

# Main Diatom Taxa in the Natural Diet of Juvenile *Haliotis fulgens* and *H. corrugata* (Mollusca: Gastropoda) in Bahía Tortugas and Bahía Asunción, B. C. S., México<sup>1</sup>

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**Abstract:** To determine the main diatom taxa found in the natural diet of *Haliotis fulgens* Philippi (green abalone) and *H. corrugata* Wood (pink abalone) juvenile specimens and loose rocks were collected from different depths at two sites in Bahía Tortugas and Bahía Asunción, Baja California Sur. Overall 113 benthic diatom species were identified, and 98 taxa were recorded in the gut contents of both abalone species. Out of these 32 were not observed in the surrounding flora. Most diatom taxa were epiphytic forms, including the abundant taxa found in the gut contents of young abalone: *Berkeleya fennica*, *Cocconeis speciosa*, *Cocconeis costata* var. *pacifica*, *Gomphonemopsis pseudexigua*, *Grammatophora marina*, *Navicula parva*, *Tabularia investiens*, and *Thalassionema nitzschioides*. This somewhat reflected the structure of the diatom assemblages in the rocky habitat: few abundant taxa and many rare or uncommon ones, with heterogeneous values of diversity ( $H'$ ) that ranged between 1.1 and 4.2 in the gut contents and from 2.83 to 3.91 in the surrounding flora. New records for the area include *Cocconeis pseudomarginata*, *C. maxima* (common), *Licmosoma* sp., *Ardissonia formosa* (rare), and the colonial form *Berkeleya fennica* (abundant), all found within the gut contents of abalone. The actual and potential diet of young abalone on the coast of Baja California Sur is extended to 235 taxa. The abundant taxa occurred frequently in clumps of numerous individuals in the gut contents, often still attached to algal tissue. The results of this investigation suggest that the importance of diatoms in the diet of larger juvenile abalone merits reassessment.

BENTHIC DIATOMS ARE the primary diet of abalone (*Haliotis* spp.) during their early stages of development (Ebert and Houk

1984, Hahn 1989). Abalone farming has relied heavily on empirical methods, successfully growing young abalone on diatom films that develop from benthic forms occurring in the water column (tychoplankton). Most research on the subject has focused on the feeding habits of postlarvae under culture conditions (Ebert and Houk 1984, Hahn 1989, Salas Garza and Searcy Bernal 1992, Kawamura et al. 1998, Martínez Ponce and Searcy Bernal 1998, Roberts et al. 1999, Siqueiros Beltrones and Voltolina 2000, Siqueiros Beltrones 2002a). The few floristic observations on these diatom films show that many are epipelagic diatoms suspended from the sediments, whereas in the advanced stages of succession epiphytic forms eventually appear in the microalgal films (Siqueiros Beltrones 2002a). But abalone dwell in rocky environments, and when feeding diatoms to postlarvae or young abalone epilithic forms

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would seem to be the logical choice (Siqueiros Beltrones 2002a,b).

Information on the diatom flora found in abalone habitats is scarce. The few in situ studies available have noted the importance only of certain diatoms (*Cocconeis* spp.) in the diet of abalone postlarvae in rocky substrata, particularly on coralline algae (Daume et al. 1997, 1999, Kawamura et al. 1998). Also, the main diet of young abalone has been observed to change from diatoms to macroalgae at around 5–10 mm (Ebert and Houk 1984, Shepherd and Cannon 1988, Hahn 1989, Mazón-Suástegui et al. 1992, Kawamura et al. 1998).

More recent observations have shown that large juvenile (>40 mm) and adult abalone that graze on or upon fleshy and coralline algae also ingest abundant epiphytic diatoms (Siqueiros Beltrones and Valenzuela Romero 2001), suggesting that epiphytic diatoms replace epilithic forms in the diet of abalone juveniles when these grow over 10 mm. Based on these observations it was expected that abundant diatoms would be found accompanying the macroalgae ingested by these young abalone, under the premise that diatoms still play an important role in their diet. However, during a study on the macroalgal diet of *Haliotis fulgens* Philippi and *H. corrugata* Wood from Bahía Tortugas and Bahía Asunción the search for a diatom component in fresh samples of the stomach contents proved negative (Guzmán del Prío et al. 2003). This prompted observations of new samples, as well as of samples from the former study.

Our objective was to determine the species composition of the diatoms ingested by young *H. fulgens* and *H. corrugata* (<50 mm) and those numerically more important. Accordingly, we tested two hypotheses: (1) that these diatom assemblages consist mainly of epiphytic forms, because most of the rocky substratum is occupied by various macroalgae species; and (2) that the diet of these abalone consists of several important taxa and many uncommon taxa, which are ingested according to their availability, much as the benthic diatom assemblages are structured in terms of their proportional abundances.

#### MATERIALS AND METHODS

The biological material used for determining the diatom component in the diet of *H. fulgens* (green abalone) and *H. corrugata* (pink abalone) juveniles was sampled mainly at Bahía Tortugas at the same sites (Clam Bay and Morros) sampled by Guzmán del Prío et al. (2003) and in Bahía Asunción. Both are important abalone fishing localities on the western coast of the Baja California Peninsula (Figure 1). Seven juvenile specimens of *H. fulgens* and five of *H. corrugata* were collected from 5 to 10 m depth, along with four rocks (0.5–2 kg) similarly covered with coralline algae, during June 2002. Some abalone specimens were attached to these rocks, and the others were collected close by on large rocks. Abalone specimens were dissected to extract their gut contents. Diatoms attached to the rock surface and corallines were brushed off using a toothbrush, which was then rinsed with tap water. Also, fixed material from the original samples analyzed by Guzmán del Prío et al. (2003) was retrieved from storage for closer inspection. This material comprised gut contents from several specimens of *H. fulgens* and *H. corrugata*. Gut contents from two specimens of each species were closely inspected.

All new samples were observed fresh under the microscope. Afterward, these and the samples from Guzmán del Prío et al. (2003) were treated with a mixture of nitric acid and commercial alcohol (ethanol) according to Siqueiros Beltrones (2002b). The clean diatom frustules were mounted on permanent slides using Meltmount (R.I. = 1.7). The slides were inspected under 1,000× with phase-contrast microscopy for identification of taxa. This was accomplished using the works by Schmidt et al. (1874–1959), Peragallo and Peragallo (1897–1908), Cleve-Euler (1968), Foged (1975, 1984), Hendey (1964), Hustedt (1959, 1961–1966), Moreno et al. (1996), Romero and Rivera (1996), Siqueiros Beltrones (2002b), and Witkowski et al. (2000). The classification of Round et al. (1990) was followed.

The relative abundances of the surveyed diatom taxa in the gut contents and in the

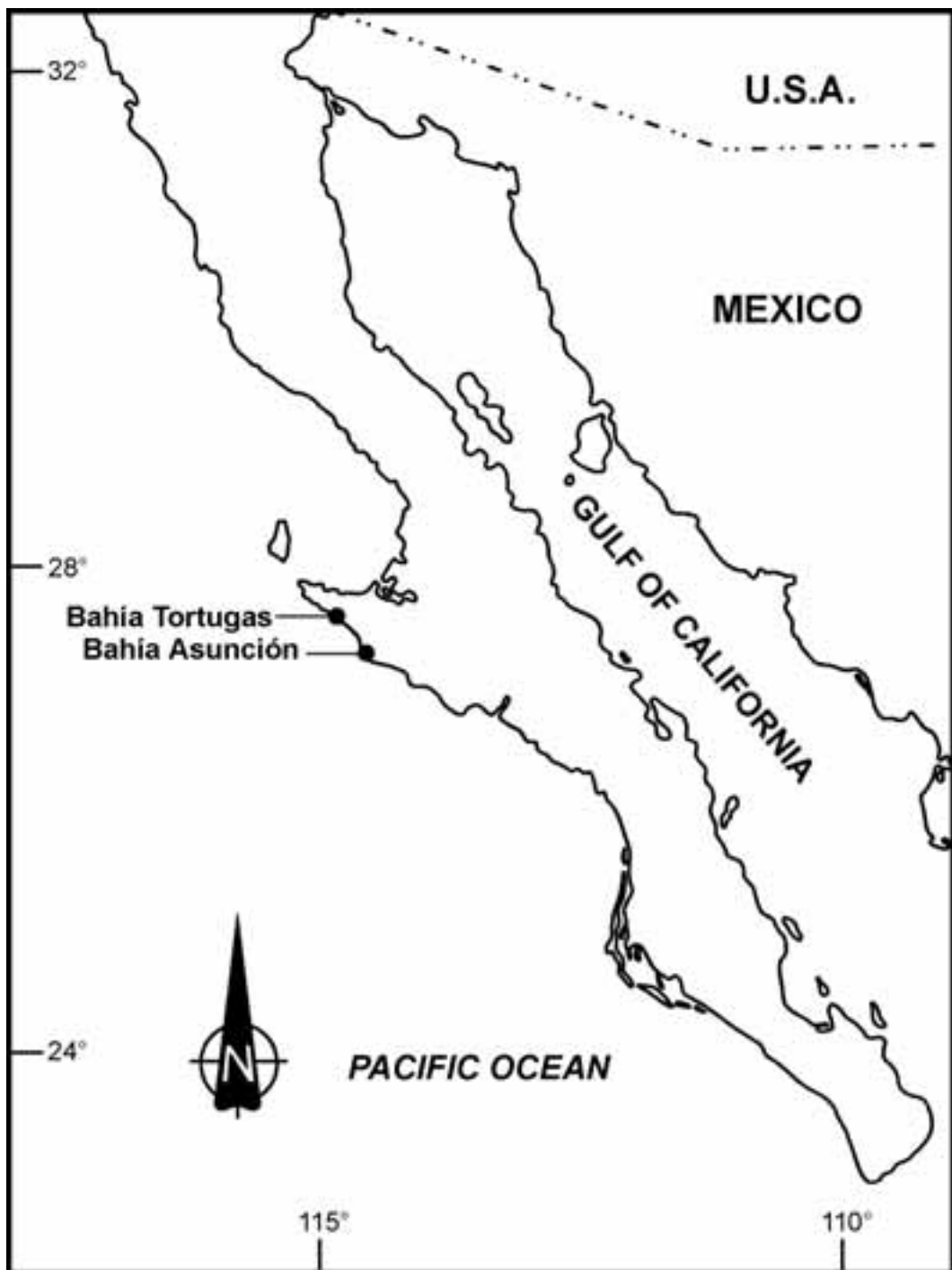


FIGURE 1. Location of sampling sites in the Baja California Peninsula, México.

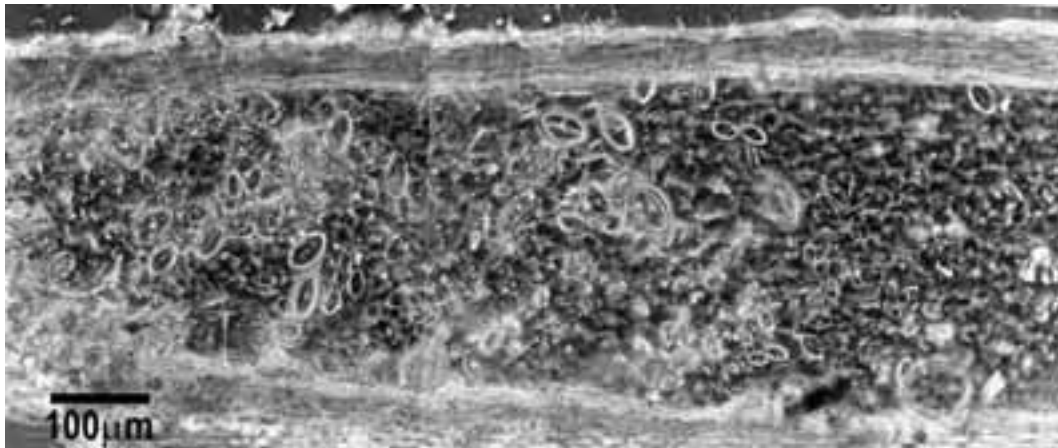


FIGURE 2. Contents of a section of a gut of a young *Haliotis* sp. (<40 mm), showing abundant *Cocconeis diminuta* Pantockzec and larger forms, cf. *Cocconeis scutellum* Ehrenberg and *C. dirupta*.

surrounding flora were determined based on a minimum number of 500 valves (McIntire and Overton 1971, Siqueiros Beltrones 2002b). These were used to estimate the parameters that served to describe the structure of the assemblages based on information theory ( $\log_2$ ), such as diversity ( $H'$ ), Pielou's evenness ( $J'$ ), and dominance (REDI), as in Brower et al. (1998). Similarity between samples was measured with the indices of Jaccard (presence/absence of species) and Bray-Curtis which also considers relative abundances of the shared taxa. Computation for samples of the surrounding flora included a repetition.

#### RESULTS

In the fresh material examined many diatom forms were observed, both from the gut contents and in the samples from the surrounding flora, but not in the samples from Guzmán del Prío et al. (2003). Few could be identified to genus level (e.g., *Cocconeis* spp.). However, the cleaned material from all samples revealed many more diatoms, all identifiable.

Overall 113 diatom species were identified in the surrounding flora and abalone gut samples from Bahía Tortugas and Bahía Asunción. Within the gut contents of both

abalone species, 98 benthic diatom taxa were recorded. In accordance with our first hypothesis most taxa were epiphytic forms, and as proposed in our second hypothesis, the distribution of the relative abundances showed few abundant species and many uncommon or rare taxa, both in the gut contents and in the surrounding flora.

Most diatoms in the gut contents (Figure 2), as in the surrounding flora, were epiphytic forms. The represented taxa were frequently observed forming aggregations of numerous individuals, either in monospecific or multispecific clumps (Figure 3a,b) or still attached to macroalgal tissue, both in the gut contents and in the surrounding flora. The diatom taxa shown in Figures 4 to 10 may be considered representative of the main diatom component in the diet of the abalone we examined. The most abundant taxa in the gut contents were *Berkeleya fennica* Juhlin-Dannfelt, *Cocconeis costata* var. *pacifica* Grunow, *Cocconeis diminuta* Pantockzec, *Cocconeis scutellum* Ehrenberg, *Cocconeis speciosa* Gregory, *Gomphonemopsis pseudexigua* (Simonsen) Medlin, *Grammatophora marina* (Lyngbye) Kützling, *Navicula parva* (Meneghini) A. Cleve, *Tabularia investiens* Wm. Smith, and *Thalassionema nitzschioides* Grunow.

According to the cumulative relative abundances ( $n = 500$ ), 37 taxa constituted 95%

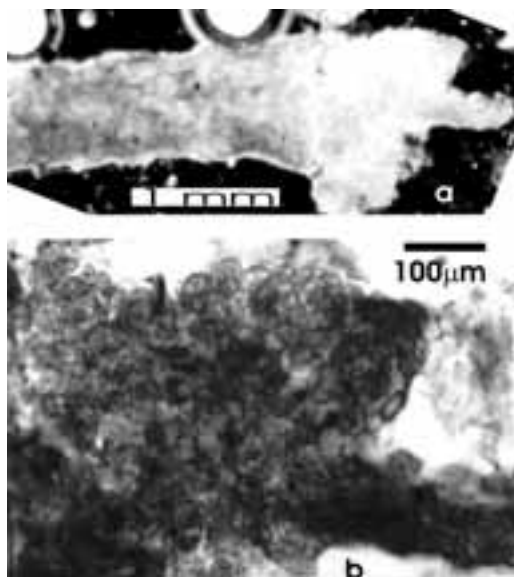


FIGURE 3. a, Clump of diatoms in the gut contents of *Haliotis corrugata* from Bahía Tortugas. b, Close-up of diatom clump showing *Cocconeis scutellum* Ehrenberg.

of the total ( $n = 7,375$ ) quantified diatoms (Table 1). Many taxa were common in one or two samples and were absent in the others. In compliance with our second hypothesis, the structure of the samples varies, although in a different manner in each sample, causing differential variation in the diversity and similarity estimations. New records for the area included *Cocconeis pseudomarginata* Gregory, large forms such as *C. maxima* (Grunow) Peragallo, *Licmosoma* sp., *Ardissonia formosa* (Hantsch) Grunow, and the colonial *B. fennica*. All were found in the gut contents of abalone.

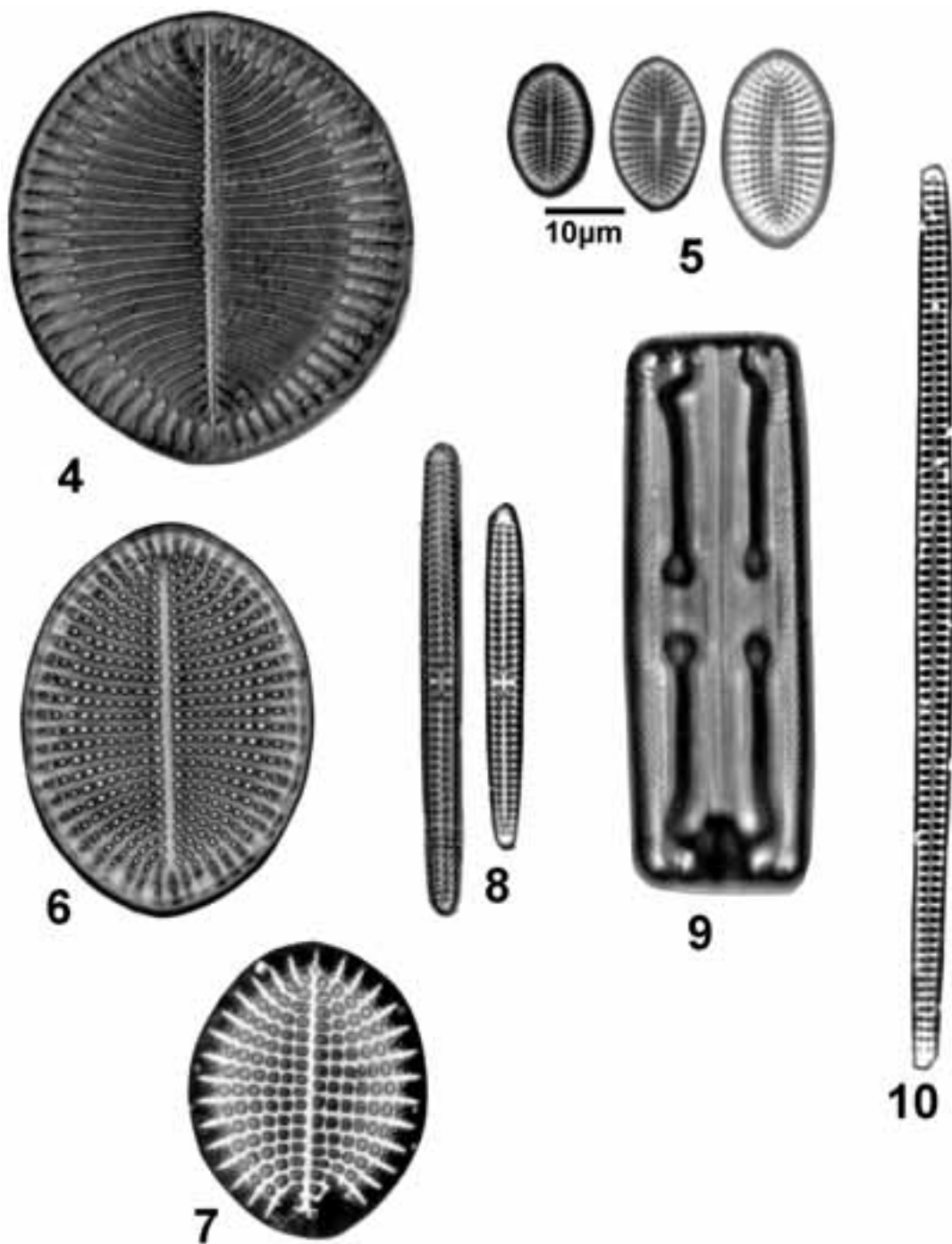
Thirty-two taxa present in the gut contents were not observed in the brushed-off material, and 15 from the surrounding flora were not observed in the gut contents. All these are mainly rare taxa, except for *B. fennica*, which was abundant in the gut contents of a *H. corrugata* specimen from Bahía Asunción but was practically absent in all the other samples. On the other hand, several common taxa from the surrounding flora (*Achnanthes groenlandica* (Cleve) Grunow, *Campylopyxis*

*garkeana* (Grunow) Medlin, *C. scutellum*, and *G. marina*) appeared with very low frequency in the gut contents. Other epiphytes such as *Gephyria media* Arnott and *Porosira* Jørgensen sp. that were abundant on the red alga *Placodium cartilagineum* (L.) Dixon (common in the area) were present but not common in the gut contents.

Based on the similarity and grouping technique using solely presence-absence of species, a low similarity was measured (i.e., <50%) in all comparisons, suggesting randomness, both in the distribution of diatom taxa on the substrate and in the diet of the abalone. On the other hand, according to the Bray-Curtis index (Figure 11), higher similarity values were caused by the shared presence of the more abundant species, mainly *C. speciosa* (Table 1), between apparently unrelated samples, such as Hf 8 versus Hc 16, whereas high dissimilarity was measured between related samples (D and E versus Hf 4). Thus, high values between related samples may not be considered as indicative of an actual similarity between the abalone diets and the available diatom flora.

Diversity of gut contents was higher for *H. corrugata* than for *H. fulgens*, because of both higher species richness and evenness (Table 2). These correspond somewhat with the values estimated for the potential diet. Some values of diversity, however, reflect the influence of a single abundant species, causing high values of dominance (REDI) and low evenness ( $J'$ ), either with a high species richness ( $S = 40-42$ ) or coupled with a low  $S$  that determined the lowest values of diversity in *H. corrugata* (minimum  $H' = 1.1$ ,  $S = 27$ ,  $J' = 0.230$ ). On the other hand, a high diversity (maximum  $H' = 4.42$ ) reflected a high species richness but mainly high evenness ( $S = 47$ ,  $J' = 0.796$ ) and low dominance (REDI = 0.245) for the same taxon.

After removal of most organic matter, the mounted samples from Guzmán del Próo et al. (2003) showed abundant diatoms, including several compact clumps with many frustules that remained even after oxidation, similar to those in Figures 2 and 3. The species composition and relative abundances were similar to the ones described earlier.



FIGURES 4–10. 4, *Cocconeis costata* var. *pacifica* Grunow; 5, *Cocconeis diminuta* Pantockzec; 6, *Cocconeis scutellum* Ehrenberg; 7, *Cocconeis speciosa* Gregory; 8, *Gomphonemopsis pseudexigua* (Simonsen) Medlin; 9, *Grammatophora marina* (Lyngbye) Kützing; 10, *Tabularia investiens* Wm. Smith.

TABLE 1

Abundance Distribution of Benthic Diatoms Making Up 95% of Cumulative Abundances in Gut Contents and Surrounding Flora at Bahía Tortugas and Bahía Asunción, B. C. S., México (Juvenile Abalone *Haliotis corrugata* and *H. fulgens* Ranged from 8 to 48 mm)

Taxa	Gut Contents			Total
	<i>H. corrugata</i> (n = 5)	<i>H. fulgens</i> (n = 5)	Rock Samples (n = 6)	
1. <i>Cocconeis speciosa</i>	727	725	659	2,111
2. <i>C. costata</i> var. <i>pacifica</i>	414	42	240	696
3. <i>Grammatophora marina</i>	147	7	418	572
4. <i>Tabularia investiens</i>	94	58	411	563
5. <i>Thalassionema nitzschioides</i>	103	320	83	506
6. <i>Gomphonemopsis pseudexigua</i>	104	307	10	421
7. <i>Navicula parva</i>	59	331	28	418
8. <i>Cocconeis scutellum</i>	7	4	350	361
9. <i>Berkeleya femica</i>	282	0	0	282
10. <i>Achnanthes groenlandica</i>	4	36	115	155
11. <i>Campylopyxis garkeana</i>	8	1	143	152
12. <i>Cocconeis dirupta</i>	8	25	81	114
13. <i>Gomphoseptatum aestuarii</i>	45	45	9	99
14. <i>Thalassiosira</i> sp. 1	85	0	2	87
15. <i>Cocconeis pseudomarginata</i>	0	71	15	86
16. <i>Cocconeis diminuta</i>	9	46	25	80
17. <i>Grammatophora hamulifera</i>	35	0	44	79
18. <i>Bacillaria socialis</i>	36	3	34	73
19. <i>Rhoicosphenia adolphii</i>	59	6	7	72
20. <i>Navicula directa</i>	32	5	35	72
21. <i>Ampbora pediculus</i>	13	6	32	51
22. <i>Cocconeis dirupta</i> small form	10	0	38	48
23. <i>Cocconeis dirupta</i> var. <i>flexella</i>	0	0	48	48
24. <i>Cocconeis costata</i> var. <i>hexagona</i>	15	5	27	47
25. <i>Cocconeis guttata</i>	10	1	34	45
26. <i>Hyalodiscus scoticus</i>	28	1	9	38
27. <i>Navicula</i> sp. 1	15	1	12	28
28. <i>Podosira stelliger</i>	3	0	23	26
29. <i>Actinopteryx</i> cf. <i>minutus</i>	21	0	4	25
30. <i>Cocconeis maxima</i>	1	21	3	25
31. <i>Paralia sulcata</i>	10	0	14	24
32. <i>Nitzschia dissipata</i>	5	3	14	22
33. <i>Rhoicosphenia marina</i>	12	7	0	19
34. <i>Thalassiosira</i> cf. <i>eccentrica</i>	16	1	2	19
35. <i>Trachyneis aspera</i>	3	4	12	19
36. <i>Raphoneis amphiceros</i>	15	0	3	18
37. <i>Fallacia oculata</i>	16	0	0	16
Nt				7,375

## DISCUSSION

Our observations strongly suggest that juveniles of *H. corrugata* and *H. fulgens* may feed on whatever diatoms are available in their environment. More than 230 diatom taxa may be available as potential diet items, as has been observed recently in a similar nearby environment (Siqueiros Beltrones and Valen-

zuela Romero 2004), including those from our study, where at least 98 taxa were observed as forming part of the *H. corrugata* and *H. fulgens* diet. However, the main benthic diatoms in the diet of *H. fulgens* and *H. corrugata* juveniles during this time of year in Bahía Tortugas and Bahía Asunción are few, specifically *Cocconeis speciosa*, *C. costata* var. *pacifica*, *C. diminuta*, *C. scutellum*, *Gomphone-*

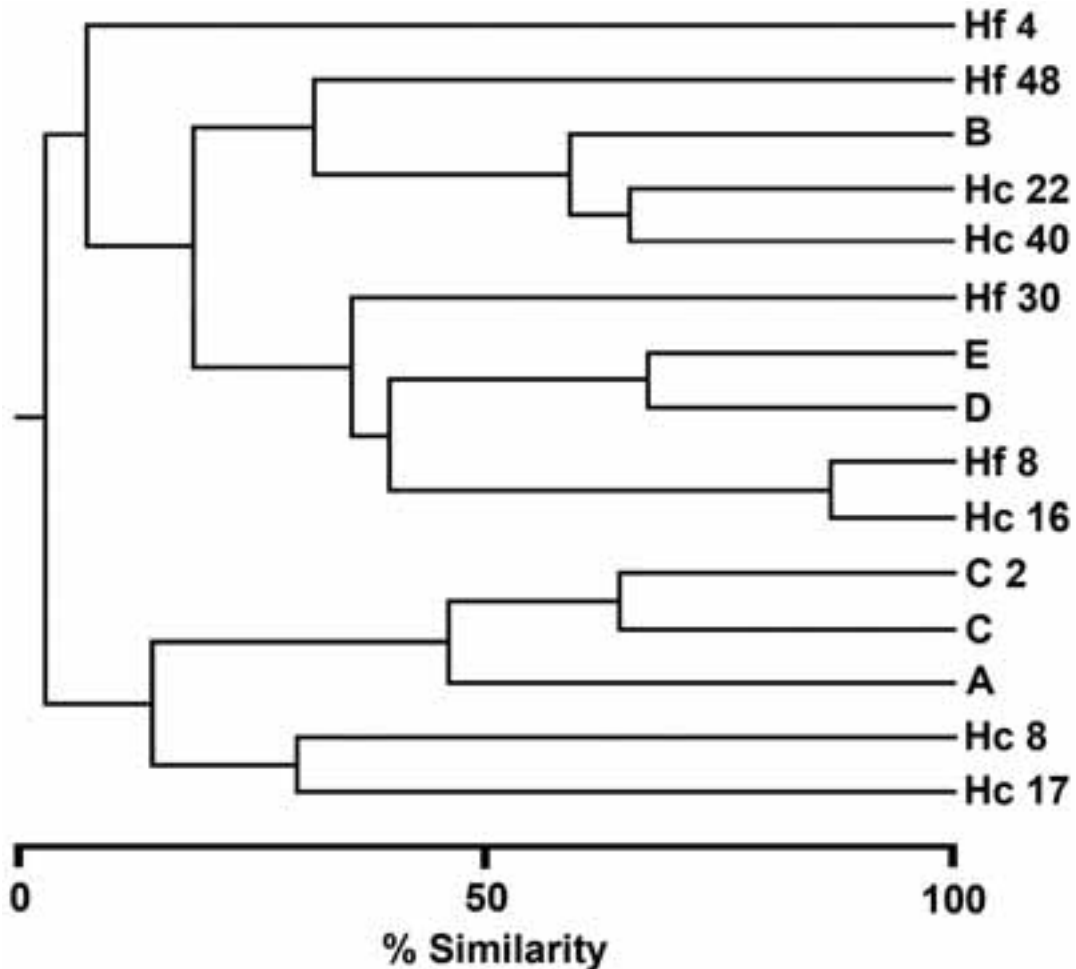


FIGURE 11. Similarity (Bray-Curtis) between the diatom taxa in the gut contents of *Haliotis corrugata* (Hc) and of *Haliotis fulgens* (Hf). Numbers = size of specimen (in mm); A, B, C, C2, D, E = surrounding flora samples and respective repetitions (rocks).

*mopsis pseudexigua*, *T. nitzschioides*, *T. investiens*, and the colonial forms *B. fennica* and *N. parva*.

Because several distinct substrata are available (rock, fleshy and coralline macroalgae), some heterogeneity occurs in the available diatom flora, which explains the observed differences in diversity and similarity values. The structure of the assemblages, although similar to the expected typical pattern, differed from sample to sample. This indicates that the abalone diet varies depending on the

diatom patches on which an abalone eventually feeds. We assume that this is shown also in the composition of the diatom diet in a particular period. Thus, more comprehensive sampling is required to detect patterns or preferences in the diet of juvenile abalone throughout the year. That is, diatoms are ingested according to their distribution, which is influenced by the available substrata, represented strongly by the macroalgal flora.

Thus, because most of the rocky substratum is covered by fleshy or coralline algae,



TABLE 2

Estimated Values of Diversity for Diatom Assemblages from Bahía Tortugas (Clam Bay [CBay] and Morros) and Bahía Asunción (Asun.) and in Gut Contents of *Haliotis corrugata* (Hc) and *H. fulgens* (Hf)

Sample <sup>a</sup>	S	H'	J'	R
CBay A	37	3.91	0.751	0.290
CBay B	46	3.65	0.662	0.399
Morros C	31	3.64	0.734	0.304
Morros C2	25	3.36	0.724	0.310
Morros D	28	3.12	0.649	0.397
Morros E	28	2.83	0.588	0.461
Hc40 CBay	40	3.06	0.574	0.499
Hc22 CBay	32	3.00	0.600	0.452
Hf48 CBay	28	2.41	0.501	0.564
Hf4 Morros	14	2.14	0.563	0.468
Hf30 Morros	18	1.37	0.329	0.730
Hf8 Morros	24	1.36	0.299	0.778
Hc16 Morros	27	1.10	0.230	0.868
Hc17 Asun.	42	2.95	0.546	0.537
Hc8 Asun.	47	4.42	0.796	0.245
Total	113			

Note: S, species richness; H', Shannon's diversity; J', Pielou's evenness; R, redundancy.

<sup>a</sup> A, B; C, C2; D, E = rock (surrounding flora) samples and repetitions; number following Hc and Hf is size of specimen (in mm).

the diatoms found in the guts of the abalone were mainly epiphytic. As many as 56 species of macroalgae have been recorded for this area (Serviere Zaragoza et al. 2003). Several of these species are known to be ingested by green abalone (McBride 1998, Serviere Zaragoza et al. 1998) and pink abalone (Guzmán del Próo et al. 2003), and epiphytic diatoms are ingested along with the tissue of their host.

In accordance with our hypothesis few important taxa constitute the main diet of young abalone, much as the benthic diatom assemblages are structured in nature. Plus, all are epiphytic forms. Some of the more frequently found taxa such as *C. speciosa* and *G. pseudexigua* were recently recorded (for the first time) growing abundantly on blades of *Macrocystis pyrifera* (L.) C. Agardh (Siqueiros Beltrones et al. 2002), a macroalga that is a preferred food of abalone and is commonly used to feed abalone under culture conditions (McBride 1998). Other diatoms recorded here for the first time were *Cocconeis britannica*

Naegeli ex Kützinger, *C. maxima*, *C. pseudomarginata*, *Licmosoma* sp., and *Ardissonia formosa*, which appeared both in the surrounding flora and in the gut contents. These extend the actual and potential diet of abalone to 235 taxa.

Although this study was directed toward larger abalone (4–48 mm), it does have some implications regarding postlarvae. Several efforts have used the monospecific (unialgal) approach when searching for their optimal diet (Martínez Ponce and Searcy Bernal 1998, Roberts et al. 1999, Correa Reyes et al. 2001). Although it seems fitting to consider diatom taxa that characterize their potential or actual diet in situ when choosing the best diet for abalone during their different stages of development, only *Cocconeis scutellum* var. *parva* and *C. pseudomarginata* (Kawamura et al. 1998) have been considered for feeding abalone postlarvae. And only *Cocconeis scutellum* has been recorded as part of the natural diet of *H. rubra* postlarvae in a similar environment (i.e., rocky substratum covered by coralline algae [Daume et al. 1997, 1999, Roberts et al. 1999]). However, in our samples the frequency of this species was not as high as that of other taxa, although several aggregations of this taxon were observed in both type of samples. The suggestion of any kind of selective feeding in the wild by abalone postlarvae (Daume et al. 1997, 1999) should also be considered; in vitro observations (Kawamura et al. 1995, Mathews and Cook 1995, Roberts et al. 1999, Siqueiros Beltrones and Voltolina 2000) may be supportive of this.

Many diatoms in the gut contents of the young abalone that we examined appeared in clumps, probably because these and colonial forms are ingested whole along with algal tissue, resisting digestion in the abalone digestive tract, and because of the cleaning technique as well. This may cause an underestimation when assessing their contribution to the abalone diet. This observation helped us to explain why no diatoms were observed in the fresh samples used by Guzmán del Próo et al. (2003), although numerous diatoms were indeed present. We thus recommend that abalone gut or stomach contents

be observed both fresh and cleaned (oxidized) when identifying diatoms.

It is widely accepted that the main diet of young abalone changes from diatoms to macroalgae at around 5–10 mm (Ebert and Houk 1984, Shepherd and Cannon 1988, Hahn 1989, Mazón-Suástegui et al. 1992, Kawamura et al. 1998), and most literature on larger juvenile abalone completely omits any reference to diatoms (S. A. Shepherd, pers. comm.). However, our observations indicate that, even in these larger abalone, diatoms are still present in an important quantity, and their proportional importance requires specific assessment. Furthermore, current investigation (by D.S.-B.) may identify more diatom taxa located in various substrata within the abalone rocky environment and in different seasons. The information provided here may yield valuable insight into the diatom-abalone trophic relationship, including possible selective feeding in both species of abalone.

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