

Copepods associated with *Phallusia mamillata* and *Ciona intestinalis* (Tunicata) in the area of Taranto (Ionian Sea, southern Italy)

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Commensal and parasitic copepods (Arthropoda: Crustacea: Copepoda) of two common species of sea squirts, *Phallusia mamillata* and *Ciona intestinalis* have been investigated in the sea of Taranto (southern Italy).

A total of 2590 specimens of copepods were collected (five families, eight genera and 13 species). The sex ratio on 2582 specimens was 1228 males (47.56%) to 1354 females (52.44%).

The species were identified by the hosts in which they were found, their sex ratio host by host, and the specific variability and affinity analysis by means of the Sørensen index and the diversity index by means of the Shannon application.

Some species of copepods have been found for the first time as in *Ciona intestinalis* (*Hermannella rostrata*) and in *Phallusia mamillata* (*Notodelphys canui*, *Doropygus pulex*, *Pachypygus gibber*, *Notopterophorus auritus*, *Lichomolgus albens*, *Hersiliodes latericia*). The association *Notodelphys canui*–*Doropygus pulex*—*Notopterophorus auritu*–*Lichomolgus albens* is noted for the first time in the Mediterranean Sea.

INTRODUCTION

Marine copepods are planktonic species and benthic ones, until depths >3000 m. Parasitic and commensal species are found in association with different taxa such as Porifera, Cnidaria, Annelida, Mollusca, Echinodermata, Tunicata, Pisces, Cetacea as well as other crustaceans. The classic papers on the species associated with invertebrates and vertebrates, were mainly morphological studies with the principal purpose to describe completely each species Monniot (1961); Bocquet & al. (1957, 1961, 1964); Gotto (1957, 1993); Bresciani & Lutzen (1961b); Illg & Dudley (1961, 1965); Gage (1966). Taxonomic, morphological and ecological studies have been made in the last 30 years by Humes & Stock (1973); Hipeau-Jacquotte (1978, 1986); Illg & Dudley (1980); Monniot (1981, 1982); Schmidt (1984); Stock & Weinberg (1985); Costello & Myers (1989). Important revisions on the taxonomy of the commensal and parasitic forms within the invertebrates are made by Gotto (1993). Along the Italian coasts, only a few studies have been carried out Costanzo (1968); Costanzo & Calafiore (1985). No study on this argument has been made before now for the Ionian Sea, so an investigation has been carried out considering the hosts *Ciona intestinalis* and *Phallusia mamillata*.

MATERIALS AND METHODS

Samples of *Ciona intestinalis* and *Phallusia mamillata* were made respectively in the Mar Piccolo Lagoon and the Mar Grande Bay of Taranto, in two stations in which these sea squirts were known to exist (Figure 1).

The collection of the materials was made monthly from May 1998 to April 1999, by means of a conventional

triangular dredge, 60 cm in side, carried out for 5 min at 800 r.p.s. In every case, ten specimens of various sizes were retained for each species with the aim of recording the relationship between the host dimension and degree of infestation. The sea squirts were preserved in diluted formaldehyde 4% with seawater, neutralized with MgCO₃ in excess. In the laboratory every specimen was dissected under a stereoscope, cutting the test from the inhalant syphon as far as the cloaca, the mantle and the branches. The copepods isolated were counted, classified and sexed. 2590 specimens were counted belonging to two suborders, five families, eight genera and 13 species. The classification of copepods was made by means of Gotto's key (1993).

RESULTS AND DISCUSSION

The list of species is reported in Table 1. The average values in size of the *Phallusia mamillata* and *Ciona intestinalis* specimens, making up each specific monthly randomized lot and the respective values of variability are reported in Table 2.

The smallest individuals of *P. mamillata* and *C. intestinalis* were caught in September 1998, while the maximum size was in July 1998 for *P. mamillata* and in March 1999 for *C. intestinalis*. This can be explained by considering that the life cycle of the two sea squirts is a year long so that the specimens caught for and before the summer were of the F1 generation and those caught from September were of the F2 generation.

The scarcity of copepods in smaller sea squirts was recorded in the September specimens for *P. mamillata* and *C. intestinalis*. The progressive number increasing up to the

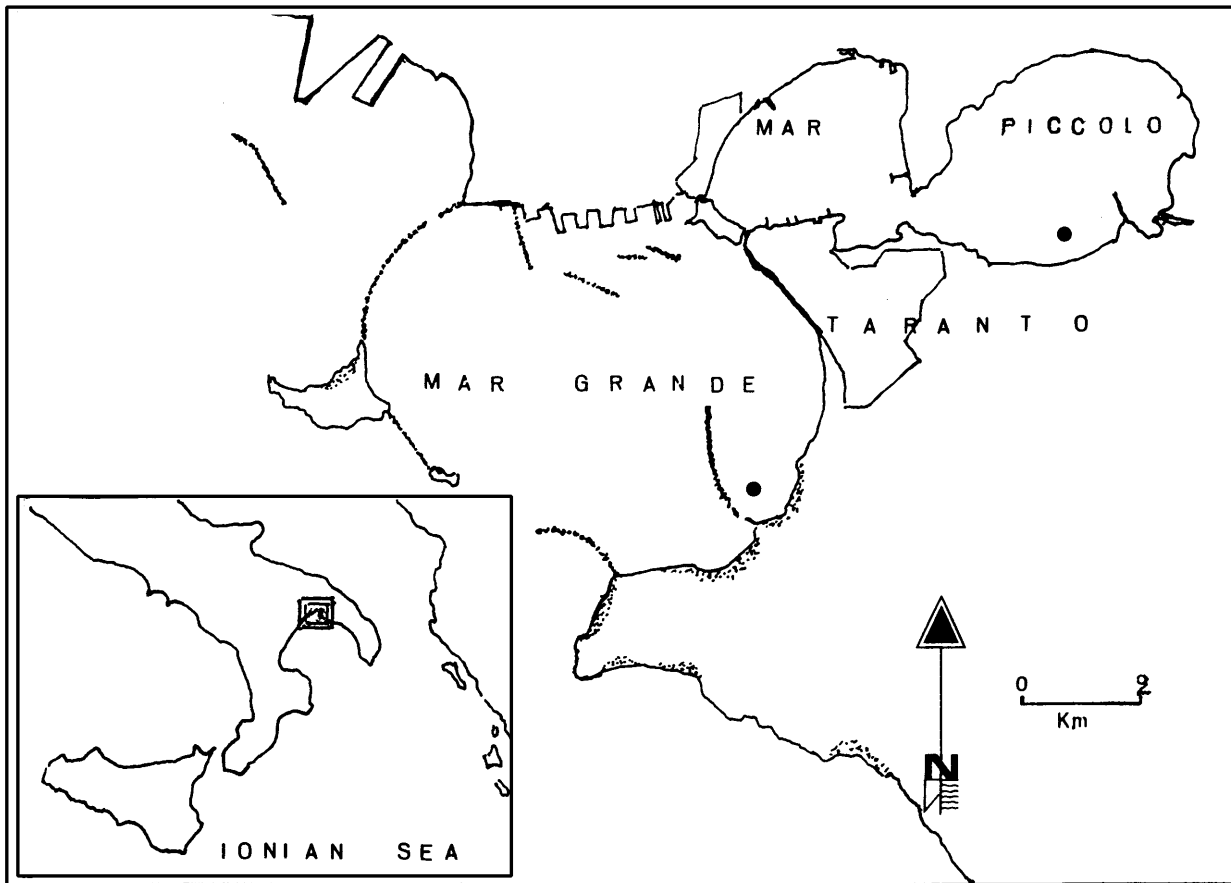


Figure 1. The records stations (black pellets) in the Mar Piccolo and Mar Grande of Taranto (Ionian Sea).

Table 1. List of the species found in the Gulf of Taranto (Ionian Sea).

COPEPODA Edwards, 1840	POECILOSTOMATOIDA Thorell, 1859
CYCLOPOIDA Burmeister, 1834	Sabelliphilidae Gurney, 1927
Notodelphyidae Dana, 1853	<i>Hermannella rostrata</i> Canu, 1891
<i>Notodelphys allmani</i> Thorell, 1859	Lichomolgidae Kossman, 1877
<i>Notodelphys canui</i> Roland, 1962	<i>Lichomolgus canui</i> G.O. Sars, 1917
<i>Doropygus pulex</i> Thorell, 1859	<i>Lichomolgus forficula</i> Thorell, 1859
<i>Pachypygus gibber</i> (Thorell, 1859)	<i>Lichomolgus furcillatus</i> Thorell, 1859
<i>Notopterophorus elongatus</i> Buchholz, 1869	<i>Lichomolgus albens</i> Thorell, 1859
<i>Notopterophorus auritus</i> (Thorell, 1859)	Clausidiidae Embleton, 1901
Ascidicolidae Thorell, 1859	<i>Hersiliodes latericia</i> (Grube, 1869)
<i>Ascidicola rosea</i> Thorell, 1859	

Table 2. Average total length (TL) dimensions and relative variability of the two species of sea squirts during the year of research.

Month	<i>Phallusia mamillata</i>		<i>Ciona intestinalis</i>	
	mTL (cm)	Variancy	mTL (cm)	Variancy
May 1998	7.77	1.25	5.85	1.11
June	8.36	4.66	5.68	0.51
July	8.69	2.39	6.26	2.86
September	3.33	0.24	2.78	0.54
October	5.43	7.52	6.11	2.19
November	6.47	19.33	5.43	3.28
December	6.16	5.69	5.74	4.57
January 1999	7.93	6.06	6.94	2.65
February	7.03	4.58	6.51	4.74
March	7.02	1.87	7.97	7.01
April	6.51	2.41	5.95	1.96

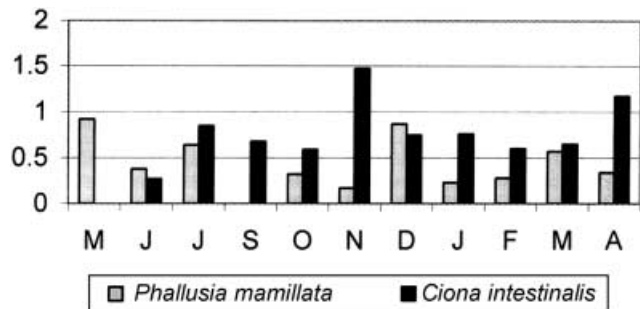


Figure 2. Diversity of *Copepod* species in the two hosts determined using the Shannon index.

Table 3. The number of copepods, T ($^{\circ}\text{C}$) and phytoplankton cell 10^3 l^{-1} values trend referred to *Phallusia mamillata* size increasing, during the investigation.

	M	J	J	S	O	N	D	J	F	M	A
Copepod no.	120	217	379	17	105	430	316	117	190	130	169
T ($^{\circ}\text{C}$)	15.2	23.5	24.6	25.8	17.8	16.2	14.2	15.7	13.5	14.6	14.8
<i>P. mamillata</i> TL	7.77	8.36	8.69	3.33	5.43	4.47	6.16	7.93	7.03	7.02	6.51
Phytoplankton	56	205	102	95	98	83	88	99	72	43	21

Table 4. The number of copepods, T ($^{\circ}\text{C}$) and phytoplankton cell 10^3 l^{-1} values, trend referred to *Ciona intestinalis* size increasing, during the investigation.

	M	J	J	S	O	N	D	J	F	M	A
Copepod no.	31	26	21	7	33	11	88	41	46	28	
T ($^{\circ}\text{C}$)	15.6	18.3	23.5	24.8	20.3	16.2	14.4	12.8	11.2	11	13.7
<i>C. intestinalis</i> TL	5.85	5.68	6.26	2.78	6.1	5.43	5.74	6.94	6.51	7.97	5.95
Phytoplankton	106	274	170	175	80.3	104	83.3	310	84.6	68.7	39.4

Table 5. Copepod species found in *Phallusia mamillata* and *Ciona intestinalis* respectively in the Ionian Sea, in comparison with the literature data referred to the same sea squirts and to the Mediterranean Sea; the species for the Ionian Sea are divided into males (m), females (f) and juveniles (j)

Copepod species	<i>P. mamillata</i>	<i>C. intestinalis</i>	Mediterranean Sea	<i>P. mamillata</i>	<i>C. intestinalis</i>
	Literature	Literature	Literature	New record	New record
<i>Notodelphys allmani</i>	*	*	*	16m, 2f, 6j	
<i>Notodelphys canui</i>					1f
<i>Doropygus pulex</i>		*		42m, 52f	9f
<i>Pachygygus gibber</i>		*	*	8m, 1f	74m, 97f
<i>Notopterophorus elongatus</i>	*		*	56m, 60f	
<i>Notopterophorus auritus</i>		*		4m, 1f	
<i>Ascidicola rosea</i>	*	*	*	3m, 12f	
<i>Hermannella rostrata</i>			*		24m, 40f
<i>Lichomolgus canui</i>		*	*	10m, 12f	57m, 70f, 2j
<i>Lichomolgus forficula</i>	*	*	*	924m, 979f	1m, 1f
<i>Lichomolgus furcillatus</i>		*	*		1f
<i>Lichomolgus albens</i>			*		6m, 11f
<i>Hersiliodes latericia</i>			*	3m	

next spring. Tables 3 & 4 show the relationship between progressive increase in the size of the hosts with the water temperatures, as an indirect influence regarding the higher/lower presence of plankton.

The presence of copepods species, divided into males, females and juveniles for each host, is reported in Table 5, compared with data from the literature with particular reference to the Mediterranean Sea.

The relationship between host size and different number of copepod species per host, reported in Table 6, allows to calculate the correlations. The correlation is significant for *P. mamillata* ($r^2=0.62$; $P<0.05$; $N=11$), but not for *C. intestinalis* ($r^2=0.05$; $P<0.05$; $N=11$). Tables 7 and 8 report the percentage values of the copepod species found each month in *P. mamillata* and *C. intestinalis*, respectively.

As can be seen, *Lichomolgus* genus was the most frequent with four species; *Nothodelphys* and *Notopterophorus* were represented with two species respectively; *Doropygus*, *Pachygygus*, *Ascidicola*, *Hersiliodes* and *Hermannella* with one

Table 6. Correlations between host size (total length, TL) and number of different copepod species per host.

<i>Phallusia mamillata</i> m TL (cm)	Copepod species	<i>Ciona intestinalis</i> m TL (cm)	Copepod species
7.77	4	5.85	1
8.36	6	5.68	2
8.69	6	6.26	3
3.33	1	2.78	2
5.43	4	6.1	4
6.47	2	5.43	5
6.16	5	5.74	3
7.93	2	6.94	3
7.03	2	6.51	3
7.02	3	7.97	2
6.51	4	5.95	4
$r^2=0,6174$; $P<0,05$; $N=11$		$r^2=0,0494$; $P<0,05$; $N=11$	

Table 7. Per cent values referred to the species found in *Phallusia mamillata* during the research.

Months	M	J	J	S	O	N	D	J	F	M	A
Copepods											
<i>Notodelphys allmani</i>	11.7	1.84	1.58								
<i>Notodelphys canui</i>		0.46									
<i>Doropygus pulex</i>		1.38	7.92				18				1.18
<i>Pachypygus gibber</i>								5.98			
<i>Notopterothorus elongatus</i>	15	2.3	4.49		2.86	3.95	4.11		7.89	14.6	5.33
<i>Notopterothorus auritus</i>	4.17										
<i>Ascidicola rosea</i>		0.46	1.58				0.32			3.85	1.18
<i>Lichomolgus canui</i>					1.9		6.33				
<i>Lichomolgus forficula</i>	69.2	92.6	84.2	100	93.3	96.1	71.2	94	92.1	81.5	92.3
<i>Hersiliodes latericia</i>		0.26		1.9							

Table 8. Per cent values referred to the species found in *Ciona intestinalis* during the research.

Months	M	J	J	S	O	N	D	J	F	M	A
Copepods											
<i>Doropygus pulex</i>				57.1							7.35
<i>Pachypygus gibber</i>	100	92.3	52.4		6.06	27.3	50	61	19.6	35.7	17.7
<i>Ascidicola rosea</i>			4.76			36.4	1.14		2.17		
<i>Hermannella rostrata</i>					84.9						52.9
<i>Lichomolgus canui</i>		7.69	42.9	42.9	6.06	9.09	48.9	36.6	78.3	64.3	
<i>Lichomolgus forficula</i>						18.2					
<i>Lichomolgus furcillatus</i>					3.03						
<i>Lichomolgus albens</i>						9.09		2.44			22.1

Table 9. Affinity of the different copepod species in *Phallusia mamillata* and *Ciona intestinalis*.

		<i>Ciona intestinalis</i>										
		M	J	J	S	O	N	D	J	F	M	A
<i>Phallusia mamillata</i>	M	0	0	0	0	0	22	0	0	0	0	0
	J	25	22	40	22	18	50	40	20	40	22	18
	J	0	0	22	25	0	36	22	0	22	0	20
	S	0	0	0	0	0	33	0	0	0	0	0
	O	0	33	29	33	25	44	29	29	29	25	0
	N	0	0	0	0	0	29	0	0	0	0	0
	D	0	29	50	29	22	60	50	25	50	29	22
	J	67	50	40	0	33	29	40	40	40	50	33
	F	0	0	0	0	0	0	0	0	0	0	0
	M	0	0	0	0	0	25	0	0	0	0	0
	A	0	0	29	33	0	44	29	0	29	0	25

species each. For the sex ratio, among 2582 specimens there were 1228 males and 1354 females (47.56 and 55.44% respectively). The sex of six specimens of *Notodelphys allmani* and two specimens of *Lichomolgus canui* could not be identified. In the two hosts the common species were: *Doropygus pulex*, *Pachypygus gibber*, *Ascidicola rosea*, *L. canui*, *L. forficula*. In *Phallusia mamillata*, *L. forficula* is the dominant species and *Pachypygus gibber* an occasional species present only in January 1999. In *C. intestinalis*, *L. forficula* seems to be an occasional species, present only in October 1998, and *P. gibber* is the dominant species. *Notopterothorus elongatus* in *Phallusia mamillata* (absent only in September 1998 and January 1999), and *L. canui* in *C. intestinalis* (absent only in May 1998 and April 1999), are companion species, respectively. All

other species in the two hosts may be considered occasional species.

The affinity values among the copepod species in the two hosts have been calculated using the Sørensen index ($P=2c/(a+b)100$) and are reported in Table 9. The mean is equal to 19.36 and, as can be seen, the highest affinity (40–50%) was found in the winter months (December 1998 and January 1999) with very scarce or no affinity in all the other months. The maximum value (67) was found for copepod species in *P. mamillata* in January 1999 and in *C. intestinalis* in May 1998. The different degree of affinity depends not only on the monthly specific composition of relative copepods in the two hosts but also on the influence of environmental conditions.

The Shannon index ($H_s = -\sum[(n_i/N) \ln(n_i/N)]$) has been used to measure the diversity of the copepod species in each of the two hosts. The results are reported in Figure 2 and Figure 4 shows that the complexity of the guest copepod species in *C. intestinalis* was greater than in *P. mamillata*. The parasitic and commensal copepods found in *P. mamillata* of the Mar Grande and *C. intestinalis* of the Mar Piccolo (Gulf of Taranto) amount to 13 species.

In these sea squirts some copepod species are recorded for the first time: *Notodelphys canui*, *D. pulex*, *Pachypygus gibber*, *Notopterophorus auritus*, *L. canui* and *Hersiliodes latericia* for *Phallusia mamillata*; *Hermannella rostrata* for *C. intestinalis*.

The association among *Notodelphys canui*, *D. pulex*, *Notopterophorus auritus* and *L. albens* is new for the Mediterranean Sea. In respect to the relationship of the copepods species with *P. mamillata* and *C. intestinalis*, it is interesting to note that only five species are common in the two hosts: *D. pulex*, *Pachypygus gibber*, *A. rosea*, *L. canui* and *L. forficula*. Data on the life history or population dynamics of guest copepods in invertebrates are rare although some parasitic or symbiotic forms have been studied in molluscs (Humes & Cressey, 1960; Costanzo & Calafiore, 1987).

Earlier studies on copepods associated with sea squirts show a constant seasonal trend and a strong correlation between life history of the host and life cycle of copepods (Gage 1966). In particular *A. rosea* and *Notodelphys allmani* were found to be abundant in *Ascidella aspersa* at the end of summer and autumn, in relation to the new generation of the sea squirt and the number of *L. canui* associated with *Halocynthia pyriformis* increases in a linear correlation with the host size (Briggs 1976). The present study confirms this aspect. The same seasonal trend among copepod species and hosts is noted; in particular for *Phallusia mamillata* there is a progressive increase in *Notopterophorus elongatus* and *L. forficula* specimens in correlation with the increasing size of the host, from October 1998 to January 1999. In the same manner for *C. intestinalis*, the population of *Pachypygus gibber* and *L. canui* increases from October 1998 to February 1999.

By means of the affinity and diversity analysis on the copepod species group associated with the two sea squirt forms, in this study, in agreement with the literature leave us can affirm that there is no sea squirt species having a homogeneous group of associated copepods.

Another point of interest is the confirmed presence of *L. forficula* in *Phallusia mamillata* observed by Gotto (1993) of which we note that the copepod is the most constant and frequent guest. Finally, *Hersiliodes latericia* and *Hermannella rostrata* have been previously signalled, in different hosts and in particular: *Hersiliodes latericia* associated with the *Leichone clypeata* (Annelida); *Hermannella rostrata* associated with mollusc species of the genera *Cardium*, *Macra*, *Spisula*, *Lutraria*, *Paphia*, *Macoma*, *Scrobicularia*, *Meretrix*, *Solen* and *Ensis* (Bivalvia). The first species has been found in *Phallusia mamillata* and the second also in *Ciona intestinalis*.

Finally, by statistical analyses referred to correlations between host size and number of different copepod species per host, we note that there is a correlation for *P. mamillata* but not for *C. intestinalis*.

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