Assessing the food habits of *Crassostrea tulipa* (L. 1819) in a commercial fishery of a Ramsar site in Ghana

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Abstract

The development of wild commercial oyster culture in Ghana is of recent concern. Reduction in feed cost through reliance on natural feed sources is an important sustainable aquaculture management strategy. In many parts of sub-Saharan African countries including Ghana, there is limited documented information on the natural food items fed on by *Crassostrea tulipa* (Lamarck, 1819). The oyster in the Densu estuary was studied for food habits using the Numerical, Frequency of Occurrence, and Index of Relative Importance (IRI) methods and condition index for a duration of 18 months. The diet of the oyster was dominated by golden algae (Phylum Ochrophyta; IRI=595), red algae (Phylum Rhodophyta; IRI=209), green algae (Phylum Chlorophyta; IRI=131.37) and diatoms (Phylum Bacillariophyta; IRI =172). There was a high species diversity (D = 6.60 ± 0.10 - 7.01 ± 0.03; H '= 0.30 ± 0.12 - 0.32 ± 0.05) among the food items ingested by *C. tulipa*. Golden algae was the most abundant (IRI= 56%), followed by red algae (IRI = 16%) and the diatoms (IRI = 13%). The less abundant phyla were Chlorophyta (IRI= 8 %), Cynophyta (IRI = 6 %) and Arthropoda (IRI = 1 %). The oyster was in a good state of wellbeing (Mean Condition index = 139.50 ± 0.11) and feeds on a wide range of plankton species. This information is essential in the development of natural feed for the oyster industry in Densu estuary. The presence of green algae in the diet requires further investigation to help minimize the possible toxic effects it may have on the fishery and humans.

Keywords: Aquaculture, Algae, Estuary, Gut, Filter feeder, Sustainable

Introduction

The West African Mangrove Oyster (WAMO, *Crassostrea tulipa* L. 1819) is widely cultivated in West Africa. In Ghana, the (WAMO) is widely distributed, occurring in mangroves, sediments and compact substrates of coastal water bodies (Obodai, 1999; Ampofo -Yeboah, 2014). According to Yankson and Obodai, (1999), 48 % in number of the estuaries and lagoons in Ghana were found to be suitable for commercial cultivation of oysters suggesting a high potential for their use for aquaculture to augment catches from the wild which the Densu estuary is no exception. Therefore, the mangrove oyster, provides a good opportunity for diversifying Ghana's aquaculture sector. According to Quayle (1989), though feeding behaviour of many species of oysters have been studied globally, little is known about what actually constitutes usable food meanwhile information obtained on one species in an area may not be applicable elsewhere. Furthermore, the fishery remains unregulated and its large-scale culture has not been attempted due to the paucity of relevant scientific information. Feed source and type is an important area for consideration in every aquaculture venture. It is therefore imperative to conduct this study in a quest to contribute to the development of the fishery.

Therefore, the study aimed at the possible provision of reliable documented data on species fed by the oyster and their biotic metrices in the estuarine environment necessary for the sustainability of the fishery.

Materials and Methods

Study Area

The study was conducted in the Densu estuary located between latitudes 5°30¹N and 5°31¹N and longitudes 0°17¹W and 0°18¹W (Figure 1) approximately 11km west of Accra. The Densu estuary takes its source from the River Densu. The study site is a complex open lagoon that is found in the river valley between the Aplaku-Takuse and Weija McCarthy hills. Besides Songor, Keta, Muni near Winneba, and Sakumo lagoon near Tema, the Densu estuary is one of the unique RAMSAR sites in Ghana.

Sampling Design

The study was conducted for a period of 18 months from March 2019 to August 2020. First, following the approach of King (1995), the distribution of the oysters was mapped and measured into an area of 520,000 m² (Figure 1) using Grid tool in QGIS (Remote sensing Laboratory, DMAFS). The area was subsequently divided into a total of fifty-two (52) grids of equal size (100 ×100) m. On each monthly visit, a sub-sample of 10 grids was randomly selected from randomly generated numbers in Microsoft Excel (Version 12) and oysters, collected and measured.

Data Collection for Analysis of Food Habits

Monthly oyster samples were obtained from an artisanal oyster collector who handpicked into a wooden basket throughout the sampling period. Oyster samples were immediately preserved in 10 % formaldehyde solution to reduce post mortem digestion immediately after capture and sent to the laboratory. At the laboratory, morphometric parameters (weight to the nearest 0.01g, length, height and width to the nearest 0.1 cm) of specimens were measured and recorded. Oysters were shucked and the tissues removed using a pair of forceps. The gut were carefully removed and the content emptied into petri dishes. A 10 ml volume of distilled water was added to the content and a sub sample of 5 ml taken and placed under a slide cover, observed and identified under either a dissecting or compound microscope depending on the size of the food item at varying magnifications. Food items were identified up to the genus level wherever possible using identification keys by Shiel (1995) for zooplankton and Bellinger and Sigee (2015) for phytoplankton. Stomach contents were analyzed using the frequency of occurrence and numerical composition methods (Bagenal and Braum,

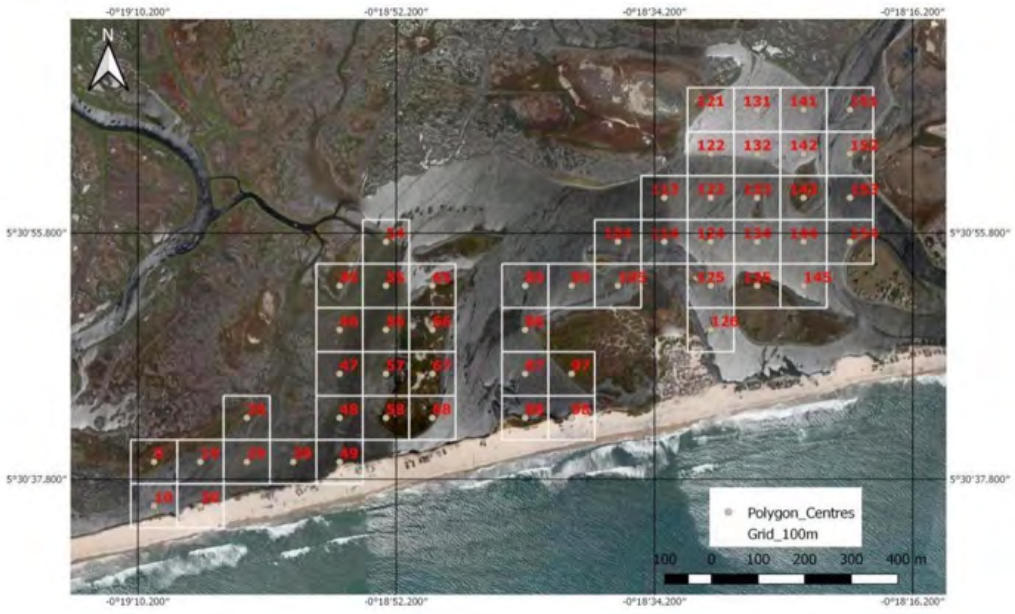

Fig. 1 Map of Densu Estuary showing grids where Sampling of oysters was done

1978; Hyslop, 1980; Lima Junior and Goitein, 2001). The frequency of occurrence method estimates the percentage of stomachs in a sample containing a given food item whereas the numerical composition method determines the total number of a particular food item recorded and expressed as a percentage of total number of all food items:

$$\text{Frequency of Occurrence} = \frac{\text{Total number of stomachs with a particular food item}}{\text{Total number of stomachs with food}} \times 100$$

$$\text{Numerical} = \frac{Total\ number\ of\ \ a\ particular\ food\ item}{Total\ number\ of\ \ food\ items} \times\ 100$$

The measures of frequency of occurrence (F) and number of food items (N) recorded in this study were integrated into an 'index of relative importance' (IRI) as described by Clark (1985) to determine the principal food item of *C. tulipa*:

$$IRI = F \times N \text{ (Clark, 1985)}$$

Where N = Numerical percentage,
F = Frequency of occurrence percentage and
IRI = Index of relative importance.
Checklist of species in the gut content and water medium, was generated from identification of monthly samples. Diversity indices were used to describe and compare the diversity of the species of food items in the estuary and the oyster. These were:
(i) Margalef's Index (D) for species richness (Margalef, 1968),

$$D = S - 1/\ln N$$

where S = number of species and N = number of individuals (2) The Shannon Wiener's
Index (H') of species richness (Shannon and Wiener, 1963),
(ii) The degree of similarity between gut contents and food items in water was determined as,

$$Cs = \frac{2j}{a+b} \text{ (Krebs, 1999)}$$

where Cs is Sorensen's index which ranges from 0 (dissimilar) to 1 (completely similar), *j* is the number of species common to a given pair of stations, and a and b are the number of species occurring in either of the pairs.
(iii) Species richness and equitability were assessed using Shannon -wiener diversity index

$$H' = -\sum Pi \ln Pi$$

where Pi is the proportion of the total number of individuals occurring in species i. Pi = ni/N; ni = Number of individuals of each species in the sample. N = Total number of individuals of all species in the sample. Range from 0 to 5.

Determination of Condition Factor
Oyster samples of not less than 60 individuals were randomly collected monthly from the field and transported to the laboratory for detailed analyses. In the laboratory the weights of individuals were measured to the nearest 0.01g using a weighing balance whereas length and height were taken using a pair of vernier calipers (to the nearest 0.1cm). Diseased and empty shells were excluded. Oysters were shucked to obtain tissues and weight of fresh flesh taken. The total fresh weight: (Pt) was the body weight of the individual after withdrawal of the foreign bodies of the shell; the weight of the fresh flesh (Pch): fresh visceral weight, drained during at least 30 minutes on filter paper and the weight of the empty shells (Pc): weight of the two valves after insulation of the visceral mass.
The weight of wet oyster tissue was taken and the tissue oven dried to constant weights at temperatures of 95-98 °C. Condition factor was determined using the Hopkins' formula (Quayle, 1989)

$$\text{Condition Index} = \frac{(Dry\ tissue\ weight\ in\ g) * 1000}{(Internal\ cavity\ volume\ in\ \text{cm}\,3)} \text{ (Quayle, 1989)}$$

In determining cavity volumes, the relation suggested by Lawrence and Scott (1982) was followed.

$$\text{Cavity volume} = Weight\ of\ \ valve - weight\ of\ \ intact\ oyster\ (both\ in\ g)$$

This method is valid because the effective density of the cavity of the contents is close to 1 g per cm^3 (Lawrence and Scott, 1982).
A condition factor value of up to 150 indicates a high condition while a low value of about 75 and below indicates a very poor condition (Quayle, 1989).

Results

Food Habits

The occurrence and abundance of food items identified in the gut of the West African Mangrove Oyster and in the estuary are presented in Tables 1 and 2 respectively. In Table 1, species occurrence in the gut of the mangrove oyster are shown. A total of 43 species from 8 Phyla and 33 Families were identified in the gut contents of the species. Out of this number, except for the larvae of stonefly and the protozoan, Vorticella sp., all other food items recorded in the study were found in the gut of C. tulipa. (Table 1). All the 43 species were found in the water samples examined for food items (Table 1).
In Table 2, golden algae, comprising five (5) species was the predominant (Occurrence

TABLE 1
Species Occurrence in the Guts of *C. tulipa* and Water of Densu estuary (2019 and 2020)

Family	Food item (phylum/species)	Gut	Water
	Bacillariophyta (Diatom)		
Hemidiscaceae	*Azpeitia neocrenulata*	+	+
	Aspeita sp.	+	+
Achnanthidiaceae	*Achnanthidium minustissmum*	+	+
	Achanthidium sp.	+	+
Fragilaraciaceae	*Aesterionella formosa Hassall*	+	+
Aulacoseiraceae	*Aulacoseira granulate*	+	+
	Aulacoseira sp.	+	+
Cocconeidaceae	*Cocconeis pediculus*	+	+
Tabellariaceae	*Meridion* sp.	+	+
	Unidentified sp.	+	+
Melosiraceae	*Melosira* sp.	+	+
Naviculaceae	*Nanoneis hasleae*	+	+
	Nanoneis sp.	+	+
Bacillariaceae	*Pseudo-nitzschia inflatula*	+	+
Stephanodiscaceae	*Stephanodiscus* sp.	+	+
	Unidentified sp.	+	+
Thalassionemataceae	*Thalassionema bacillare*	+	+
	Thalassionema javanicum	+	+
	Cyanophyta (Blue green algae)		
Nostocaceae	*Anabaena* sp.	+	+
Chroococcaceae	*Anacystis* sp.	+	+
Oscillatoriaceae	*Lyngbya* sp.	+	+
Microcystaceae	*Microcystis viridis*	+	+
	Microcystis sp.	+	+
Microcoleaceae	*Planktotrix agardhiii*	+	+
Oscillatoriaceae	*Oscillatoria* sp.	+	+
	Unidentified sp. 1	+	+
	Unidentified sp. 2	+	+
Rivulariaceae	*Rivularia* sp.	+	+
	Unidentified sp.	+	+
Microleaceae	*Trichodesmium* sp.	+	+

TABLE 1 *cont.*
Species Occurrence in the Guts of *C. tulipa* and Water of Densu estuary (2019 and 2020)

Family	Food item (phylum/species) Bacillariophyta (Diatom)	Gut	Water
	Cyanophyta (Blue green algae) *cont.*		
Chlorellaceae	*Nannochloris sp.*	+	+
Oedogoniaceae	*Oedogonition sp.*	+	+
	Ochrophyta (Golden algae)		
Mallomonadaceae	*Synura petersenii*	+	+
	Synura uvella	+	+
Tabellariaceae	*Tabellaria flocculosa*	+	+
Toxariaceae	*Toxarium undulatum*	+	+
	Rhodophyta (Red algae)		
Solieriaceae	*Agardhiella* sp.	+	+
	Arthropoda		
	Unidentified sp (Amphipod)	+	+
	Unidentified species (stonefly larvae)	–	+
	Ciliophora (protozoan)		
Vorticellidae	*Vorticella* sp.	–	+
	Odonata (Damselfly)		
	Bradinopyga alpogastra	–	+

=35 %, IRI= 595 %) food items filtered by the shellfish. The phylum Rhodophyta comprising one species, was the second abundant (Occurrence =20.9 %, IRI= 209 %, n = 1080) group of food ingested by *C. tulipa*. Green algae occurred in 13.4 % of the gut examined and composed of 9.8% of the food ingested. Out of the 1080 stomachs examined, blue green algae were found in 10.8 % of *C. tulipa* diet making up a composition of about 3.9%. The larvae of amphipods and damselfly made up less than 3% (IRI= 16.20) of the gut contents of the mangrove oyster.

In Fig 2 the abundant taxa examined in surface water (per 500 ml) of Densu were Ochrophyta, Rhodophyta, Bacillariophyta, Chlorophyta and Cyanophyta. They occurred in 32 %, 15.2 %, 17 %, 11 % and10.8 % of sampled water

TABLE 2
Frequency of Occurrence and Numerical Percentages of Gut Contents of *C. tulipa* in Densu estuary

Food item (phylum/species)	Frequency of occurrence (%)	Numerical percentage (%)	IRI (n =1080)
Bacillariophyta (Diatom)	**17.2**	**10**	**172**
Azpeitia neocrenulata			
Achnanthidium minustissmum			
Aesterionella formosa hassall			
Aulacoseira granulata			
Cocconeis pediculus Meridion sp.			
Melosira sp.			
Nanoneis hasleae			
Pseudo-nitzschia inflatula Stephanodiscus sp.			
Thalassionema bacillare			

TABLE 2 *cont.*
Frequency of Occurrence and Numerical Percentages of Gut Contents of *C. tulipa* in Densu estuary

Food item (phylum/species)	Frequency of occurrence (%)	Numerical percentage (%)	IRI (n =1080)
Cyanophyta (Blue green algae)	**10.8**	**3.9**	**42.12**
Anabaena sp.			
Anacystis sp.			
Lyngbya sp.			
Microcystis viridis			
Planktotrix agardhiii			
Oscillatoria sp.			
Rivularia sp.			
Trichodesmium sp.			
Chlorohyta (Blue green algae)	**13.4**	**9.8**	**33.32**
Chlamydomonas caudata			
Coelastrum sp.			
Cylindros permopsis			
Gonatozygon sp.			
Nannochloris sp.			
Oedogonition sp.			
Ochrophyta (Golden algae)	**35**	**17**	**595**
Synura petersenii			
Synura uvella			
Tabellaria flocculosa			
Thalassionema javanicum			
Toxarium undulatum			
Rhodophyta (Red algae)	**20.9**	**10**	**209**
Agardhiella sp.			
Arthropoda	**1.8**	**7.3**	**13.14**
Amphipod			
Odonata (Damselfly)	**0.9**	**4**	**3.6**
Bradinopyga alpogastra			

**n= number of stomachs examined; IRI= Index of Relative Importance*

respectively. The damselflies were the least abundant group (IRI= 4) (Figure 2).

Fig 3, gives further illustration on the IRI of all groups of food items fed on by *C. tulipa*). While the least group was the Odonata (IRI=0 %), the most important groups were the Ochropyhyta (IRI = 56 %), Rhodophyta (IRI =16 %) and Bacillariophyta (IRI = 13 %) and

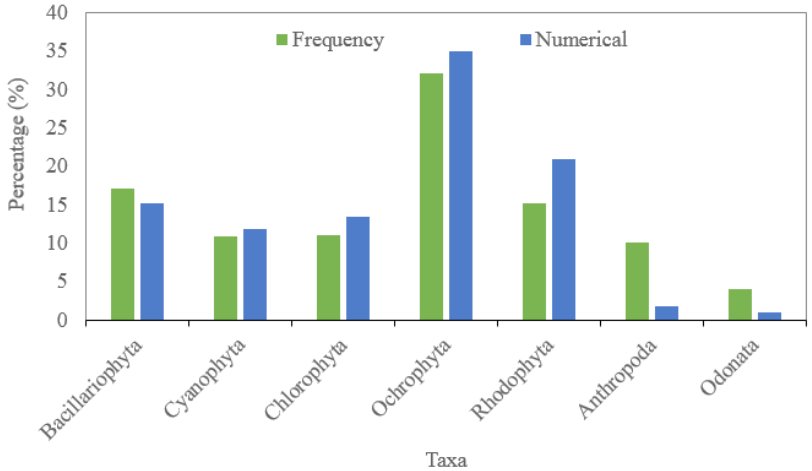

Fig. 2 Percentage Abundance and Occurrence of Food 1tems In Water of Densu estuary

the remaining were distributed among the Chlorophyta (IRI= 8 %), Cynophyta (IRI = 6 %) and Arthropoda (IRI = 1 %).

Species Richness, Diversity, Evenness and Similarity

The number of species, genera, families, species richness, diversity and evenness for the plankton and insect communities recorded in the stomach contents and water samples of the Densu estuary are presented in Table 3.

Where

CF denotes Condition Factor;

D =Species Richness;

H'= Species Diversity;

J = Evenness

Cs= similarity

Among the 43 species belonging to 33 genera and 33 families identified in the gut and water samples of the estuary, species diversity was not significantly different ($p < 0.05$) between gut contents ($H = 0.30 \pm 0.12$) and contents in water medium ($H= 0.32 \pm 0.05$) ($t = 2.16$, $p < 0.05$) (Table 3). Similarly, 40 species were found in the gut and 43 in water samples implying higher number of species in water than in the gut. A Mann Whitney U - test performed on results obtained from Margalef's index analysis indicated no significant difference in species richness between the food items in gut ($D = 6.60 \pm 0.10$) and water ($D = 7.01 \pm 0.03$) ($t = 0.15$, $p > 0.05$). Individuals were evenly distributed among species in the gut ($J' = 0.60$) and water

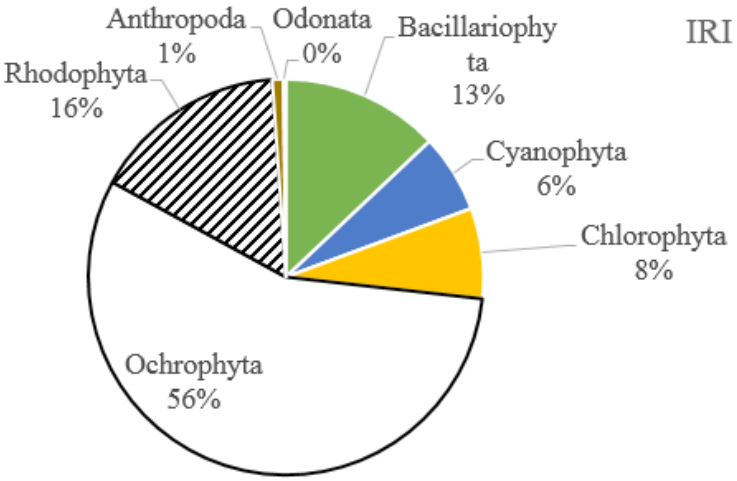

Fig. 3 1ndex of Relat1ve 1mportance of Food 1tems 1n Water of Densu estuary

TABLE 3

Diversity Indices of food items in the gut of *C. tulipa* and water in Densu estuary

Biotic index	Gut	Water
No. of Phyla	7	8
No. of Families	31	33
No. of Genera	31	33
No. of species	40	43
D	6.60 ± 0.10	7.01 ± 0.03
H'	0.30 ± 0.12	0.32 ± 0.05
J'	0.60 ± 0.02	0.68 ± 0.15
CF	139.50 ± 0.11	
Cs	0.970	

(J' = 0.68) communities (Table 3).

Similarity of Food Items in Gut and Water

Most of the species found were represented in the gut and water of the estuary (Table 3). In comparing similarity between the two, a Sorensen's similarity index value of 0.970 was obtained suggesting a high similarity among food items found in the gut and surface water of the Densu estuary.

General wellbeing of the oyster

The mean monthly (CI) of *C. tulipa* in Densu was 139.50 ± 0.11 (Table 3).

Discussion

The predominance of the diet of *C. tulipa* in Densu by golden algae (IRI=595), red algae (IRI=209), green algae (IRI= 131.37) and diatoms (IRI =172) is similar to reports on other species of the genera *Crassostrea*. The research of Dupuy et al. (2016) in France and Kasim and Mukai (2009) in Japan showed that lagoon population of *C. gigas* preferred micro phytoplanktonic benthic diatoms.

However, the preponderance of the golden and red algae in gut of *C. tulipa* and the water column implies their possible abundance and proliferation in the Densu estuary estuary. Conversely, the Densu mangrove oyster ingested less of the blue green algae, arthropods, protozoans and damselflies which is similar to that reported on the lagoon pearl oyster population in France by Loret (2000). In Densu, a high similarity was observed among food items found in the gut and surface water of the Densu estuary which did not conform with the findings of Kasim and Mukai (2009). The authors stated no significant correlations between diet items in gut of the oyster and water column (Kasim and Mukai, 2009). Also, in the study of Densu population, species diversity and richness were not significantly different between contents in the gut and water medium with individuals evenly distributed among species in the gut (J' = 0.60) and water (J' = 0.68) communities (Table 2). However, more species were found in the water column than that found in the gut. Kasim and Mukai (2009) encountered low algal species diversity and abundance in water column of the Akkheshi-Ko estuary in Japan in comparison with species found in the gut which is dissimilar to this report on the Densu estuary in Ghana.

However, in the study of Kassim and Mukai (2009), some groups of algae (diatoms) were found concurrently in high concentrations in the gut and water column. Rouillon et al. (2005) also identified abundance of phytoplankton (dinoflagellates) in the stomach contents than the water samples. Conclusively, probably the pattern of continual presence of some taxa of food items in both water and gut of *C. tulipa* in Densu may be an indication of their abundance in the water column and benthic regions of the system. In literature, a suit of factors for example cell size, ontogeny, capture rate, selection and consumption influences feeding habits among wild bivalve oysters (Metian et al., 2020; Rosa and Padilla, 2020).

In furtherance to that, the authors explained that bivalves do not feed what is filtered but that they have the ability to sort the cleared particles and reject them prior to ingestion in the form of pseudofaeces, depending on various factors such as the concentration of particles filtered from suspension, the surface properties of the trapped particles, low nutritional value, or particle chemical properties (Metian et al., 2020; Rosa and Padilla, 2020) Another study by Galimany (2020) established in an experiment that filtered and cleared algae species in diet of *Crassostrea* were also assimilated by the bivalves. Therefore, the Densu population may not be ingesting high quantities of diatoms, golden and red algae but rather probably selected, consumed and retained these preferred groups. Low retention of food particles has been reported by Loret (2000) where the research found low counts of cyanobacteria in the gut of pearl oyster. The presence of some species of Cyanobacteria is of potential danger to the fishery. This is because some species of cynaobacteria produce toxins which may bio-accumulate in

C. tulipa posing threats to aquatic and human life.

Conclusions

The WAMO was in a good condition during the study indicating possible favourable conditions for the growth and survival of the species. The mangrove oyster feeds on a wide range of plankton species and few individuals of amphipods and damselfly species which categorises it as a planktivore omnivore. This implies in the development of the oyster culture industry in the Densu estuary, feeding cost could be minimized by relying on feed sources from the wild and supplementing with artificial feeds. Therefore, this information is important for development of feed types for aquaculture purposes among stakeholders.

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