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The abundance of prospective natural food for sea cucumber *Holothuria atra* at Karimunjawa Island waters, Jepara, Indonesia

RETNO HARTATI[1,2♥], WIDIANINGSIH[2], AGUS TRIANTO[2], MUHAMMAD ZAINURI[3], AMBARIYANTO[2]
[1]Doctoral Program in Marine Science, Department of Marine Science, Faculty of Fisheries and Marine Science, Universitas Diponegoro. Jl. Prof. Dr. Soedharto, SH. Kampus Tembalang, Semarang 50275, Central Java, Indonesia. Tel./Fax. +62-24-7474698, ♥email: retnohartati.undip@yahoo.com
[2]Department of Marine Science, Faculty of Fisheries and Marine Science, Universitas Diponegoro. Jl. Prof. Dr. Soedharto, SH. Kampus Tembalang, Semarang 50275, Central Java, Indonesia.
[3]Department of Oceanography, Faculty of Fisheries and Marine Science, Universitas Diponegoro. Jl. Prof. Dr. Soedharto, SH. Kampus Tembalang, Semarang 50275, Central Java, Indonesia.

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Abstract. *Hartati R, Widianingsih, Trianto A, Zainuri M, Ambariyanto. 2017. The abundance of prospective natural food for sea cucumber* Holothuria atra *at Karimunjawa Island waters, Jepara, Indonesia. Biodiversitas 18: 947-953. Holothuria atra,* as one of the most abundant and widely distributed sea cucumber species, is an omnivore, consuming detritus, uneaten food, and algae in the substrate. For sea ranching purpose, the objective of present works was aimed to determine potential food availability for *H. atra* in Karimunjawa Island of Jepara. Biotic environment factors in proposed location were assessed including seaweed and seagrass associated communities, epiphytes on the seagrass, phyto and zooplankton, benthic organism, microphytobenthic as chlorophyll-a, phaeophytin and chloropigmen as well as sediment characteristics. There were six species seagrasses and 10 species of seaweeds found in Karimunjawa waters, with *Enhalus acoroides* and *Thalassia hemprichii* provide shelter and food for *H. atra*. Algal epiphytes use seagrass leaves and rhizome as substrate and there were 28 genera of them belong to Chrysophyta, Chlorophyta, and Cyanophyta. The variety and abundance of phytoplankton, zooplankton the benthos enrich the location. The chlorophyll-a, phaeophytin and total chloropigment in the sediment of Karimunjawa waters could be representative of microphytobenthos. The substrate of Karimunjawa waters consisted of fine sandy-coarse sediment and the content of organic matter may be important for feeding activity of *H. atra*.

Keywords: *Holothuria atra*, Indonesia, Karimunjawa Island, natural food, sea cucumber, teripang keling

INTRODUCTION

Holothuria atra was commonly known as black sea cucumber, or locally named as Teripang Keling or Teripang Hitam is one of the most abundant and widely distributed sea cucumber species in most parts of the Indo-Pacific region. They inhabit a wide range of depths and a broad variety of habitat ranging from rocky reefs to mudflats and commonly associated with seagrass beds. *H. atra sometimes* is naked and black but usually, covers itself with a coating of light coral sand held in place by tube feet. The integument is thick and firm. This species lacks cuvierian tubule and thus may be handled without the annoyance of adhesive threads.

Holothuria atra prefers lives in coastal seagrass beds, soft and hard substrates of coral reefs as having been previously reported by Kinch et al. (2008); Dissanayake and Stefansson (2012); Setyastuti (2014) and Asha et al. (2015). Just like other sea cucumbers, *H. atra* is omnivore-consuming detritus, uneaten food, and algae in the substrate. It ingests sand grains, digests the nutrient, and then expels sand pellets both in day and night time.

Sea cucumber has high demand market and value because not only for culinary delicacy but people believe that sea cucumber has many purposes for human health such as traditional medicine and aphrodisiac (Ramon et al. 2010). A study showed that sea cucumber has potential activity as anti HIV (Human Immunodeficiency Virus) due to its content of the CCR5 (Chemokine receptor 5) antagonist compounds (Hegde et al. 2002). However, the natural stock of the sea cucumber decreased year by year due to overfishing. One possible effort to overcome this problem is sea ranching. To date, none has been done in sea ranching of sea cucumber in Indonesia. Karimunjawa Island is known as a source of sea cucumber production (Pradina et al. 2010), one species of them is *H. atra*. According to Giraspy and Walsalam (2010), one of the keys to successful sea ranching is site selection. Along with three other locations (Teluk Awur, Bandengan, and Ujung Piring waters), Karimunjawa Island is proposed as sea ranching location for *H. atra*. Naturally, the abundance of sea cucumber is very strongly affected by the availability of natural food. As macrozoobenthic organisms or secondary or higher consumers, they may consume the sedimentary material derived from various decomposed autotrophic organisms (Wardiatno et al. 2015). The objective of the present research was to determine potential food availability for *H. atra* in its habitat.

MATERIALS AND METHODS

Study area

The research was conducted at *H. atra* habitat (Hartati et al. 2002; Pradina et al. 2010) i.e. Karimunjawa Island in Jepara Regency. The map is presented in Figure 1 and the position of each station is presented in Table 1. The determination of potential food for the sea cucumber was carried out in Marine Biology Laboratory, Department of Marine Sciences, Faculty of Fisheries and Marine Science, Universitas Diponegoro, Semarang, Central Java, Indonesia.

The prospective habitat and natural food in research location were assessed including seaweed and seagrass associated communities, epiphytes on the seagrass, phytoplankton, and zooplankton, benthic organisms, microphytobenthos as chlorophyll-a, phaeophytin and chloropigmen. Five transect lines and three quadrate transects were established at the site running perpendicular to the shore and ending at approximately 500 meters deep edge of the bed. The edge of the bed was defined as the furthest growing sea grass from shore. The quadrate consisted of 1-m^2 of PVC-pipe separated into 100 squares. If any seagrass was found in a square, then it was identified and counted for their shoot. Percent cover for each species was calculated from the shoot density according to English et al. (1994). The number colony of macroalgae found in each quadrate was also recorded. A sample of each species was saved for later identification in the lab. Ten or more leaves of seagrass were selected at random; the epiphytes were carefully scraped from these blades, identified and counted.

Tabel 1. Geographical position of sampling stations and sub-stations at Karimunjawa waters, Jepara, Central Java, Indonesia

Station	South	East
L1	5° 53' 19.19"	110° 26' 46.17"
L2	5° 53' 14.15"	110° 26' 41.95"
L3	5° 53' 10.48"	110° 26' 36.29"
L4	5° 53' 6.67"	110° 26' 30.80"
L5	5° 53' 2.73"	110° 26' 25.45"

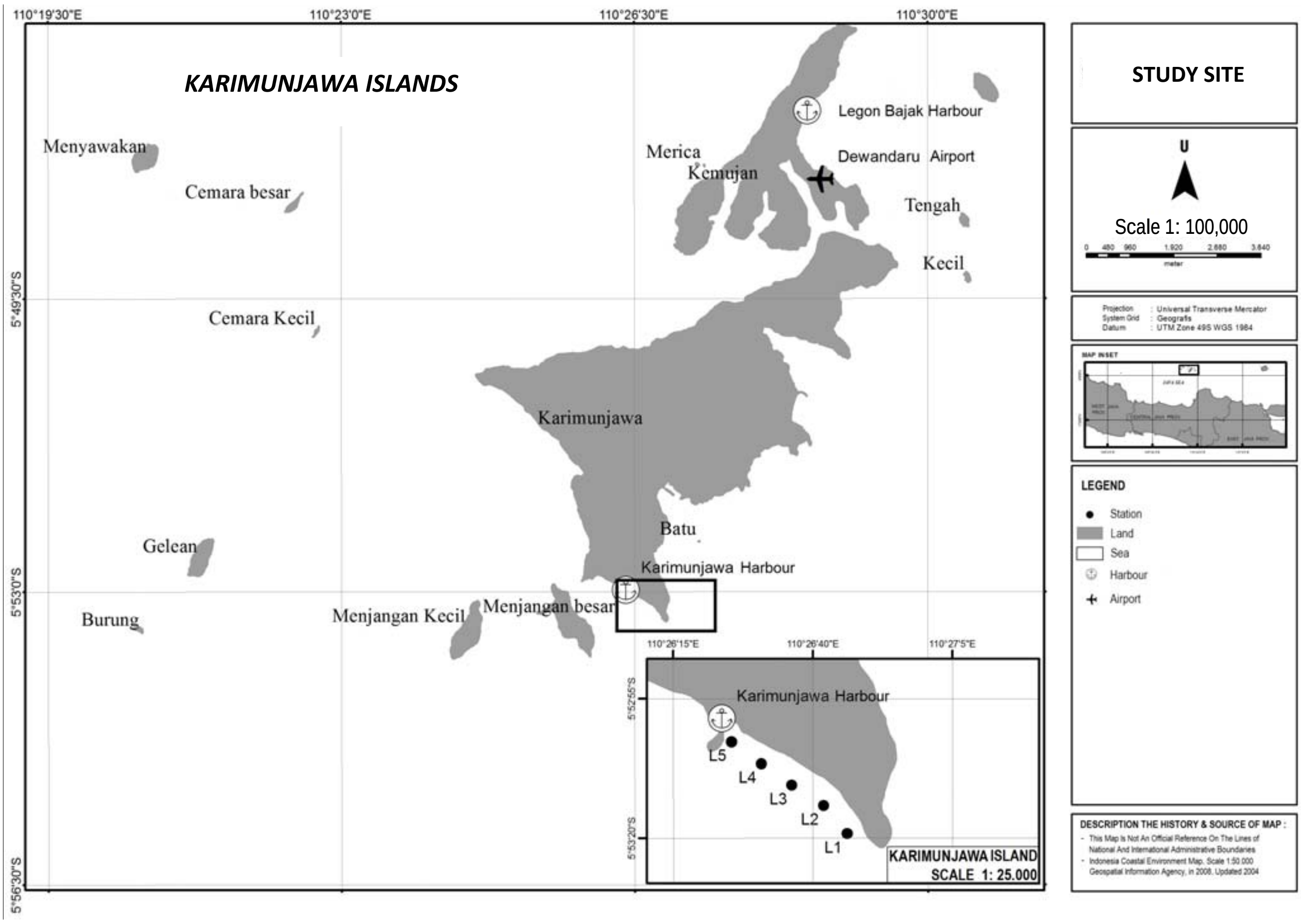

Figure 1. Map of sampling stations of Karimunjawa waters, Jepara, Central Java, Indonesia

Plankton was collected from the surface waters (0.5-1 m depth) using a plankton net with a 37-µm mesh (for phytoplankton) or 300 µm (for zooplankton) and a 20cm opening. The net was pulled behind a boat at the end of a 10-m-long nylon rope for 10 minutes. The distance covered was about 500 m and the volume of seawater filtered for each plankton haul was 350 m^3. After each haul, the sample was taken to the laboratory after preserving with 4% buffered formalin. Plankton cell identification and counting were done using a Sedgwick rafter counting cell under a light microscope. Macroinvertebrate as benthic organism communities was sampled at each of the five stations using core sampler. The samples were separated from the sediment using sieves (500 µm). All the animals collected were immediately fixed in formaldehyde (4%) in the field and then transferred to 70% ethyl alcohol. The macroinvertebrates were sorted, identified to the lowest possible taxon (species, genus or families) and counted under a stereomicroscope.

Seawater and sediment samples were taken to measure chlorophyll a and total chloropigments to determine the phytobenthic biomass. Sediment was taken from the surface (up to 3 cm depth) and in the laboratory, they were dried in an oven at 60°C for 72 h. Total Organic Matter (TOM) analysis was done using the 'ash method' (Slater and Carton 2009). For each sample, 12 g were taken and burned at 450°C for five h in a combustion chamber. Samples were then cooled in a desiccator and the final weight was recorded. TOM content was then calculated as a loss on ignition. The chlorophyll was extracted with a standard ethanol extractions and pre- and post-acidification measurements (Lorenzen 1967). A Perkin-Elmer Lambda 3BUV/VIS spectrophotometer with a 1 nm spectral bandwidth and optically matched 4 cm micro-cuvettes are used in the present work. Total chloropigments was calculated as the sum of chlorophyll a and phaeophytin. Abiotic environmental factors, such as total organic matter and grain size of sediment were analyzed (Robert 1979).

RESULTS AND DISCUSSION

Results

There were six species of seagrasses found in Karimunjawa Waters (Table 2), i.e. *Enhalus acoroides, Thalassia hemprichii, Halophila ovata, Halodule pinifolia, Cymodocea rotundata,* and *C. Serrulata.* Seaweed was also found in the same location of the seagrasses. Average density of seagrass was 12.0-19.6 shoot.m^{-2} and coverage of 12.3-29.4 shoot.m^{-2} (Tabel 2). *T. hemprichii* and *E. acoroides* coverage were high (42.2 and 30.8 % cover). While there were 10 species of seaweed in Karimunjawa waters i.e. *Padina australis, Halimeda opuntia, H. macroloba, Halimeda* sp.*, Turbinaria decurrens, Caulerpa racemosa, Dyctiota* sp.*, Udotea argentea, U. flabellum* and *Sargassum cristaefolium,* with an average density of 0.2-4.6 colony.m^{-2}. The two highest species abundances were *P. australis* (5 colony.m^{-2}in line 1) and *H. macroloba* (2.4 colony.m^{-2} in line 1).

Table 2. Seagrass and seaweed-associated composition, shoot or colony density abundance (A = shoot.m^{-2} or colony.m^{-2}) and coverage (B=%/m^2) in Karimunjawa waters, Jepara, Central Java, Indonesia

	Station									
Species	**L1**		**L2**		**L3**		**L4**		**L5**	
	A	**B**	**A**	**B**	**A**	**B**	**A**	**B**	**A**	**B**
Seagrass										
Enhalus acoroides	32	26.6	27.2	30.8	29.6	13.8	3.2	12.4	8	19.9
Thalassia hemprichii	17.6	42.2	20	29.1	35.2	14.1	30.4	15.4	45.6	24.0
Halophila ovata	8.8	19.5	1.6	9.4	10.4	20.1	7.2	40.6	0.8	3.1
Halodule pinifolia			8	19.5	3.2	9.4				
Cymodocea rotundata			3.2	37.5						
Cymodocea serrulata						4.2				
Average	19.5	29.4	12.0	25.3	19.6	12.3	13.6	22.8	18.1	15.7
Seaweed										
Padina australis	1.2		4		5		4.6		4.4	
Halimeda opuntia	2		1.2		0.4		0.6		0.8	
Halimeda macroloba	2.4		1.8		0.6		0.8		0.4	
Halimeda sp.			0.2				0.4			
Turbinaria decurrens			0.4							
Caulerpa racemosa					0.4				0.2	
Dyctiota sp.							0.6			
Udotea argentea	0.8									
Udotea flabellum	0.6				0.4					
Sargassum cristaefolium									0.2	
Total colony.m^{-2}	7.0		8.8		6.8		7.0		6.0	

In this study, many kinds of algae were also found in seagrass beds. Algal epiphytes use seagrass leaves and rhizome as substrate. The species composition of epiphyte is presented in Tabel 3. There were 27 genera belong to Division of Chrysophyta, Chlorophyta, and Cyanophyta. Generally, the genera of Chrysophyta were more abundant than other Division. In all research location, the epiphytes density (cell.cm^{-2}) and a number of genera (n) were high in seagrass of *E. acoroides,* due to their larger leaves.

There were 26 genera of phytoplankton and 10 genera of zooplankton found in the research locations (Table 4). The diversity and evenness of phyto and zooplankton were high, and there are no dominance genera (Tabel 5).

The number of genera and abundance of benthic community varies with stations. The number of genera and average abundance of macrozoobenthos found in Karimunjawa waters was 22-25 genera and 584-4.416 indv.m^{-2}, respectively consisted of polychaeta (70%), crustaceans (23%) and bivalves (7%). The community structure of macrozoobenthos revealed that the diversity and evenness were high, i.e. 3.79 and 0.85 respectively. This indicated a balance in macrozoobenthos communities in the locations.

The concentration of chlorophyll a and phaeophytin in the seawater and sediment of every sampling station of Karimunjawa were shown in Table 6. The highest chlorophyll a and phaeophytin of sea water occurred in stations L3 and L1. The highest chlorophyll a and phaeophytin of sediment were found in stations L4 and L1.

Table 4. The average abundance of phytoplankton (cell.L^{-1}) and zooplankton (indv.L^{-1}) number of genera (n) found at Karimunjawa waters, Jepara, Central Java, Indonesia

Phytoplankton	Density cell.L^{-1} (number of genera)	Zooplankton	Abundance, indv.L^{-1} (number of genera)
Chromophyta	730.000 (27)	Average abundance	2.320
Cyanophyta	14.000 (1)	Number of genera	10
Total	744.000 (28)		

Table 5. Community structure indices of phytoplankton and zooplankton in Karimunjawa waters, Jepara, Central Java, Indonesia

Plankton	Community structure indices		
	Diversity	Evenness	Dominance
Phytoplankton	3.22	1.01	0.01
Zooplankton	3.59	0.97	0.02

Table 3. Average epiphytes density (cell.cm^{-2}) and number of genera (n) in seagrass blades in Karimunjawa waters, Jepara, Indonesia

Seagrass species	Chrysophyta	Chlorophyta	Cyanophyta	Total Density (No. of genera)
Enhalus acoroides	2.226 (20)	182 (2)	725 (4)	3.133 (27)
Halophila ovata	732 (17)	62 (1)	521 (4)	1.315 (22)
Thalassia hemprichii	800 (12)	46 (1)	704 (4)	1.244 (18)
Halodule pinifolia	649 (7)	43 (1)	704 (4)	1.396 (12)
Cymodocea rotundata	727 (12)	50 (1)	541 (4)	1.318 (17)
Cymodocea serrulata	725 (10)	46 (1)	548 (4)	1.319 (16)

Table 6. The concentration of chlorophyll-a, Phaeophytin and Total chloropigment in the seawater (ug.L^{-1}) and sediment (µg.g^{-1}) of each sampling station of Karimunjawa waters, Jepara, Indonesia

Station	Seawater			Sediment		
	Chlorophyll-a (ug.L^{-1})	Phaeophytin (ug.L^{-1})	Total chloropigment (ug.L^{-1})	Chlorophyll-a (µg.g^{-1})	Phaeophytin (µg.g^{-1})	Total chloropigment (µg.g^{-1})
L1	0.000168	0.061700	0.061868	0.003200	0.115080	0.11828
L2	0.000161	0.048300	0.048461	0.001960	0.092920	0.09488
L3	0.000178	0.050997	0.051175	0.002670	0.087580	0.09025
L4	0.000172	0.057940	0.058112	0.007120	0.080630	0.08775
L5	0.000170	0.056340	0.05651	0.004810	0.110810	0.11562

Note: The bottoms substrate of Karimunjawa waters was fine sandy-coarse substrate type, which have higher organic matter content (see Table 7).

Table 7. The sediment character of Karimunjawa waters, Jepara, Indonesia

Station	Percentage (%)					Total organic matter (%)
	Gravel	Coarse sand	Fine sand	Silt	Clay	
L1	12.57	43.20	40.30	3.93	0.00	2.21
L2	10.35	43.21	42.56	4.05	0.00	2.72
L3	10.92	43.78	41.78	3.85	0.00	2.68
L4	19.03	41.27	36.62	3.08	0.00	6.03
L5	10,98	46.60	35.93	4.20	0.00	3.28

Discussion

There were 6 species seagrasses and 10 species of seaweed found in Karimunjawa waters, *E. acoroides* and *T. hemprichii* may provide shelter and food for *H. atra*. Seagrass serves as diverse habitat for many marine species including fish and many invertebrates, such as sea cucumber (Tuapattinaja et al. 2014). The seagrass beds area did not only serve as primary and secondary production in the coastal ecosystems, they also provide as nursery areas for many fish and invertebrate species. Because it provides protection as well as a source of nutrient that helps to sustain many complex food chains (Rasheed et al. 2006). According to Dissanayake and Stefansson (2012) the highest density of *H. atra* in the northwest coast of Sri Lanka was reported in the seagrass habitat followed by the sandy habitat with rocks/corals, rocky habitat with algae/seagrass. Dense aggregation of *H. atra* in the shallow water seagrass area in Bama beach, Baluran National Park was also observed by Setyastuti (2014). *H. atra* were found mostly in association with seagrass *E. acoroides*, *C. rotundata,* or a mix of both species and small number exposed to the bare sandy substrate with no association. The observed affinity of *H. atra* with seagrass may be related to the sheltering effect of the seagrass canopy and nutritional factor in the bottom of seagrass. Based on Komatsu et al. (2004), the seagrass will trap more nutrient in the baseline and bring benefit to the benthic community including *H. atra*.

Algal epiphytes use seagrass leaves and rhizome as substrate and there were 28 genera of them belong to Division of Chrysophyta, Chlorophyta and Cyanophyta in Karimunjawa waters. The variety and abundance of phytoplankton, zooplankton the benthos enrich the location. The chlorophyll-a, phaeophytin and total chloropigment in sea water and sediment of Karimunjawa could be representative of microalgae and microphytobenthos. Variability in the food supply is a major controlling factor in the population dynamics of benthic animals, in particular, holothurians. Deposit feeders are among the most important consumers of detritus on the ocean floor. According to Dar and Ahmad (2006) *H. atra* consumed large amounts of benthic sediments, they also absorbed particulate materials from the water column and swept the organic biofilm off the top 5 mm of sediment.

The average density of phytoplankton in the area was 744.000 cell.L^{-1} and consisted of 28 genera. Although phytoplankton does not have a direct use for deposit feeder such as *H. atra*, they play very important role in the feeding of holothurian planktonic larva (Hartati et al. 2017). The benthic community of study site consisted of polychaete worms (70%), pelecypods (7%), and crustaceans (23%). Polychaetes are an important component of the macrobenthic community as they often dominate in term of abundance, biomass, and species or taxa. They play an important role in the stability and functioning of the benthic community and the ecological in general (Wardiatno et al. 2017). Deposit-feeding sea cucumbers process large volumes of benthic sediments, from which they assimilate bacterial, fungal and detrital organic matter (Kitano et al. 2003; Slater et al. 2011; Navarro et al. 2013; Yokoyama 2013). Holothurians feed on various organic detritus, and benthic organisms such as bacteria, cyanophycean, and foraminiferans (Wiedemeyer 1992), as sources of dietary lipids, particularly fatty acids. Mfilinge and Tsuchiya (2016) showed that *H. atra* selected algal (bacteria, diatoms, dinoflagellate and green macroalgae) and detrital particles coated with bacteria for food. Since the algal and detrital particle found in present work *H. atra* might select both.

Estimating the concentration of chlorophyll-*a* is the most common method for assessing algal biomass. The concentration of chlorophyll-*a* has also been shown to relate to primary productivity and can be used to assess the physiological health of algae by examining its degradation product, phaeophytin. Chlorophyll-a concentration in sea water of Karimunjawa in the ranged from 0.000161-0.000178 ug.L^{-1} but the concentration of phaeophytin and total chloropigments were from 0.048300-0.000 to 0.048300-0.0761 ug/L respectively. Chlorophyll-a in sediment could be representative of microphytobenthos, i.e. benthic microalgae which are important and resilient ecosystem modifiers and are also important in coastal food webs because of their high accessibility to consumers, such as sea cucumber. The chlorophyll-a, phaeophytin and total chloropigment in the sediment of Karimunjawa waters was 0.001960-0.007120; 0.188770-0.007120, and 0.08775-0.118828 μg.g^{-1} respectively. It may be due to the high abundance of microphytobenthic organisms. Singh et al. (1999) stated the importance of chloropigment concentration in the feeding activity of sea cucumbers. In his experiment, Uticke (1999) also found that *H. atra* significantly reduced the chlorophyll contents of the sediment which was dominated either by diatoms or by cyanobacteria. In present work, the diatom was the most abundance.

The sediment character and total organic matter of the sediment are presented in Table 7. The bottom substrate of Karimunjawa waters was fine sandy-coarse substrate type. Sediment characteristics (i.e., grain size and organic content), biotic relationships and habitat variable such as shelter availability influences the aggregation and distribution of sea cucumber (Conand 1989; Dissanayake and Stefansson 2012; Eriksson et al. 2012). The feeding selectivity of *H. atra* between the coarse sediments and the fine sediments depend on their biogenic needs and food availability (Dar and Ahmad 2006). It was found varied size of *H. atra* in Karimunjawas waters, and they might prefer various sediment particles. The organic matter of sediment was in the range of 4.21-6.03%. According to Moriarty (1982) the detritus (non-living matter) constituted 60-80% of the organic matter in the sediment and thus the food of *H. atra*. As in Aspidochirote holothuroids, *H. atra* are prominent benthic representatives in many littoral ecosystems (Harrold and Pearse 1987; Birkeland 1988), and ingest superficial sediment and feed on non-living detritus and associated microorganisms (Massin and Jangoux 1976; Moriarty 1982; Birkeland 1988; Namukose et al. 2016). Due to their feeding activity, holothuroids must have strong effects on the environment: they are active sediment reworkers that alter the bottom stability (Massin 1982), promote the return of nutritive elements to the water column (Rhoads and Young 1971) and enhance the production of sediment-associated bacteria (Amon and Herdel 1991; Namukose et al. 2016), in turn will be useful for their food.

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