Revista Nordestina de Zoologia
Issn 1808-7663

STOMACH CONTENTS AND FEEDING HABIT OF *Plagusia depressa* (Fabricius, 1775) (CRUSTACEA: DECAPODA: PLAGUSIIDAE) IN SANDSTONE REEFS OF NORTHEAST BRAZIL

Julianna de Lemos Santana[1,3]*, Victor Andrei R. Carneiro[2], Wagner José dos Santos[3] and Tereza Cristina dos S. Calado[3]

[1] Laboratório de Carcinologia – Museu de Oceanografia Prof. Petrônio Alves Coelho (MOUFPE), Universidade Federal de Pernambuco (UFPE). Av. Arquitetura, s/n, Cidade Universitária, Recife, Pernambuco, Brazil.

[2] Laboratório de Algas Marinhas Édison José de Paula – Departamento de Botânica, Instituto de Biociências, Universidade de São Paulo (USP). Rua do Matão, 277, Cidade Universitária, Butantã, São Paulo, São Paulo, Brazil.

[3] Laboratórios Integrados de Ciências do Mar e Naturais (LABMAR/UFAL). Rua Aristeu de Andrade, 452, Farol, Maceió, Alagoas, Brazil.

* Corresponding author; E-mail: julilemos@live.com

ABSTRACT

The aim of this study was to investigate the natural diet of the species *Plagusia depressa*, to unravel the relationship of the species with the environment and their natural diet. A total of 60 individuals were collected and examined between December 2013 and November 2014, of which 33 were males, 26 females and 1 individual of undeterminate sex. The samples were taken by hand during the night at Sonho Verde reefs, Northeastern Brazil, during low tide. Individuals were sent to the laboratory to remove of stomachs for further analysis. The stomachs were ranked in three stages of fullness to denote the volume, separating a low filling (21.43%), medium filling (26.79%) and full (51.78%). In all stomachs, there were great amount not identified material, a result of the advanced stage of digestion of the items. The items found had their frequency of occurrence calculated being Algae the most representative item. The method of points and feeding index reinforce the predominance for algae items. In the environment was observed that the species is mostly herbivorous, but accidentally and opportunistically ingests animal items.

Key words: Brachyura, crab, diet, ecology, herbivory.

RESUMO

O objetivo deste estudo foi investigar a alimentação de *Plagusia depressa*, para desvendar a relação da espécie com o ambiente e a dieta natural. Um total de 60 indivíduos foram coletados e examinados entre Dezembro/2013 e Novembro/2014, sendo 33 machos, 26 fêmeas e 1 indivíduo de sexo indeterminado. A coleta foi realizada manualmente durante a noite, nos recifes de Sonho Verde, Nordeste do Brasil, durante a maré baixa. Os indivíduos foram encaminhados ao laboratório para a remoção dos estômagos. Os estômagos foram classificados em três estágios de repleção estomacal para determinar o volume, separando-os em pouco cheio (21.43%), enchimento médio (26.79%) e cheio (51.78%). Em todos os estômagos foram encontradas grandes quantidades de material não identificado, resultado de um avançado estágio de digestão dos itens. Os itens encontrados tiveram a frequência de ocorrência calculada sendo o item Alga o mais representativo. Além disso os métodos de pontos e índice alimentar reforçaram a predominância de itens algais. No ambiente, foi observado que a espécie é majoritariamente herbívora, mas acidentalmente e oportunisticamente ingere itens animais.

Palavras-chave: Brachyura, caranguejo, dieta, ecologia, herbivoria.

INTRODUCTION

Decapoda (Crustacea) have many feeding habits such as predation, saprophagy, detritivory, and filtration. Thus, they occupy many trophic positions in various aquatic food chains (Carqueija & Gouvêa 1998). Among decapods, crabs (infraorder Brachyura) have great sizes and great architecture of apparatuses of the digestive system. Internal and external structures are adapted to the type of diet and the behavior employed in food capture (Collins *et al.* 2004). When feeding on macroscopic material, it is necessary to use quitinized internal structures to break the food into smaller particles. This propensity for quitinization is due to the ectodermal origin of the foregut. Thus, decapods developed ossicles in their stomach, which, together with the mouth parts, reduce food into small fragments (Brösing 2010). Among studies on stomach contents of Brachyura, the

Portunidae family stands out due to their economic importance (Caine 1974, Hill 1976, Paul 1981, Laughlin 1982, Williams 1982). Nowadays, the number of studies regarding Plagusiidae feeding is scanty, with only two studies on genus *Plagusia* Latreille, 1804, the two herbivorous species *Plagusia capensis* De Haan, 1835 (Bacon 1971), in northern New Zealand, and another *Plagusia dentipes* (De Haan, 1835) (Samson *et al.* 2007) in Tateyama Bay, Japan. About the diet habits of *Plagusia depressa* (Fabricius, 1775), there is a unique contribution given by Hartnoll (1965) who made notes in a comprehensive study on this and other species of grapsoids. According to field observations from Hartnoll (1965) the species feeds by scraping off encrusting algae from the reef, making a relation with the feeding habit and the spoonlike chelae.

The “cliff crab” *Plagusia depressa*, is distributed in the Western Atlantic from North Carolina, USA (Williams 1965) to Bahia, Brazil (Almeida and Carvalho 2014), Central Atlantic in Ascension and St. Helena Islands (Manning & Chace 1990), and in the Eastern Atlantic from Azores, Portugal (Powers 1977) to Angola (Kensley & Penrith 1973). It inhabits intertidal zones in fissures of rocks and corals, and tide pools although the occurrence of this species in reefs is common (Melo 1996). There is a small commercial fishery that targets *P. depressa* (Coelho *et al*. 2004, Rosa *et al*. 2018). There are three studies published about *P. depressa* in Brazil. These studies were performed in the Northeastern region and investigated the general biology, the population structure (Freitas & Santos 2002, 2007) and the individual growth of the species (Coelho *et al.* 2004).

Researches on feeding habit of crustaceans cover many factors of ecologic importance such as the relationship with other species and with the environment, characteristics of digestive anatomy and feeding habits (Oliveira *et al.* 2006). Moreover, the available food play important roles in migration patterns, distribution, ecdysis and reproduction of Brachyura (Laughlin 1982, Oliveira *et al.* 2006). This study investigates feeding habits and analyzes the stomach content of *Plagusia depressa* for the first time and to compare with the already registered in the literature for the other species of the genus.

MATERIALS AND METHODS

The study was conducted along the Sonho Verde sandstone reefs, in the city of Paripueira, state of Alagoas, Northeast Brazil (09º26'35 "S 35º31'31" W and 09º27'15 "S 35º31'45" W) (Figure 1). The climate of the region is tropically, hot and humid and minimum temperature equal to or higher than 18°C. Sonho Verde beach presents reefs in barrier. In this formation, reefs are formed parallel to the coastline, and are separated from the beach by a channel formed when the tide is high (Correia & Soviersozki 2008).

Figure 1. Map of the study area, sandstone reefs at Sonho Verde, Paripueira, Alagoas, Brazil, with arrow indicating the reef line (Adapted from Google Earth).

Monthly sample of *Plagusia depressa* were carried out from December 2013 to November 2014, and five individuals per month were randomly captured by hand at low tide with the aid of flashlights. Specimens were conditioned alive in a thermal box containing ice to avoid stomach content regurgitation, as well as to diminish enzymatic activity in the stomachs (adapted from Carqueija & Gouvêa 1998). Air and water temperature were measured in order to correlate with biotic data. In the laboratory, specimens were identified following Melo (1996) and then dried on absorbent paper, sexed and measured with a 0.01 mm precision digital steel caliper (Carapace Length – CL). A dorsal cut was performed to dissect the digestive tract, using tweezers and scissors, for subsequent analysis (modified from Carqueija & Gouvêa 1998). Stomachs were classified into three states of repletion (RE) was classified into three classes: 1 (less filled), 2 (medium filled) and 3 (full), as suggested by Pontes & Arruda (2005). Empty stomachs were not considered. Subsequently, stomachs were weighed with a 0.01 g precision digital scale, and stomach contents were removed with water jets and deposited in Petri dishes for stereomicroscope analysis (Stevens *et al.* 1982). Food items were identified to the lowest possible taxonomic level and preserved in 4% formaldehyde-saline solution. Digested organic residues of material were classified as “digested material” (DM) (Branco & Verani 1997, Mantelatto & Christofoletti 2001).

The frequency of occurrence of each food item was calculated according to $FO = \left(\frac{bi}{N}\right).100$ to determine the percentage frequency of the stomachs (b) where a food item ($_i$) occurs in relation to the total number of stomachs that are no empty (*N*) (Oliveira *et al.* 2006). To calculate the number of points of each item, the contribution (%) of this item in each stomach was estimated during the analysis and points were assigned to each stage of stomach repletion (ER1 - 0.35 point, ER2 - 0.65 point, ER3 - 1 point). The points method was applied to calculate the percentage volume (*V*) of every food item. We followed Williams’ formula (1981): $\sum_{j=1}^{n}\left(\frac{aij}{A}\right).100$, where *A* is the total number of points for all items, *n* the total number of stomachs and *aij* the number of points of the item found in the

stomachs examined. The contribution and points of the repletion stage were grouped according to each item. In order to understand the relevance of each food item, the food index proposed by Kawakami & Vazzoler (1980) was used. This index combines the two methods, and is calculated according to the formula $FIi = \frac{FOi \cdot Vi}{\sum_{i=1}^{n}(FOi \cdot Vi)}$ where *FIi* is the food index of item *i*, *n* the total number of stomachs, *FO* the frequency of occurrence (%) and *V* the relative contribution of volume (%) of item *i*. The chi-square test (χ^2) at a 5% significance level ($\alpha = 0.05$) was used to verify whether there are differences in the frequency of each food item consumed between sexes, being used for this analysis in points of each category of prey.

RESULTS

A total of 60 individuals were collected and examined between December 2013 and November 2014, being 33 (55%) males, 26 (43.33%) females and 1 (1.66%) individual whose sex was not determinate. The CL varied between 12.80 - 48.77 mm (30.13 ± 9.39 mm on average) for males, 14.12 - 48.40 mm (29.93 ± 11.48 mm on average) for female and the individual of undetermined sex had 12.00 mm of CL. Among the females collected, seven (26.92%) were ovigerous. Air temperature varied between 26° C and 28° C, water temperature between 26.8° C and 28.5º.

Natural Observation

In samples started immediately after the sunset, individuals were still hiding in slits in the reef, being a few found in puddles and therefore needed more time for capture. Also, it was observed that in the sample that preceded the sunrise specimens were found within only 5 minutes, all in pools, feeding on algae. When there were no dashes or slits in the reef, specimens were caught in reentrances on the side of the reef facing the ocean, under wave action. The analyzed specimens do not get very distant from the water, which indicates a greater vulnerability to desiccation and, therefore, these animals remain in the moist

surface of the reef, which has the humidity increased by the associated algae (Masunari & Dubiaski-Silva 1998).

Of the 60 analyzed stomachs, 56 (93.33%) presented content for identification of which 12 (21.43%) were classified as ER 1, 15 (26.79%) were classified as ER 2 and 29 (51.78%) as ER 3. Among females, 4 (15.39%) had ER 1, 7 (26.93%) in ER2 and 15 (57.68%) ER 3. Regarding males, 7 (24.14%) were found in ER1, 8 (27, 59%) in ER2 and 14 (48.27%) in ER 3. Four empty stomachs were collected in July (two individuals), in August (one individual) and in September (one individual). The latter was found in pre-ecdysis, and so it had consumed minute amounts of food which were already in the midgut.

The total frequencies of occurrence of food items identified were: Algae (100%), DM (100%), Mollusca (66.07%), Sediment (57.14%), Crustacea (25.00%), Ascidiacea (17.85%) and Polychaeta (7.14%). Items were grouped (Table I) and had their respective frequencies calculated (Table II). The predominant food items in the diet of *P. depressa* were algae and animals that usually live in or close to algae such as ascidians, polychaetes, tanaids, mollusks and amphipods.

The volumes and occurrences of amphipods (1.7% and 16.07%), polychaetes (0.6% and 7.14%) and tanaids (0.2% and 1.78%) living between algae and sediments are relatively low and likely represent accidental ingestion next to vegetal feed grounds. Amphipods are quite representative because they have larger body mass and thus occupy a large volume. They are mainly found in algae when compared to the other groups and are more common in the sediment and inside encrusting invertebrates. It is difficult to determine the contribution of each item to the total volume of the stomach due to the different stages of repletion that they present. In order to understand these values, the points method was applied, and the food index of each item was calculated. Frequencies of each item relative to the total volume of the stomachs confirm the result of frequency of occurrence, emphasizing the importance of algae and DM for *P. depressa* diet (Table III). There were no significant differences between

sexes according to chi-square test ($\alpha = 0.05$; $\chi^2 = 3.84$) applied on the volume percentage (V) (Table IV).

Table I. General description of the items found in the stomachs of *Plagusia depressa* from sandstone reefs at Sonho Verde, Paripueira, Alagoas, Brazil.

Group	**Type of fragments**
Rhodophyta	Fragments
Cyanobacteria	Whole algae or fragments that allowed identification
Dinozoa	
Heterokontophyta	
Mollusca	
Gastropoda	Shell fragments
Annelida	
Polychaeta	Fragments
Arthropoda	
Crustacea	
Amphipoda	Anterior part fragments and whole animals
Decapoda	Chellipod fragments
Tanaidacea	Whole animal
Chordata	
Ascidiacea	Pharynx fragments

Table II. Description of most frequent food items and their frequency of occurrence in the analyzed stomachs (DM – Digested Material; ND – Not Defined) of *Plagusia depressa* from sandstone reefs at Sonho Verde, Paripueira, Alagoas, Brazil.

Food item	**FO%**	**Habit**
Rhodophyta		
Antithamnion sp.	10.71	Macroalgae/Epiphyte
Antithamnionella breviramosa (E.Y.Dawson) E.M.Woll.	8.92	Macroalgae/Epiphyte
Ceramium sp.	16.07	Macroalgae/Epiphyte
Dipterosiphonia dendritica (C.Agardh) F.Schmitz	8.92	Macroalgae/Epiphyte
Gracilaria sp.	7.14	Macroalgae/Bentic
Hypnea pseudomusciformis Nauer, Cassano & M.C.Oliveira	41.07	Macroalgae/Epiphyte
Polysiphonia sp.	48.21	Filamentous/Epiphyte
Rhodophyta ND	16.07	
Ceramiacean ND	16.07	
Coralinacean ND	21.42	
Cyanobacteria		
Lyngbya majuscula Harv. ex Gomont	8.92	Filamentous/Epiphyte
Lyngbya sp.	19.64	Filamentous/Epiphyte
Oscillatoria sp.	7.14	Filamentous/Epiphyte
Dinozoa		
Peridinium quinquecorne Abé	16.07	Unicellular/Planctonic
Peridinium sp.	7.14	Unicellular/Planctonic
Heterokontophyta		
Amphora angusta W.Greg.	7.14	Unicellular/Epipelic
Campyloneis grevillei (W.Sm.) Grunow	14.28	Unicellular/Planctonic
Climacosphenia moniligera Ehrenb.	7.14	Unicellular/Planctonic
Cocconeis scutellum Ehrenb.	28.57	Unicellular/Epiphyte
Coscinodiscus sp.	12.50	Unicellular/Planctonic
Fragilaria oceanica Cleve	7.14	Unicellular/Planctonic
Frustulia rhomboides (Ehrenb.) De Toni	7.14	Unicellular/Planctonic
Grammatophora marina (Lyngb.) Kütz.	32.14	Colonial/Epiphyte
Grammatophora sp.	25.00	Colonial/Epiphyte
Melosira nummuloides C.Agardh	12.50	Colonial/Epiphyte
Melosira sp.	8.92	Colonial/Epiphyte
Licmophora sp.	8.92	Colonial/Epiphyte
Navicula sp.	19.64	Unicellular/Planctonic
Ascidiacea	17.85	
Amphipoda	*16,07*	
Mollusca ND	*66,07*	
Eulithidium affine (Adams, 1850)	*32,14*	
Polychaeta	*7,14*	
DM	100.00	

Table III. Volume (V) and Feeding index (FI) of food items found in the stomachs of *Plagusia depressa* from sandstone reefs at Sonho Verde, Paripueira, Alagoas, Brazil.

Food Item	V%	FI
Algae	63.7	0.6824
DM	22.6	0.2421
Mollusca	9.9	0.0700
Amphipoda	1.7	0.0032
Ascidiacea	1.0	0.0015
Polychaeta	0.6	0.0005
Decapoda	0.2	0.00007
Tanaidacea	0.2	0.00003

Table IV. Volume (V) of each food item and chi-square test (χ^2) between the frequencies for males and females of *Plagusia depressa* from sandstone reefs at Sonho Verde, Paripueira, Alagoas, Brazil.

Food Item	Male V (%)	Female V (%)	χ^2
Algae	62.64	64.17	0.02
DM	24.54	20.65	0.33
Polychaeta	-	1.38	-
Mollusca	10.32	9.66	0.02
Tanaidacea	0.37	-	-
Amphipoda	0.99	2.38	0.57
Decapoda	0.37	-	-
Ascidiacea	0.77	1.76	0.38

DISCUSSION

The great variety of phytoplankton ingested by *P. depressa* as well as epiphytic algae ingested together with macroalgae, may also be related to filtration of suspended food by bristles of their buccal parts, as suggested by Schmalfuss (1976) for *Percnon gibbesi* (H. Milne Edwards, 1853). Red algae were the predominant macroalgae in stomach contents (being the most frequent

Polysiphonia sp. and *Hypnea musciformis*) and green algae (chlorophytes, here represented only by *Cladophora glomerata*) were the least type found. The same was observed for *Plagusia dentipes* by Samson *et al.* (2007) in Japan, although this species prefers Corallinaceae. In the present study, fragments of coralline algae were relatively frequent in the contents (FO = 21.42%). Another plagusid crab, *P. capensis,* also exhibited predominance of brown and red algae (Bacon 1971). This great amount probably reflects the almost year-round availability and abundance of these algae in the environment (Wolcott & O'Connor 1992, Samson *et al.* 2007). Almost all Macroalgae consumed by *P. depressa* was filamentous, with exception of *Hypnea pseudomusciformis*. The preference of grapsoids for filamentous algae was also observed by Kennish (1996) for *Grapsus albolineatus* Latreille, in Milbert, 1812.

Whereas most species of marine brachyurans, mainly Portunidae, as, for example, all species of the genus *Callinectes* (Stimpson, 1860) (Paul 1981, Laughlin 1982, Hsueh *et al,* 1992, Branco & Verani 1997, Mantelatto & Christofoletti 2001) are omnivorous and that most herbivorous species has abundant food, a species such as *Plagusia* would hardly ever be unsuccessful, unless the area had suffered some anthropogenic interference or undergone catastrophic events that decimated all plant diversity. The swimming crabs of Portunidae family prefers animal matter such as crustaceans, fish and mollusks (Hill 1976, Wear & Haddon 1987), this is due to greater voracity, activity and growth rate of this family (Edgar 1990), requiring a greater amount of nutrients to perform these functions satisfactorily.

Mollusk shells were frequently found in the diet of *P. depressa* and constitute an important component in the stomach contents of most brachyurans such as *Scylla serrata* (Forskål, 1775) (Hill 1976) and *Portunus pelagicus* (Linnaeus, 1758) (Josileen 2011). The consumption of mollusks shells and not only soft parts is directly related to the strength of the chelipeds of the species. For example, Devi *et al.* (2013) studied the grapsoid *Varuna litterata* (Fabricius, 1798), and found no remains of mollusks in the stomach contents.

The considerable amount of digested material raises the assumption that *P. depressa* ingest animal items such as fish and soft mollusks. It also reflects a rapid digestion process off materials of animal origin, as they contain endogenous digestive enzymes that can accelerate digestion at higher temperatures, such as in *Plagusia depressa*. This type of chelae has a regular face (without elevations). Most ingested items that could be identified were sessile organisms or organisms of reduced mobility, which may indicate that the DM found in the stomachs of *P. depressa* could be prey which would soon be debilitated or dead.

There was variation only among ovigerous females, which had stomachs at maximum repletion stage (ER3), and not ovigerous females, which had varied stages. Ovigerous females also presented a slightly higher volume of DM than those that did not carry eggs. According to Kennish (1996), consumption of organic matter (possibly by animal origin), even if opportunistic, appears to be a key factor for reproductive success. Tanaidacea and Decapoda appear only in stomach contents of males, and Polychaeta only in females, although these results are not significant due to possible accidental or opportunistic intake of these items, as well as the total number of analyzed individuals.

Reduction of food activity during ecdysis events is necessary since the foregut will also be eliminated during ecdysis because of its ectodermal origin such as the carapace (Abelló 1989). The same was found by Carqueija & Gouvêa (1998) with the portunid *Callinectes marginatus* (A. Milne-Edwards, 1861). According to Williams (1982), after leaving their old carapace, crabs ingest large abundance of mollusk shells and limestone materials to replenish the supplies of nutrients needed to form a new rigid shell. This study focused on *Portunus pelagicus* crab, that fills the stomach with a lot of limestone and less organic matter. The same was observed by Hill (1976) with *Scylla serrata*, another species of crab. In the present study, no specimens of *P. depressa* were found in immediate post-molten phases (with the carapace still soft), so this aspect cannot be compared with other species.

Sediment intake was observed but was not considered relevant because although it was present in more than half of the stomachs analyzed, the small

volume of sediments found indicates that the intake is accidental. Other brachyurans, especially Portunidae, which have crustaceans and mollusks among their main food items, ingest large amounts of sediment to aid mechanical digestion of more rigid parts (Mantelatto & Christofoletti 2001, Ferreira *et al.* 2011). In addition, sediment components like shells of mollusks are a source of minerals to form a new carapace (Williams 1982).

Brachyurans rarely ingest their prey intact due to the manipulation of food by chelae and buccal parts, so the number of individuals ingested is not as important as the volume of food ingested (Branco & Verani 1997). The high importance of algae and DM in *P. depressa* diet, revealed by the point methods, is similar to that seen by Samson *et al*. (2007) for *Plagusia dentipes*. These results are different from those reported for portunid crabs such as *Callinectes danae* (by Branco & Verani 1997) and *Ovalipes catharus* (by Wear & Haddon 1987), since point methods and food index (FI) analysis indicated the importance of crustaceans and mollusks. For *Hepatus pudibundus*, an Aethridae crab, Mantelatto & Petracco (1997) observed that the contribution of crustacean volume and DM were the most expressive among the stomach contents, when using the point method. The same was found by Mantelatto & Christofoletti (2001) for the portunid crab *Callinectes ornatus*.

All field surveys reported *Plagusia depressa* feeding exclusively on algae, corroborating the asssumptions of Hartnoll (1965) observation made in individuals from Jamaica, who also reported this species scraping a thin layer of crustose algae with a spoon-like chela. Other species resident of this site such as *Pachygrapsus transversus* (Gibbes, 1850) and *Eriphia gonagra* (Fabricius, 1781) were reported feeding on shellfish. The same was observed by Christofoletti *et al.* (2010) which pointed out that *P. transversus* feeds invertebrates rather than on algae. Cannicci *et al.* (2002) reported a similar behaviour between *P. transversus* and its congener species *Pachygrapsus marmoratus* (Fabricius, 1787). This food habits of species that cohabit in the same environment that *P. depressa* favour the abundance of food for the species during the whole year since, there is a little consumption algae. The results

corroborate with that found by Samson *et al.* (2007) for *Plagusia dentipes* (De Haan, 1835) collected in Tateyama Bay, Japan. This relationship among of animal items and algae found in the contents was also recorded for *Plagusia squamosa* (Herbst, 1790) from Pacific coast of Mexico and Hawaiian Islands, observed by Frick *et al.* (2011), because the amphipods observed in the stomach contents of this species lived on the algae that grows in the carapace of the turtle *Lepidochelys olivacea* (Eschscholtz, 1829). Thus, as seen for other congener species, *Plagusia depressa* is mostly herbivore and items of animal origin are opportunistically ingested. Animals with this habit maintain the algae cover of the reef, which is of great importance for the installation of other algae species and associated animals.

ACKNOWLEDGEMENTS: The authors would like to thank Fernando Luis Medina Mantelatto, Marcio Paiva Guimarães, Marina de Sá Leitão Câmara de Araújo and Victor Ribeiro Cedro, and all the members of LABMAR-UFAL for your important contributions in this research. We would like to thank the reviewers for the suggestions.

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Submetido: 15/03/2019

Aceito: 10/04/2019