur in the region, since petrogenic hydrocarbons have already been detected in fronds of *Sargassum* and in amphipods in that region, including amphipods (LOURENÇO *et al.*, 2019).

Laboratory studies that evaluated the effect of petrogenic hydrocarbons on amphipods recorded behavioural changes (SATBHAI *et al.*, 2017) and increased mortality (DRISCOLL *et al.*, 1998), which can potentially affect populations that occur in contaminated areas. Reproductive patterns and life cycles of amphipods are still poorly known and may vary according to different environmental conditions (LEITE, 1996; APPADOO & MYERS, 2004; JACOBUCCI & LEITE, 2006), including contamination by petrogenic hydrocarbons. In this context, this study aimed to make an initial exploratory survey, in order to determine the population and reproductive aspects of *C. filosa* in a region with pollution by petrogenic hydrocarbons and to compare the data obtained to works carried out in other locations of Brazil and abroad.

## MATERIALS AND METHODS

**Study area.** Sampling was conducted along the rocky shoreline of São Sebastião Island, one of the largest coastal islands in Brazil, located in the northern coast of state of São Paulo, southeastern of the country (Fig. 1). The coast in the region has alternating rocky shores and sandy beaches, with many other coastal islands. The climate is subtropical, and water temperatures range from 12°C at the bottom (50 m deep) to 28°C on the surface in summer. The infralittoral rocky shore communities are dominated by algal beds of *Sargassum furcatum* Kützing, 1843 (Phaeophyceae).

In the São Sebastião Channel is located a large international harbour which includes the Dutos and Terminais Centro Sul (DTCS), formerly the Almirante Barroso Petroleum Terminal (TEBAR), responsible for intense traffic of ships transporting petroleum products. The São Sebastião Channel has also an intense movement of touristic and fishing boats (MARQUES *et al.*, 2013). The presence of aliphatic and polycyclic aromatic hydrocarbons (PAHs) was detected in sediments, algae and amphipods collected in the São Sebastião Channel in January, August and November 2008. Total PAHs ranged from 33.4 to 2010 ng g<sup>-1</sup> dry weight of *Sargassum* with the predominance of low molecular weight PAHs, mostly of naphthalene and alkyl-naphthalenes. In amphipods PAHs concentrations ranged from 71.2 to 989 ng g<sup>-1</sup> dry weight of amphipods. The concentrations of PAHs in amphipods obtained from São Sebastião Channel were higher than those reported in studies with amphipods in other regions (SILVA & BÍCEGO, 2010; LOURENÇO *et al.*, 2019).

**Sampling and processing.** Sampling was conducted in March, September and December 2007 and February 2008, on four rocky shores located on São Sebastião Island (permit SISBIO no. 29765). Among the rocky shores sampled, two are located to the northeast and two to the southeast of the Dutos and Terminais Centro Sul (DTCS) (Fig. 1). On these rocky shores, the collections were performed by means of boat

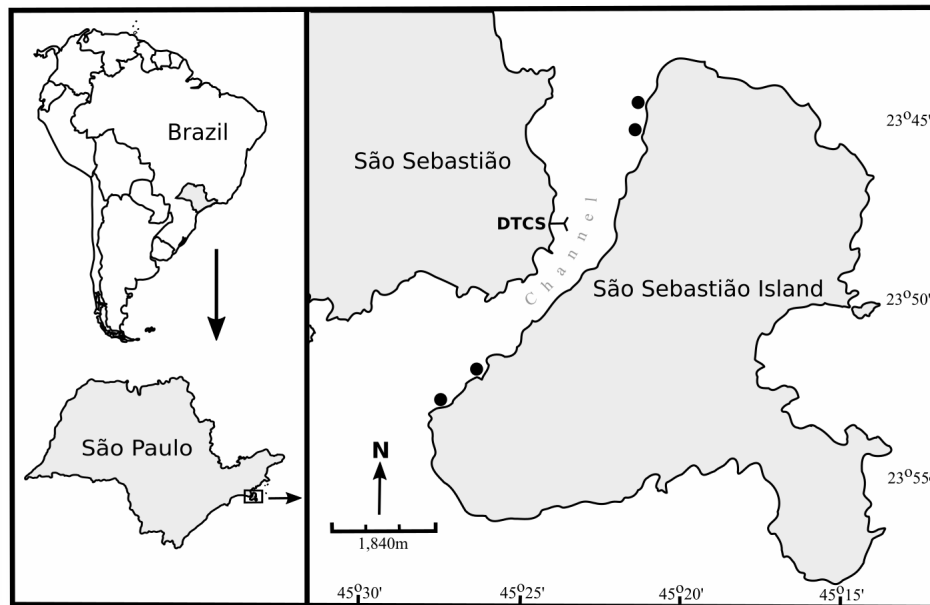


Fig. 1. Location of the sampling stations in the São Sebastião Channel, state of São Paulo, southeastern coast of Brazil (DTCS- Dutos and Terminais Centro Sul, Almirante Barroso Petroleum Terminal).

incursions; material was collected via snorkeling between 1 and 2 m depth. In the *Sargassum furcatum* beds of each rocky shore, a 50 m transect was delimited, in which 10 algal fronds were randomly collected. The fronds were covered with 0.2 mm mesh fabric bags to prevent the organisms from escaping and were transported to the Marine Biology Centre laboratory at the University of São Paulo (CEBIMAR-USP).

In the laboratory, the algae were kept in the freezer at  $-20^{\circ}\text{C}$  until the animals showed no vital signs. After this cooling, the algae were subjected to four successive washes with sea water and the water resulting from this process was filtered through a 0.2 mm mesh to retain the macrofauna, then was subsequently fixed in 70% ethanol. The dry biomass of the *Sargassum* fronds was obtained after drying in an oven at  $60^{\circ}\text{C}$  for 72 hours (JACOBUCCI *et al.*, 2009).

With the aid of a stereoscopic microscope, all individuals of *C. filosa* were separated from the macrofauna, identified, and counted. The sexual differentiation between males and females was performed due to secondary sexual characteristics such as the size of the gnathopods (Figs 2-5) and the presence of oostegites (JACOBUCCI & LEITE, 2006). Females carrying eggs or embryos were considered ovigerous and individuals with no secondary sexual traits were considered juveniles until the minimum size of ovigerous females (JACOBUCCI & LEITE, 2006). The eggs of the females were classified into four stages of development: stage I, eggs with an oval shape without indications of body structures; stage II, eggs with a marked oval shape with the appearance of the caudal cleft and cephalic appendages; stage III, differentiation and appearance of the head, body segments and appendages; stage IV, free juveniles at the marsupium.

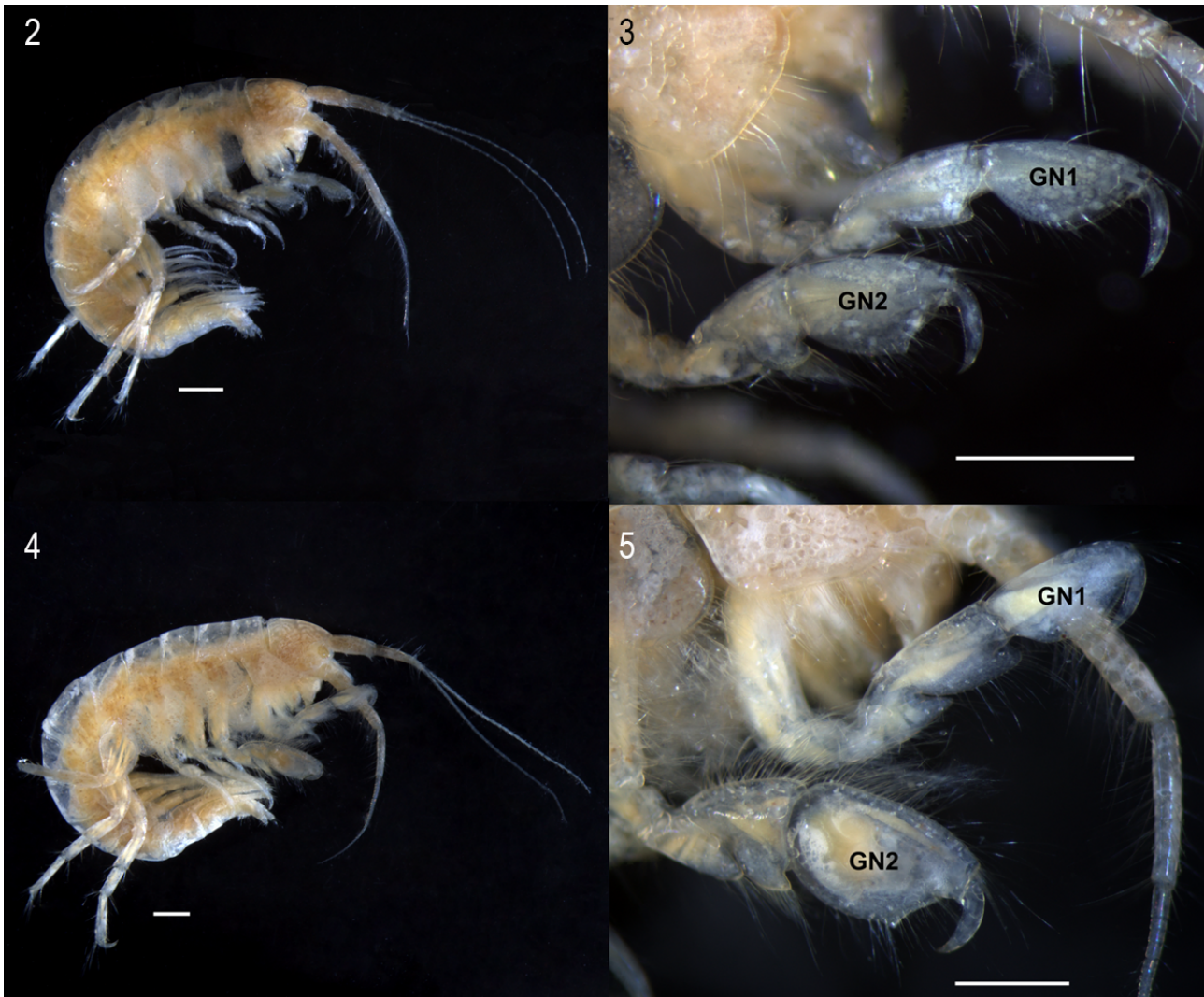
For ovigerous females with intact marsupium, the total number of eggs was counted (JACOBUCCI & LEITE, 2006). The amphipods were photographed with a digital camera (Nikon Coolpix 938 3.1 megapixel) coupled with a Zeiss SV11 stereoscopic microscope. The images were analysed using the UTHSESA Image Tool for Windows (Version 3.0), determining the individuals' total body length from the distance between the insertion of the first pair of antennas and the telson.

**Data analysis.** The individuals of *C. filosa* collected from the four rocky shores were grouped by sampling periods and organized into size classes. Size distributions were analysed adopting the 1.8 mm class interval according to the Sturges formula ( $K = 1 + 3.322(\log N)$ ; where  $K$  = number of classes and  $N$  = total number of individuals analysed) (STURGES, 1926).

The density of associated amphipods in each frond was obtained by dividing the total number of individuals by the dry weight of the frond in grams.

To compare the *Sargassum* biomass between sampling periods one-way ANOVA was used, followed by the a posteriori Newman-Keuls test. The number of eggs of the ovigerous females at different stages of development was also compared using one-way ANOVA, followed by the a posteriori Newman-Keuls test. Amphipod density was compared using Kruskal Wallis followed by the a posteriori Dunn test. The sizes of males and females were compared through the analysis of variance (ANOVA) of two factors (sex and period), where the size of the individuals was considered the response variable. Prior to analysis, the data





Figs 2-5. Sexual differentiation between male and female of *Cymadusa filosa* Savigny, 1816: 2, female; 3, gnathopods 1 and 2 of the female; 4, male; 5, gnathopods 1 and 2 of the male (bars: 1 mm).

were assessed for normality and homogeneity of variance (ZAR, 2010).

The sex ratio was estimated as the quotient between the number of males and the total number of individuals in the population (males plus females) (WILSON & HARDY, 2002). Deviations from a 1:1 sex ratio were tested using  $\chi^2$  test ( $p < 0.05$ ). Sex proportion values higher or lower than 0.5 indicated populations skewed toward males or females, respectively.

The reproductive period of *C. filosa* was assessed by the relative frequency of ovigerous to non-ovigerous females. The minimum, maximum and mean values of fecundity were determined. The fecundity was evaluated through the relationship between the number of eggs, classified in the 1st stage of development, and the size of the females, using simple linear regression (ZAR, 2010).

For all analyses, a significance level of 5% ( $\alpha = 0.05$ ) was considered. Tests for homoscedasticity and normality of the contrasted data sets (after ln-ln transformation) were performed and found to be satisfactory (ZAR, 2010).

## RESULTS

The biomass of *Sargassum furcatum* fronds was significantly lower in September 2007 compared to the other sampling periods ( $F = 15.73$ ,  $df = 3$ ,  $p < 0.001$ ) (Fig. 6). A total of 1372 specimens of *Cymadusa filosa* were recorded, including 934 juveniles, 274 females (60 ovigerous) and 164 males. Population mean density of *C. filosa* ranged from 1.27 to 6.40 ind/g with higher mean values in March 2007 compared to the other sampling periods ( $H = 25.59$ ,  $df = 3$ ,  $p < 0.001$ ) (Fig. 7).

The mean total body length (mean  $\pm$  se) recorded for the sampled population was  $5.64 \pm 0.10$  mm. The length of males varied from 4.11 to 20.75 mm ( $9.87 \pm 0.25$  mm) and of females from 3.58 to 20.22 mm ( $9.09 \pm 0.21$  mm). No significant difference was detected between male and female body length ( $F = 0.68$ ,  $df = 3$ ,  $p = 0.55$ ).

The overall sex ratio significantly differed from a 1:1 proportion and was skewed toward females (0,60 male: 1 females) ( $\chi^2 = 11.78$ ,  $df = 3$ ;  $p < 0.01$ ). The same pattern was observed in each sampling period (Tab. I).

The population was dominated by juveniles in March and September 2007 and February 2008, whereas in December 2007 there was a predominance of adult individuals, more specifically females. Juveniles were distributed in the size

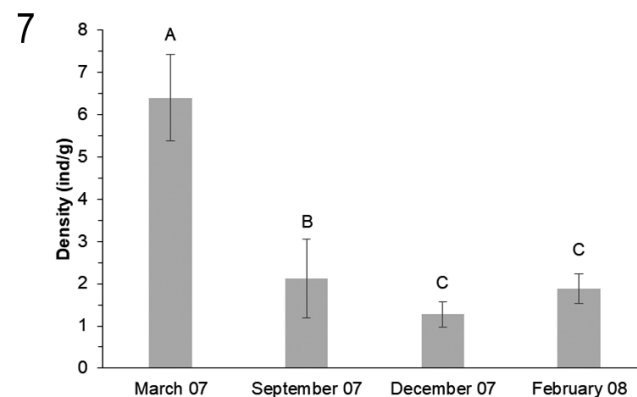
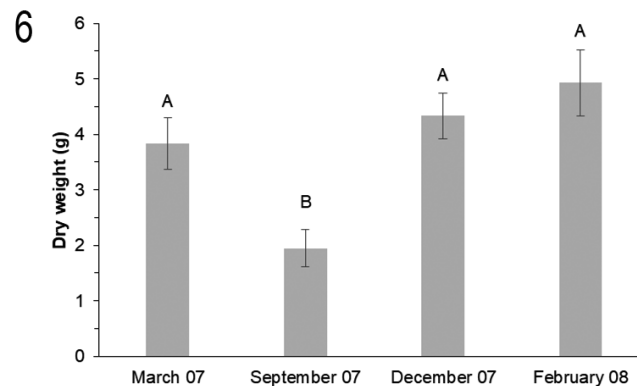
classes 1.32–3.11 to 4.91–6.71 mm. Males and females were recorded in all size classes except the first. Ovigerous females were recorded in intermediate size classes from 4.91–6.71 to 17.52–19.31 mm. (Fig. 8).

Sixty ovigerous females were recorded, with a higher proportion in relation to the total females in February 2008 ( $\chi^2 = 15.32$ ,  $df = 3$ ;  $p < 0.01$ ) (Tab. I; Fig. 9).

Ovigerous females showed great variation in the number of eggs found in the marsupium (between 1 and 56) with  $26.01 \pm 1.84$  eggs/ovigerous female (mean  $\pm$  se). Females with eggs were found in the four stages of development. Significant difference in the number of eggs occurred only between the 1<sup>st</sup> and 4<sup>th</sup> stages of development, with a lower average in the last stage ( $F = 3.84$ ,  $df = 3$ ,  $p < 0.05$ ) (Tab. II).

Tab. I. Mean body length of males and females, sex ratio and number of juveniles and ovigerous females in the population of *Cymadusa filosa* Savigny, 1816 in the São Sebastião channel in the São Sebastião Channel, state of São Paulo, southeastern coast of Brazil.

Month	Male ( $\bar{x} \pm se$ )	N	Female ( $\bar{x} \pm se$ )	N	Sex ratio (M:F)	Number of juveniles	Number of ovigerous females
March 2007	$9.20 \pm 0.28$	66	$8.90 \pm 0.23$	101	0.65:1	544	22
September 2007	$11.87 \pm 1.24$	9	$9.80 \pm 0.71$	21	0.43:1	54	0
December 2007	$9.60 \pm 0.53$	29	$9.09 \pm 0.31$	60	0.48:1	51	8
February 2008	$11.70 \pm 0.44$	60	$10.84 \pm 0.39$	92	0.65:1	285	30
Total		164		274	0.60:1	934	60



Figs 6, 7. Dry weight in grams ( $\pm$ standard error) of *Sargassum furcatum* Kützing, 1843 fronds (Fig. 6) and mean density ( $\pm$ standard error) (number of individuals/gram of algal dry weight) of *Cymadusa filosa* Savigny, 1816 (Fig. 7) in the São Sebastião Channel, state of São Paulo, southeastern coast of Brazil. Bars with the same letter indicate absence of significant difference ( $p > 0.05$ ).

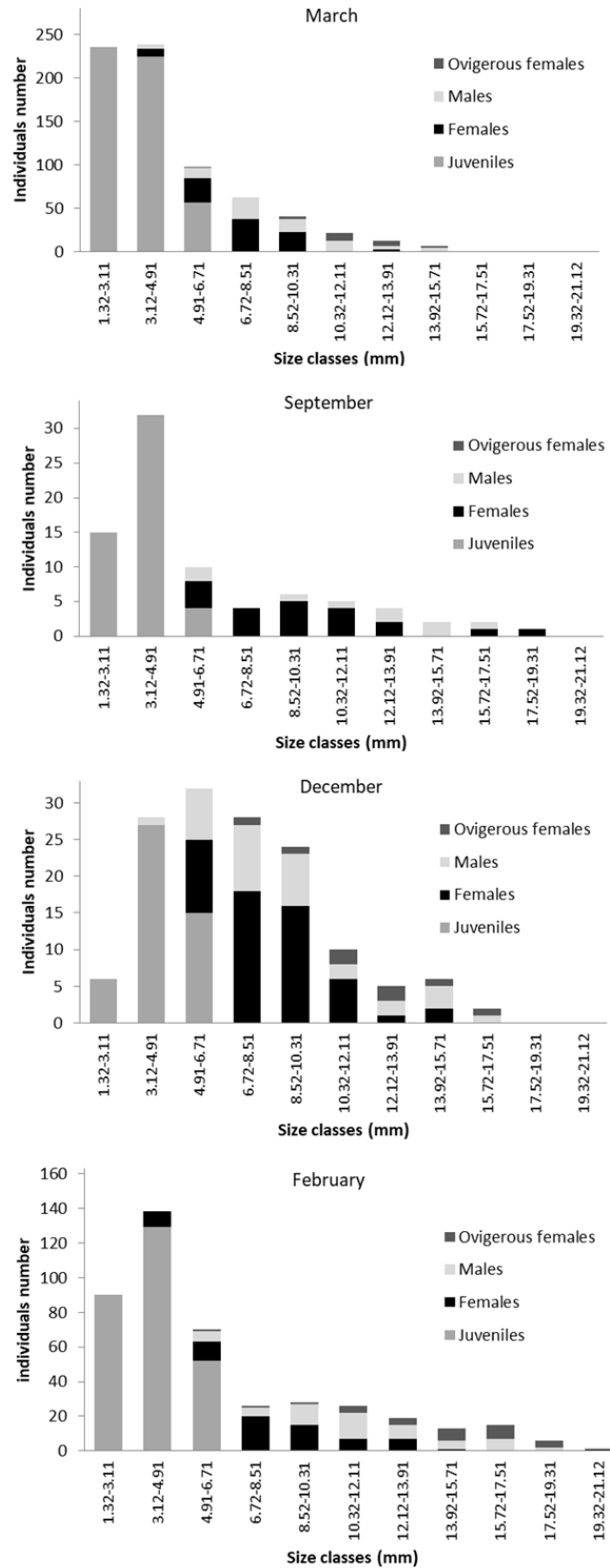


Fig. 8. Histograms of total body length (mm) of males, females and juveniles of *Cymadusa filosa* Savigny, 1816 in the São Sebastião Channel, state of São Paulo, southeastern coast of Brazil.

Fecundity showed that the number of eggs increased with female size ( $y = -28.0812 + 4.1551x$ ,  $R^2 = 0.6458$ ,  $p < 0.001$ ) (Fig. 10).

## DISCUSSION

We compared population data of *Cymadusa filosa* in regions with high organic and heavy metal contamination in Ubatuba (VICENTE *et al.*, 2021) and other sites with reduced contamination (APPADOO & MYERS 2003; JACOBUCCI &

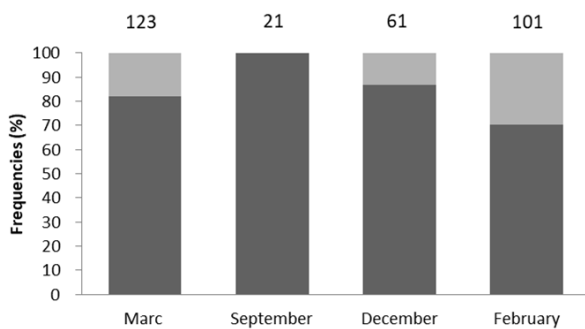


Fig. 9. Relative frequency of ovigerous (light grey) and non-ovigerous (dark grey) females of *Cymadusa filosa* Savigny, 1816 in the São Sebastião Channel, state of São Paulo, southeastern coast of Brazil.

Tab. II. Number of ovigerous females of *Cymadusa filosa* Savigny, 1816 found by egg development stage and average number of eggs in each stage, in the São Sebastião Channel state of São Paulo, southeastern coast of Brazil (\*Same letters indicate non significant differences).

Stage	N	$\pm$ se (*)	Min.	Max.
1 <sup>st</sup>	24	25.00 $\pm$ 3.50 (a)	3	56
2 <sup>nd</sup>	6	16.00 $\pm$ 4.12 (ab)	8	32
3 <sup>rd</sup>	6	14.50 $\pm$ 3.14 (ab)	2	28
4 <sup>th</sup>	25	13.02 $\pm$ 2.05 (b)	1	50

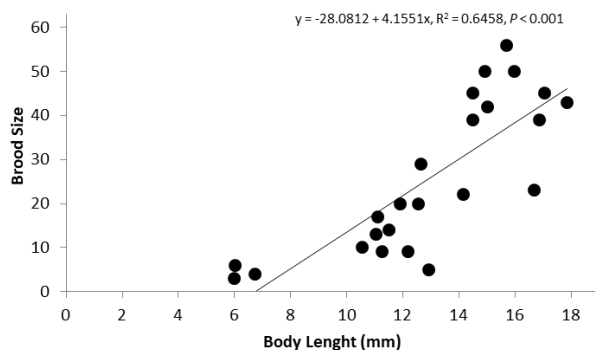


Fig. 10. Regression analyses between ovigerous females' length (mm) and brood size (number of eggs in the marsupium) of *Cymadusa filosa* Savigny, 1816, in the São Sebastião Channel state of São Paulo, southeastern coast of Brazil.

LEITE, 2014) to the present study. A summary of biological characteristics in the different sites are present in Tab. III.

No clear variation of density, adult size, proportion of ovigerous females and number of eggs in brood were verified considering the data available in those studies.

Amphipods in tropical regions are characterized by continuous reproduction and multivoltinism (SAINTE-MARIE, 1991). APPADOO & MYERS (2004) studying the life history of three amphipods in the Indian Ocean, including that of *Cymadusa filosa*, indicated that this species had continuous reproduction and multivoltinism, also observed by JACOBUCCI & LEITE (2006) in Ubatuba in the coast of state of São Paulo.

In the present study, the presence of young *C. filosa* in all months of the study and ovigerous females in three of the four months indicates that this amphipod follows the pattern observed for tropical species. However, a reproductive peak was observed in March 2007 and February 2008, indicated by the high presence of juveniles and ovigerous females. According to SASTRY (1983), reproductive peaks are frequent in different species and locations in tropical regions in response to small environmental variations as observed by APPADOO & MYERS (2004). In other study conducted in the northern coast of the state of São Paulo in a nearby region, density peaks of the amphipods *A. ramondi*, *S. pelagica* and *C. filosa* also occurred in warmer months and coincided with high values of epiphytic algae biomass (JACOBUCCI *et al.*, 2009). The increase in epiphyte biomass favours detritus accumulation in *Sargassum* fronds and tube-building amphipods, like *C. filosa* that can use detritus for tube construction (APPADOO & MYERS, 2003), can be favoured. Epiphytes are also an important food resource for *C. filosa* (JACOBUCCI & LEITE, 2008). These factors could favour the sexual differentiation and female maturation.

A common characteristic among amphipods is the difference in terms of body sizes between males and females, as occurs, for example, in amphipods *Apothyale media* Bousfield & Hendrycks, 2002, *Orchestia gammarellus* Pallas, 1766, *Ptilohyale crassicornis* Haswell, 1879 (LEITE & WAKABARA, 1989; DIAS & SPRUNG, 2003; TSOI & CHU, 2005) with larger males. As most of male energy is allocated for growth, they are generally larger than females. Females, in contrast, use much of their energy for egg growth and production, which slows down their growth rate (CARDOSO & VELOSO, 2001). Although this pattern has already been recorded for *C. filosa* (APPADOO & MYERS, 2004), in the present study there was no significant difference in body length between males and females.

The small proportion of individuals recorded in classes of body size greater than 12 mm, in which higher representation of males would be expected, may explain the absence of difference in size between males and females. As indicated by APPADOO & MYERS (2004) and JACOBUCCI & LEITE (2006), this may result from selective predation by fish, with preference for larger amphipods (EDGAR, 1983b).



Tab. III. Summary of biological characteristics for *Cymadusa filosa* Savigny, 1816 in this study and in other studies elsewhere (APPADOO & MYERS, 2004; JACOBUCCI & LEITE, 2006; VICENTE *et al.*, 2021). “–” = Absence of reference values.

	Present study	VICENTE <i>et al.</i> (2021)	APPADOO & MYERS (2004)	JACOBUCCI & LEITE (2006)
	Petrogenic hydrocarbons	Organic and heavy metal pollutants	No pollutants	No pollutants
Mean density (ind/g)	1.27 - 6.40	0.09 - 0.33		~ 0 - 10
Female body size (mm)	9.09 ± 0.21	11.09 ± 0.59 (highest mean)	10.6 ± 1.5 (largest mean)	–
Male body size (mm)	9.87 ± 0.25	10.92 ± 0.87 (highest mean)	10.1 ± 0.9 (largest mean)	–
Proportion ovigerous/non ovigerous females (%)	21.9	–	32-70	8 - 45
Number of eggs in brood	1 - 56	10 - 41	8-56	15 - 69
Mean egg volume (mm <sup>3</sup> )	–	0.04 ± 0.002 (highest mean)	–	0.034 ± 0.008
Sex ratio (M:F)	0.60:1	2.70:1 - 12.04:1	0.60:1	1.25:1

In fact, the impacts of herbivorous and visually oriented fish are well known on amphipod fauna (NELSON, 1979) and may be a structuring factor in the population in the São Sebastião Channel, because there is a high diversity of fish in this region (LAMAS *et al.*, 2016). Further evidence of size restrictions in natural populations is that individuals of *C. filosa* reared in the laboratory often reach body sizes larger than animals collected in the field (unpubl. data).

The sex ratio deviation observed in the present study in relation to the 1:1 ratio, with higher frequency of females, may be related to factors dependent on the life cycle of the species (HAMILTON, 1967). Females are generally more frequent than males, a result also observed for other amphipods (KEVREKIDIS, 2005; CASTIGLIONI *et al.*, 2016) including *C. filosa* (APPADOO & MYERS, 2004). This pattern could be related with differences in the mortality rate, longevity, growth rate, behaviour and habitat use between the sexes (WENNER, 1972; GABLE & CROOKER, 1977). Variation in mortality can occur due to the higher susceptibility of one of the sexes to predation, due to the reproductive behaviour of the species (differential migration). BOROWSKY (1983) experimentally studying the reproductive behaviour of the amphipods *Jassa falcata* Montagu, 1808, *Ampithoe valida* Smith, 1873 and the tanaid *Tanais cavolini* Milne Edwards, 1829, observed that males are more active than females and often look for new females for mating. LEITE *et al.* (2003) suggested that males of the tanaid *Kalliapseudes schubarti* Mané-Garzon, 1949, in search of new copulation opportunities, could leave their tubes and be the target of predators, which would result

in a larger number of females in the population. This is likely to occur for the *C. filosa* population investigated, as males were seen swimming more frequently than females in laboratory cultures (pers. obs.). However, other factors probably affect sex ratio since JACOBUCCI & LEITE (2014), and VICENTE *et al.* (2021) recorded a higher proportion of males in the populations studied.

The mean number of eggs (26 eggs) produced by *C. filosa* found on the shores of the São Sebastião Channel was similar to that of the population of *C. filosa* (25 eggs) of Flamengo cove in Ubatuba (LEITE, 1996), and higher than the number of eggs, 20.6, produced by this species in Mauritius in the Indian Ocean (APPADOO & MYERS, 2004). However, JACOBUCCI & LEITE (2006) found slightly higher mean fecundity (29 eggs) on the rocky shore of Fortaleza beach, also located in the municipality of Ubatuba. Fecundity of *C. filosa* was positively related to the body size of ovigerous females. This positive relationship was found in populations of *C. filosa* studied by LEITE (1996) and APPADOO & MYERS (2004) but was not recorded by JACOBUCCI & LEITE (2006), possibly due to the small number of ovigerous females collected in this study. This positive relationship between fecundity and female body size is common among malacostracan crustaceans (JENSEN, 1958) and has already been identified in other species of amphipods (BOROWSKY, 1986; FONSECA *et al.*, 2000; JACOBUCCI & LEITE, 2006).

The reduction in the number of eggs in the fourth stage of development may reflect the active removal of unviable eggs from the marsupium, as suggested for the



amphipod *Apherusa jurinei* Milne-Edwards, 1930 (DICK *et al.*, 2002). With advanced stages of egg development, there is an increase in their volumes; the space in the marsupium becomes restricted, making the development of all eggs unfeasible (CARDOSO & VELOSO, 2001).

Therefore, it was expected that striking differences in the number of eggs, individual body length and population density were found but the population structure and reproduction presented by *C. filosa* in the studied region were similar to those of populations in other locations, indicating that pollution by petrogenic hydrocarbons in the São Sebastião Channel did not affect the population and reproductive parameters investigated in this region.

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