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Dietary analysis of caprellids *Caprella penantis* and *Caprella grandimana* (Crustacea: Amphipoda) in southern Spain

G. Martínez-Laiz[1] · J. M. Guerra-García[1]

Abstract The trophic ecology of two dominant intertidal caprellids, *Caprella penantis* and *Caprella grandimana*, was studied by examining gut contents. Samples were collected from Tarifa Island (Punta Marroquí, 36°00′00.7″N 5°36′37.5″W) bimonthly from December 2005 to December 2006 to explore seasonal and sex-related differences in diet. The gut contents of the two species included detritus, metazoan prey (crustaceans, polychaetes, hydroids, nematodes, and turbellarians), macroalgae, microalgae, and dinoflagellates. The dominant component was detritus, followed by crustaceans, mainly copepods, and macroalgal tissues. A three-way analysis of variance was conducted using the factors "species" (*C. penantis* vs. *C. grandimana*), "sex" (males vs. females), and "month" (seven dates, December 2005–December 2006). Males of both species had a significantly higher proportion of prey and a lower proportion of detritus in their guts than females. The larger body and gnathopod 2 of males appeared to be related to a more carnivorous diet. Both species fed more on crustaceans and macroalgae during the summer (April, June, and August), probably owing to fluctuations in prey availability and seasonal variations in algal abundance. Moreover, *C. penantis* was significantly more predatory than *C. grandimana*, but both species had a diet based mainly on detritus and crustaceans. *Caprella penantis* dwells at the lower intertidal and *C. grandimana* at the mid-tide level, which may reduce competition between the species.

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Introduction

Amphipods are among the most diverse benthic fauna (Dauby et al. 2001; Lourido et al. 2008), constituting a dominant group in soft bottom and marine rocky habitats (de-la-Ossa-Carretero et al. 2010). They represent the most diverse group of crustaceans with respect to lifestyle, habitat, and size (De Broyer and Jazdzewski 1996). The trophic diversity of amphipods at the interspecific level indicates that they have species-specific functional roles as mediators in trophic pathways (Jeong et al. 2012); hence, they play a key role in energy flow through food webs (Vazquez-Luis et al. 2013). Despite the importance of amphipods in marine ecosystems, little is known about the feeding habits of these crustaceans; indeed, functional types have been studied in fewer than 10 % of amphipod species (Guerra-García et al. 2014).

Caprellids, also called skeleton shrimps, are small marine amphipods common to many littoral habitats that inhabit algae, hydroids, ascidians, anthozoans, bryozoans, sponges, and seagrasses. They cling to artificial (buoys, ropes, and litter) and natural (macroalgae) floating material, which facilitates their dispersal. Caprellids are also useful bioindicators of marine pollution and environmental stress (Guerra-García and García-Gómez 2001; Guerra-García and Koonjul 2005), as they have the capacity to accumulate trace metals (Guerra-García et al. 2009) and are extremely sensitive to tributyltin (Takeuchi et al. 2004; Aono and Takeuchi 2008). These amphipods form an important trophic link between primary producers and higher trophic levels (Woods 2009) by feeding on

a wide variety of organisms, such as copepods, gammarids, hydroids, sponges, polychaetes, macroalgae, and diatoms (see Guerra-García and Tierno de Figueroa 2009; Alarcón-Ortega et al. 2012; Baeza-Rojano et al. 2014). Moreover, it has been concluded that these amphipods are detritivores (see Guerra-García et al. 2014), but it remains controversial whether they should be considered omnivores. Some recent studies have contributed to knowledge of Caprellidae from the Iberian Peninsula and nearby areas, particularly the Strait of Gibraltar, where there is a very high proportion of endemic caprellid species, contributing 30.8 % to the endemism of the Mediterranean Sea (Guerra-García et al. 2002). Although some research has considered the feeding habits of peracarids in the intertidal zone of the Strait of Gibraltar (Torrecilla-Roca and Guerra-García 2012), seasonal dietary variations of caprellids and intraspecific differences have not been explored.

We chose *Caprella penantis* and *Caprella grandimana* to conduct a diet analysis because they are the most common species in the calcareous macroalgae *Corallina elongata*, which is dominant in the shallow waters of the Mediterranean (Baeza-Rojano et al. 2011). Previous studies in this area reported that *C. penantis* and *C. grandimana* are the dominant taxa in the intertidal rocky ecosystems of the Strait of Gibraltar and are associated with *Gelidium corneum* and Corallinaceae algae (*C. elongata* and *Jania rubens*, respectively; see Guerra-García et al. 2011). *Caprella penantis* requires unperturbed areas subjected to exposure and strong currents that reduce sedimentation and suspended organic matter and solids (Guerra-García and García-Gómez 2001). *Caprella penantis* has also been proposed as a suitable bioindicator, as it accumulates trace metals at higher concentrations than other amphipods (Guerra-García et al. 2009). The taxonomy of *C. penantis* is considered one of the most problematic of caprellids worldwide, as it shows wide intraspecific morphological variations due to multihabitat preferences. Cabezas et al. (2013) confirmed cryptic speciation and at least four distinct monophyletic mitochondrial lineages. Only individuals from Tarifa Island were sampled in the present study, which all belonged to the same species of *C. penantis* sensu stricto. *Caprella grandimana* is also common throughout the Mediterranean Sea (Krapp-Schickel 1993) and along the Atlantic African coast (Bellan-Santini and Ruffo 1998). This species is mainly associated with the algae *C. elongata* and *J. rubens* in the intertidal zones of the Strait of Gibraltar, and it reproduces year around. This species grows and reproduces in captivity on a diet of diatoms alone (Baeza-Rojano et al. 2011), and it is a potential aquaculture resource because of its higher omega-3 fatty acid and polar lipid contents compared with gammarids (Baeza-Rojano et al. 2014).

Amphipod feeding preferences have been traditionally assessed using in situ and laboratory observations, feeding experiments, gut content analyses, and studies of the functional morphology of feeding appendages (Legeżyńska et al. 2012). Knowledge of amphipod feeding ecology has recently expanded owing to the use of biomarkers, such as fatty acid biochemical profiles (Jaschinski et al. 2011; McLeod et al. 2013; Baeza-Rojano et al. 2014) and stable nitrogen (δ^{15}N) and carbon (δ^{13}C) isotopes (Olabarria et al. 2009; Mancinelli 2012; Jeong et al. 2012). However, these techniques are indirect indicators, requiring fresh material and preventing the use of specimens fixed in ethanol or formalin. They also depend on an abundance of available material (particularly in small species) to have sufficient quantities for chemical analyses. In contrast, a gut content examination is a direct indicator, which is an advantage. Traditional studies that directly examine digestive contents are limited in number because they are time-consuming (Guerra-García et al. 2014); therefore, we used the methodology proposed by Bello and Cabrera (1999) to study gut contents. This method makes the specimens transparent so gut contents can be observed.

The objective of this study was to gain insight into the feeding ecology of Caprellidae in the rocky intertidal areas of a relatively pristine ecosystem (Tarifa Island). The fact that the two caprellid species dwell at different heights in the intertidal associated with different macroalgal species could affect their role in the trophic chain of this ecosystem. A series of anatomic features, such as the gnathopod 2 and swimming setae on antenna 2, have been used previously to define the feeding habits of different caprellid species. Moreover, it has been demonstrated that larger crustaceans of the same species with stronger feeding appendages tend to consume more prey (Kapiris et al. 2010); therefore, sex-related differences in carnivorous behavior are expected as a reflection of the sexual dimorphism in *C. penantis* and *C. grandimana*. Finally, seasonal variations are a major feature in the dynamics of the rocky intertidal community and have been investigated in other amphipods, such as mesograzer species in an eelgrass community (Jaschinski et al. 2011) or Talitridae (Olabarria et al. 2009). Some caprellid species are known to be opportunists that exploit different organic matter sources throughout the year. The feeding plasticity of the species in our study was assessed as we examined seasonal shifts in their diet.

Materials and methods

Study area and sampling

This study was conducted in the Strait of Gibraltar, an important biogeographic zone, in which fauna of the Mediterranean Sea and the Atlantic Ocean along one axis and fauna of Europe and Africa along the other overlap. The

Spanish side of the Strait is protected under the marine terrestrial Strait Natural Park (Parque Natural del Estrecho), which was declared a protected area in 2003. Tarifa Island is a marine reserve inside the park with a unique biogeographic position in which long-term military access restrictions have contributed to maintain the richest rocky shore intertidal ecosystems in southern Spain (Guerra-García and García-Gómez 2000). This study was conducted at the most southern point of Tarifa Island (Punta Marroquí, 36°00′00.7″N 5°36′37.5″W). The tidal range at this location is approximately 250 cm (Guerra-García et al. 2011). We considered five levels to establish the zonation of these two species (level 1: from zero tidal level to 0.5 m; level 2: 0.5–1 m; level 3: 1–1.5 m; level 4: 1.5–2 m; and level 5: 2–2.5 m). Previous studies on vertical distribution of peracarid fauna in this area revealed that caprellids are the dominant taxa in levels 1 and 3 (*C. penantis* and *C. grandimana,* respectively; Guerra-García et al. 2011). Three replicates (20 × 20-cm quadrats) were sampled every 2 months (from December 2005 to December 2006). The surface was scraped at each level, and all macroalgae and associated fauna were collected.

All samples were fixed immediately after collection in 80 % ethanol to avoid excretion of fecal pellets. The samples were washed over a 0.5-mm mesh sieve in the laboratory, and the amphipods were separated from the algae. The caprellids from the three sampling replicates were sexed using a stereomicroscope and analyzed following the methods of Bello and Cabrera (1999) with slight variations. Individuals with an empty gut were discarded. We examined the digestive contents of 560 specimens (280 *C. penantis* and 280 *C. grandimana* or 20 specimens of each sex/month). Specimens of each sex and species were placed in vials containing Hertwig's liquid (270 g chloral hydrate, 19 ml 1 N HCl, 150 ml distilled water, and 60 ml glycerin) and heated in an oven at 65 °C for 2–5 h depending on cuticle thickness of the specimens. This treatment dissolved the fat in the tissue so the specimens became transparent and their gut contents could be observed. Then, the specimens were mounted on slides for study under the microscope. The percentage of absolute gut content (%Abs) (at 40× or 100×), as the total area occupied in the entire digestive tract, and relative gut content (at 100× or 400×), as the area occupied by each component within all gut contents, were estimated using a microscope equipped with an ocular micrometer.

Data analysis

Mean and SE were calculated to determine the percentages of absolute gut contents and each component of the diet throughout the year. Univariate and multivariate analyses were used to identify potential differences among species and sexes, as well as seasonal fluctuations in the diet, based on the null hypothesis of no change in the diet.

We considered the percentages of detritus and prey to test whether the diet was similar for the sexes of *C. penantis* and *C. grandimana* throughout the year. To determine the quantity of prey in the diet, we pooled the percentages of crustaceans, polychaetes, turbellarians, nematodes, and hydroids. We used analysis of variance (ANOVA) with the following factors: "species," a fixed factor with two levels, *C. penantis* and *C. grandimana*; "sex," a fixed factor with two levels, males and females; and "month," a fixed factor with seven levels, December 2005, February 2006, April 2006, June 2006, August 2006, October 2006, and December 2006. All factors were orthogonal. Twenty replicates were used. Prior to the ANOVA, heterogeneity of variance was tested with Cochran's C test. Variances remained heterogeneous, even when the data were transformed. Therefore, untransformed data were analyzed, as ANOVA is a robust statistical test and is relatively unaffected by heterogeneity of variances, particularly in balanced experiments with a relatively large number of replicates (Underwood 1997). However, to reduce type I error, the level of significance was reduced to <0.01. When ANOVA indicated a significant difference, the source of the difference was identified using the Student–Newman–Keuls (SNK) test. Univariate analyses were conducted with GMAV5 (Underwood et al. 2002).

Nonparametric multidimensional scaling (MDS) was used as the ordination method to explore differences in the caprellid diets (Clarke and Warwick 1994). Data were square root transformed, and the similarity matrix was calculated using the Bray–Curtis index.

Results

We examined the digestive contents of 560 specimens (280 *C. penantis* and 280 *C. grandimana*). The average area occupied by the contents of the entire digestive tract ranged from 42 to 84 % in *C. penantis* and from 25 to 73 % in *C. grandimana* (Tables 1, 2). Gut contents of the caprellid species included detritus, metazoan prey (crustaceans, polychaetes, hydroids, nematodes, and turbellarians), macroalgae, microalgae, and dinoflagellates. The dominant component was detritus, which represented 87.6 ± 1.6 % (*n* = 280) [mean ± SE, *n*] of the *C. penantis* diet and 90.1 ± 1.2 % (*n* = 280) of the *C. grandimana* diet, followed by crustaceans, mainly copepods, and macroalgae. Crustaceans represented 10.4 ± 1.6 % (*n* = 280) in *C. penantis* and 6.27 ± 1.37 % (*n* = 280) in *C. grandimana*. Macroalgal tissues represented 1.23 ± 0.43 % (*n* = 280) in *C. penantis* and 2.42 ± 0.65 % (*n* = 280) in *C. grandimana*. Diatoms and polychaetes were present occasionally.

Table 1 Gut contents of *Caprella penantis* ($n = 280$) studied throughout the year (December 2005–December 2006)

	%Abs	Components (100 %)							
		%Det	%Cru	%Pol	%Hyd	%Tur	%Mac	%Mic	%Din
Dec. 2005									
Males	67.9 (4.4)	89.4 (3.5)	9.7 (3.2)	–	–	–	0.6 (0.5)		0.3 (0.3)
Females	60.5 (9.3)	99.5 (0.5)	0.5 (0.5)	–	–	–	–	–	–
Feb. 2006									
Males	53.5 (8.8)	89.8 (5.2)	10.0 (5.2)	–	–	–	–	–	0.2 (0.1)
Females	42.7 (6.3)	96.8 (2.1)	0.7 (0.7)	2.0 (2.0)	–	–	–	0.5 (0.4)	–
Apr. 2006									
Males	72.0 (5.0)	80.5 (4.3)	16.4 (4.2)	0.3 (0.3)	–	0.3 (0.3)	1.0 (0.6)	1.5 (1.5)	–
Females	76.0 (4.1)	88.4 (2.8)	7.2 (1.8)	–	–	–	3.4 (2.5)	1.0 (0.6)	–
Jun. 2006									
Males	74.2 (2.8)	81.8 (3.9)	16.4 (3.7)	–	0.3 (0.3)	–	0.9 (0.5)	0.6 (0.3)	0.1 (0.1)
Females	70.0 (4.0)	93.7 (1.9)	4.6 (1.7)	–	–	–	1.2 (0.5)	0.4 (0.4)	0.1 (0.1)
Aug. 2006									
Males	39.2 (5.0)	78.4 (3.8)	20.8 (3.8)	–	–	–	0.5 (0.3)	0.3 (0.3)	–
Females	42.0 (3.3)	87.0 (3.3)	11.0 (3.5)	–	–	0.5 (0.5)	1.2 (0.7)	0.3 (0.3)	–
Oct. 2006									
Males	66.2 (4.0)	89.8 (1.8)	8.7 (1.9)	–	–	–	1.0 (0.6)	0.5 (0.3)	–
Females	52.4 (4.7)	85.1 (2.7)	8.5 (2.2)	–	–	–	5.7 (1.7)	0.7 (0.5)	–
Dec. 2006									
Males	78.2 (7.0)	82.3 (3.4)	17.1 (3.4)	–	–	–	0.5 (0.5)	0.1 (0.1)	–
Females	84.4 (5.9)	84.3 (2.2)	13.9 (2.3)	0.3 (0.3)	–	–	–	1.5 (0.9)	–

Mean values with SE (in parentheses) are shown

% Abs total area occupied by all digestive tract contents, *Det* detritus, *Cru* crustaceans, *Pol* polychaetes, *Hyd* hydroids, *Tur* turbellarians, *Nem* nematodes, *Mac* macroalgae, *Mic* microalgae, *Din* dinoflagellates

Dinoflagellates, together with the remaining prey, such as hydroids, turbellarians, and nematodes, were scarcely represented, with percentages of 0.03 % (hydroids) to 0.07 % (turbellarians) in *C. penantis* and 0.01 % (hydroids and dinoflagellates) to 0.04 % (turbellarians) in *C. grandimana*. Polychaetes were detected by the presence of their setae, and hydroids were detected by the presence of their cnidocytes. No nematodes were found in *C. penantis* (Tables 1, 2). Therefore, these two caprellids species fed mainly on detritus, but they complete their diet with a number of different prey and vegetal tissues.

The percentages of prey in the diets of *C. penantis* and *C. grandimana* differed (significant effect of species, $F = 20.77$, $P < 0.001$), and they were consistent over time and between sexes (no significant species × month interaction, $F = 1.56$, $P > 0.01$; or species × sex interaction, $F = 0.79$, $P > 0.01$) (Table 3). *Caprella penantis* had a higher percentage of prey (10.7 ± 0.8 %, $n = 280$) than that of *C. grandimana* (6.63 ± 0.64 %, $n = 280$). The pattern for the percentage of detritus was the opposite: *C. penantis* had a significantly lower percentage of

detritus (87.6 ± 1.6 %, $n = 280$) than that of *C. grandimana* (90.6 ± 1.2 %, $n = 280$) (significant effect of species, $F = 8.54$, $P < 0.01$), and these differences were consistent over time and between sexes (no significant species × month interaction, $F = 2.03$, $P > 0.01$; or species × sex interaction, $F = 0.01$, $P > 0.01$) (Table 4).

Values for the same variables also differed throughout the year (significant effect of month, $F = 6.26$, $P < 0.001$ for prey and $F = 6.66$, $P < 0.001$ for detritus), with a similar pattern for both species that was consistent in both sexes (no significant month × sex interaction, $F = 2.24$, $P > 0.01$ for prey and $F = 2.76$, $P > 0.01$ for detritus) (Tables 3, 4). Lower percentages of prey and macroalgal tissues tended to be found in the diet during cold months and a higher percentage during warmer months. October showed an intermediate value (Fig. 1). December 2005 and February 2006 showed significantly lower prey values (5.13 ± 1.13 %, $n = 80$ and 5.04 ± 0.95 %, $n = 80$, respectively) than April (9.69 ± 1.51 %, $n = 80$) (SNK, $P < 0.05$), June (9.31 ± 1.38 %, $n = 80$) (SNK, $P < 0.05$ but not significant for February 2006), and August (12.9 ± 1.7 %, $n = 80$)

Table 2 Gut contents of *Caprella grandimana* (*n* = 280) studied throughout the year (December 2005–December 2006)

	%Abs	Components (100 %)								
		%Det	%Cru	%Pol	%Hyd	%Tur	%Nem	%Mac	%Mic	%Din
Dec. 2005										
Males	70.3 (4.1)	88.9 (2.7)	7.9 (2.4)	2.1 (1.4)	–	–	–	–	1.1 (0.6)	–
Females	60.0 (6.2)	95.0 (2.3)	0.3 (0.3)	–	–	–	–	4.2 (2.2)	0.5 (0.3)	–
Feb. 2006										
Males	60.0 (6.4)	92.9 (1.8)	5.8 (1.8)	0.5 (0.5)	–	0.3 (0.3)	–	0.3 (0.2)	0.2 (0.1)	–
Females	56.3 (5.8)	96.5 (1.0)	1.0 (0.8)	0.2 (0.2)	–	–	–	0.9 (0.4)	1.3 (0.5)	0.1 (0.1)
Apr. 2006										
Males	27.0 (3.6)	85.2 (3.0)	12.8 (3.0)	–	–	–	–	1.7 (0.6)	0.3 (0.2)	–
Females	24.7 (4.7)	90.4 (3.3)	1.7 (1.0)	–	–	–	–	7.8 (3.4)	0.1 (0.1)	–
Jun. 2006										
Males	43.6 (5.2)	83.7 (2.8)	13.2 (2.6)	–	–	–	0.4 (0.3)	2.7 (0.9)	–	–
Females	24.8 (5.9)	92.9 (1.2)	2.5 (1.0)	–	–	–	–	4.3 (1.4)	0.3 (0.3)	–
Aug. 2006										
Males	36.8 (7.2)	81.9 (5.1)	15.9 (5.2)	–	–	–	–	2.2 (1.1)	–	–
Females	31.0 (4.4)	92.1 (2.3)	2.0 (1.4)	–	–	0.3 (0.3)	–	5.3 (2.2)	0.3 (0.3)	–
Oct. 2006										
Males	44.8 (4.8)	92.0 (2.0)	7.4 (2.1)	–	–		–	–	0.6 (0.3)	–
Females	40.6 (4.9)	91.8 (2.3)	4.1 (1.2)	–	–		–	4.1 (1.7)	–	–
Dec. 2006										
Males	72.5 (4.4)	88.5 (4.0)	10.7 (3.9)	0.4 (0.4)	–		–	0.2 (0.1)	0.2 (0.2)	–
Females	69.4 (5.1)	96.7 (1.1)	2.5 (1.1)	0.3 (0.3)	0.1 (0.1)		–	0.2 (0.2)	0.2 (0.1)	–

Mean values with SE (in parentheses) are shown

% Abs total area occupied by all digestive tract contents, *Det* detritus, *Cru* crustaceans, *Pol* polychaetes, *Hyd* hydroids, *Tur* turbellarians, *Nem* nematodes, *Mac* macroalgae, *Mic* microalgae, *Din* dinoflagellates

Table 3 Three-factor analysis of variance results for abundance of prey in guts of *Caprella penantis* and *C. grandimana*

Source of variation	df	MS	F	P
Species (Sp)	1	2365.7	20.77	0.0000***
Month (Mo)	6	712.8	6.26	0.0000***
Sex (Se)	1	9291	81.56	0.0000***
SpxMo	6	178	1.56	0.1559
SpxSe	1	90.4	0.79	0.3734
MoxSe	6	254.8	2.24	0.0384
SpxMoxSe	6	35	0.31	0.9331
Residual	532	113.9		
Cochran's C test		C = 0.1 (P < 0.01)		
Transformation		None		

df degrees of freedom, *MS* mean square, *P* significance level

*** Significant (*P* < 0.001)

Table 4 Three-factor analysis of variance results for the abundance of detritus in the guts of *Caprella penantis* and *C. grandimana*

Source of variation	df	MS	F	P
Species (Sp)	1	1177.4	8.54	0.0036**
Month (Mo)	6	918.4	6.66	0.0000***
Sex (Se)	1	5203.4	37.77	0.0000***
Sp × Mo	6	279.5	2.03	0.0605
Sp × Se	1	1,4	0.01	0.9198
Mo × Se	6	380	2.76	0.0120
Sp × Mo × Se	6	83.6	0.61	0.7255
Residual	532	137.9		
Cochran's C test		C = 0.1 (P < 0.01)		
Transformation		None		

*** Significant (*P* < 0.001)

** Significant (*P* < 0.01)

(SNK, *P* < 0.01). As expected, the amount of detritus in the diet also fluctuated during the year. The caprellids seemed to feed more on detritus during cold months, but this pe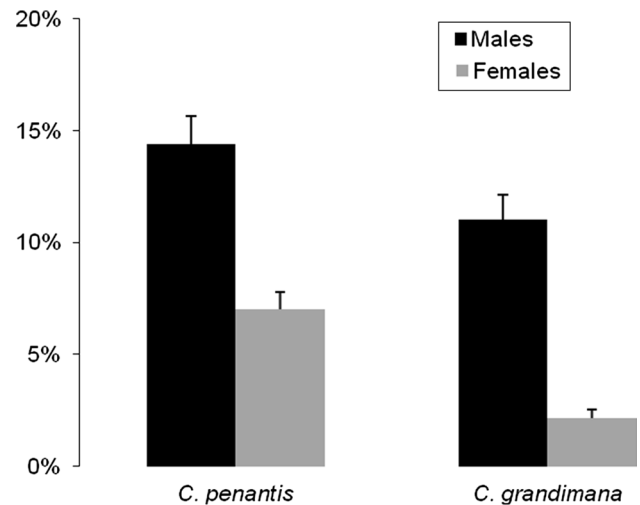rcentage decreased during summer. The percentages of detritus from December 2005 (93.2 ± 1.3 %, *n* = 80) and February 2006 (94.1 ± 0.9 %, *n* = 80) were significantly higher than those for April (86.2 ± 1.7 %, *n* = 80) (SNK, *P* < 0.01), June (88.1 ± 1.4 %, *n* = 80) (SNK, *P* < 0.05),
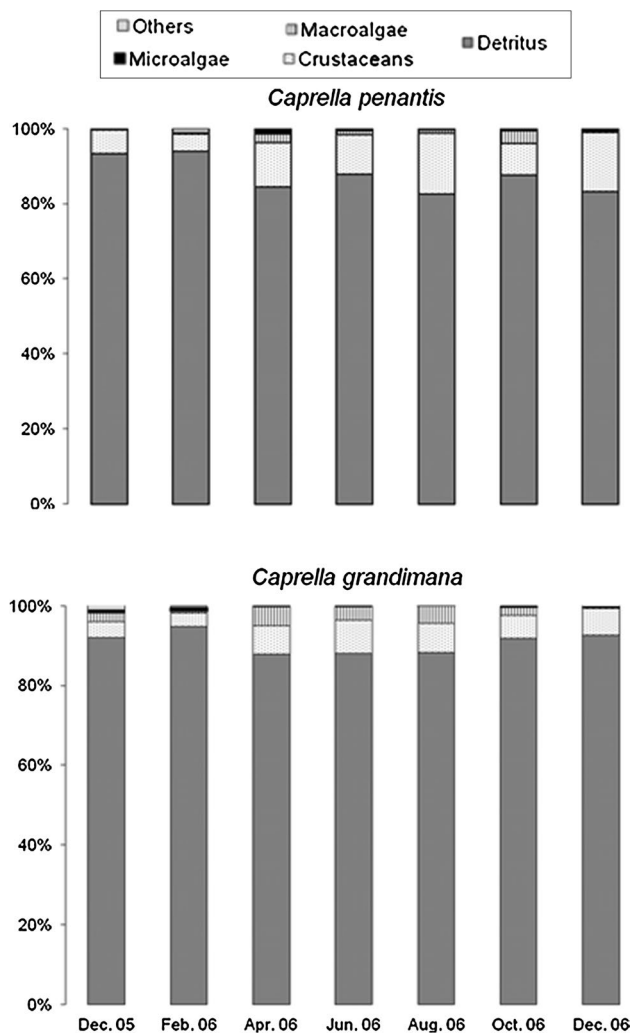
Fig. 1 Percentage of components in digestive tracts of *Caprella penantis* (*upper*) and *C. grandimana* (*lower*) on seven dates, December 2005–December 2006. All prey other than crustaceans combined as "others"

Fig. 2 Percentages of prey in guts of male and female *Caprella penantis* and *C. grandimana*. Values are mean ± SE
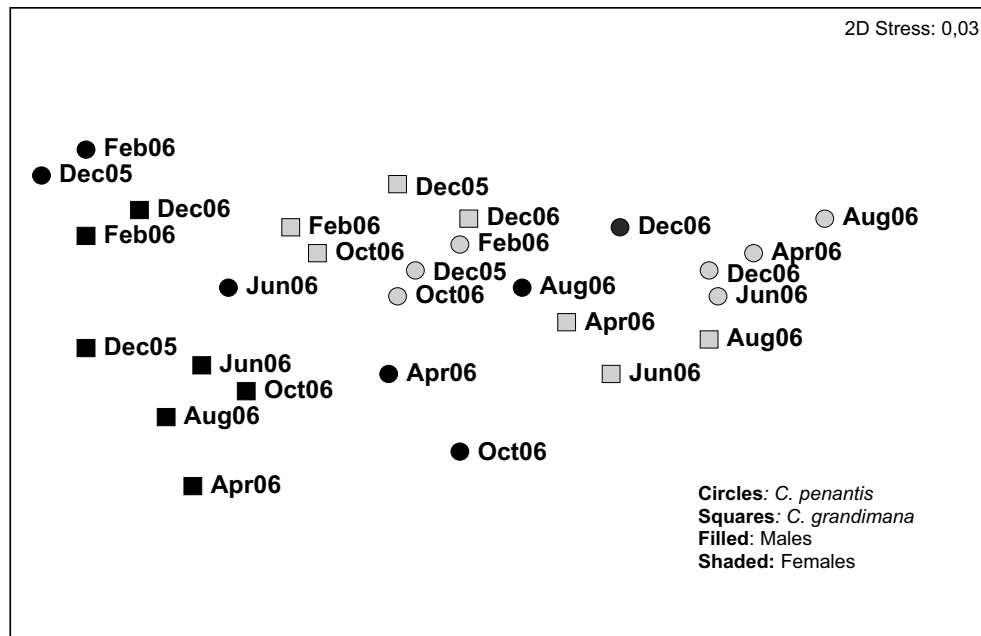
two-dimensional MDS plot (Fig. 3). Consequently, the sex of the specimens had an effect regardless of species and time of year when all components of the diet were considered.

Discussion

Our results revealed that both species feed mainly on detritus (>85 % in *C. penantis* and 90 % in *C. grandimana*) throughout the year and supplement their diets with crustaceans (>10 % in *C. penantis* and >6 % in *C. grandimana*). Macroalgal tissues (>1 % in *C. penantis* and >2 % in *C. grandimana*) and diatoms were less represented, suggesting that these species do not selectively eat microalgae under natural conditions. Dinoflagellates and a wide range of metazoan prey (polychaetes, nematodes, turbellarians, and hydroids) were also found but contributed very little to the diet. A similar pattern was reported for these two species in a previous dietary analysis (Guerra-García and Tierno de Figueroa 2009).

Detritus was always the greatest contributor to the diets of these caprellids, which was consistent with several studies (Jeong et al. 2012; Vazquez-Luis et al. 2013; Guerra-García et al. 2014). Detritus plays a key role in marine invertebrate food webs and trophic pathways in marine ecosystems (Valiela 1995). More energy and materials flow through detrital food webs than through grazer food webs in most freshwater, estuarine, and marine ecosystems, and many animals use detritus directly because it is highly nutritious after a short period of microbial colonization (Mann 1998). In addition, amphipod species richness and total abundance increase with an increase in detrital content

and August (85.2 ± 1.7 %, n = 80) (SNK, P < 0.01). Moreover, the percentages of polychaetes, turbellarians, nematodes, hydroids, and dinoflagellates remained <1 % except in February 2006 for *C. penantis* and December 2005 for *C. grandimana*. Detritus was the dominant component throughout the year in both species, followed by crustaceans.

The largest differences appeared when values were compared between sexes (significant effect of sex, $F = 37.77$, $P < 0.001$ for detritus and $F = 81.56$, $P < 0.001$ for prey). Males of *C. penantis* and *C. grandimana* were clearly more carnivorous than females (Fig. 2). Males fed more on prey (12.8 ± 0.8 %, n = 280) and less on detritus (86.1 ± 1.2 %, n = 280) than females (4.60 ± 0.47 %, n = 280 and 92.2 ± 1.2 %, n = 280, respectively). In addition, males and females were clearly separated in the

Fig. 3 Two-dimensional multidimensional scaling plot for diets of male and female *Caprella penantis* and *C. grandimana* on seven dates, December 2005–December 2006

(Vazquez-Luis et al. 2012). However, whether caprellids should be considered detritivores or omnivores has been widely debated, as the limits for distinguishing between these two strategies are difficult to establish based on percentages of each item. Many authors consider that >95 % detritus in the diet is indicative of a detritivore (Guerra-García et al. 2014), while amphipods that feed alternatively on different trophic resources, such as detritus, small invertebrates, and meiobenthos, are considered omnivores (Fanelli et al. 2009; Navarro-Barranco et al. 2013). We suggest that *C. penantis* and *C. grandimana* should be considered omnivores based on these definitions. Vazquez-Luis et al. (2013) considered *C. grandimana* to be an omnivore and Guerra-García and Tierno de Figueroa (2009) referred to both species as opportunistic.

The second most abundant component after detritus was crustaceans (mainly copepods and some gammarids), which were detected by their mouthparts, broken antennae and gnathopods, or other appendages. Both species live and graze on macroalgae, but this component can have a relatively high content of indigestible fiber and low nitrogen content (Mann 1998). Animal matter is more easily assimilated. Caprellids display cannibalism when food is inadequate, but we found no evidence of cannibalism in the guts we examined. We considered the percentage of particular prey in the gut as an indication of predation by these species. In fact, prey densities are important when investigating variations in dietary composition of benthic amphipods (Yu et al. 2003).

It is common for opportunistic amphipods to change their feeding strategy if trophic resources change owing to environmental factors. Consequently, it seems logical that prey availability can greatly influence feeding habits (Jeong et al. 2012; Ros et al. 2014) and metabolic rate, allowing for highly efficient energy utilization (Doyle et al. 2012). Thus, seasonal peaks in macroalgae and copepod abundance would coincide with greater consumption of these components by *C. penantis* and *C. grandimana*. Several studies in the intertidal areas of Tarifa Island during the same year reported seasonal fluctuations in seaweed (Guerra-García et al. 2011), which are related to cyclic variations in environmental factors, such as temperature, day length, and wave action (Neto 2000). The macroalgal contribution to the diet increased in both species from April to August, when maximum seaweed biomass occurs, whereas both species decreased their feeding on macroalgae during cold months when seaweed abundance was at its lowest. A similar seasonal abundance pattern has been confirmed for copepods in a number of freshwater (Iskaros and El-Otify 2012) and marine ecosystems worldwide (Jaschinski et al. 2011; Kurt and Polat 2012; Rajkumar et al. 2013). Therefore, the higher percentage of prey consumed during summer can be explained by an increase in crustacean availability from April to August. The ability to vary feeding preference in response to prey availability is quite common in marine vertebrates (Varo et al. 2011; Griffin et al. 2012; Matić-Skoko et al. 2014) and in several invertebrate phyla, such as gorgonians (Leal et al. 2014), nemerteans (Caplins et al. 2012),

and echinoderms (Chiantore et al. 2002). Soler-Membrives et al. (2011) found a very similar trend to ours in the tropho-dynamics of the pycnogonid *Ammothella longipes*, which appears to be carnivorous during spring and early summer, but prefers detritus when prey availability diminishes during winter. Opportunistic feeding has been reported in amphipods on several occasions (Werner and Auel 2005; Nunez-Pons et al. 2012), as well as in herbivorous species, such as some talitrids (Bessa et al. 2014). Opportunistic feeding can be a key factor in the success of an invasive species (Maazouzi et al. 2007; Cook et al. 2009). Moreover, some opportunistic, Arctic, under-ice amphipod species switch from a predominantly herbivorous diet in summer to meta-zoan prey, including planktonic copepods in winter, when ice algae are scarce and they must rely on stored lipids as an energy source (Werner and Auel 2005).

Diet composition varied between the species we studied. These differences were also observed in fatty acid-based nutritional analyses by Guerra-García et al. (2004) and Baeza-Rojano et al. (2014). *Caprella grandimana* consumed more diatoms and macroalgae than *C. penantis*, which is more car-nivorous. Caine (1977) reported that a combination of weak swimming setae on antenna 2 and a long basis of gnathopod 2 suggests a predatory mode of life, but our results suggest that *C. grandimana*, which bears these characteristics, has a less predatory mode of life than *C. penantis*. We support the con-clusions of Guerra-García and Tierno de Figueroa (2009) that there are not always clear relationships between morphologi-cal characters and digestive content. The higher percentage of crustaceans found in *C. penantis* than that in *C. grandimana* may be related to body size differences. Poltermann (2001) reported that crustaceans are present in considerably lower proportions in the guts of smaller species and young or small individuals than those of larger species of Arctic amphipods. Jaschinski et al. (2011) reached the same conclusion regard-ing significantly higher $\delta^{15}N$ values in larger amphipods, and Felten et al. (2008) also suggested that greater preda-tory behavior by freshwater amphipods is related to body size. Despite these variations, we found that both species fed mainly on detritus and crustaceans throughout the year. Nelson (1979) also reported an overlap in the diets of several amphipod species and argued that food competition does not play a large role in the structure of the amphipod community in an eelgrass ecosystem where great quantities of detritus are present year around. However, rocky intertidal habitats con-stitute steep vertical gradients, composed of a patchy mosaic of assemblages in which competition for a limiting resource is important. Therefore, these environmental characteristics can strongly affect community composition (Crain and Bertness 2006; Masterson et al. 2008). The presence of *C. penantis* in the lower intertidal (mainly associated with *G. corneum*) and *C. grandimana* at intermediate levels (mainly associated with *C. elongata* and *J. rubens*) avoids interspecific competition.

Finally, anatomic features may also be why adult *C. penan-tis* and *C. grandimana* males were more carnivorous than adult females. Significant intraspecific differences have been reported as fairly common among marine organisms (Bear-hop et al. 2006; Hoeinghaus and Davis 2007; Olabarria et al. 2009). The structures of crustacean feeding appendages have been suggested to influence the diet, as they determine han-dling ability and strength (Kolts et al. 2013). The larger gna-thopod 2 in males than females is probably related to a more predatory strategy, as it allows the male to catch and manipu-late a greater number of prey.

The gut examination methodology used in this study has been applied previously by entomologists (Tierno de Figueroa et al. 2006) and was successfully used for amphipods (Navarro-Barranco et al. 2013; Vazquez-Luis et al. 2013; Guerra-García et al. 2014). Gut content analy-ses are an accurate direct indicator, which is an advantage compared with indirect indicators, such as stable isotope and fatty acid analyses. However, a combination of these methods, together with mouthpart morphological studies and behavioral observations, is needed to obtain complete data on caprellid feeding habits and food web interactions in ecosystems where they dominate (Vazquez-Luis et al. 2013), particularly when studying seasonal variations. Although our understanding of amphipod feeding hab-its has increased during the last few years, several authors have pointed out that qualitative and quantitative character-izations of the amphipod species-specific trophic ecology are needed to better understand their potential role in the trophic dynamics, structure, and nutrient fluxes in marine ecosystems (Graeve et al. 2001; Jeong et al. 2012). In this sense, our study represents the first comprehensive dietary analysis of Caprellidae to investigate seasonal and intraspe-cific variations. Further research is necessary to establish trophic preferences of other amphipods and their roles in the food web of the highly variable intertidal zone.

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