A PRELIMINARY INVESTIGATION OF THE STRUCTURE OF DIATOM COMMUNITIES ASSOCIATED WITH
THE REEF HABITATS OF THE FLORIDA KEYS

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ABSTRACT

The dynamics of energy pathways within coral reef communities have come under increasing investigation. However, few qualitative or quantitative studies of the micro-populations of non-planktonic diatom floras associated with reef substrates have been done. The structure of such communities associated with coral reefs in the Florida Keys is analyzed and compared.

Substrate samples representing the coral sand bottom, the different zones of the reef structure, and the adjacent grass beds were collected along similar transects at Molasses, Sombrero, and Western Sambo Reefs on the open ocean side of the Florida Keys. The numbers of genera, species, and the species diversity of the diatom floras from these habitats are compared. High densities of diatoms were found associated with all substrates, and the number of species observed were found to increase with the degree of structural complexity of the habitat. Comparison of affinity measurements of all samples indicated a high degree of structural similarity between diatom communities from similar substrates. Transect sites of coral substrate from Molasses Reef are demonstrated to be unique and characterized by diatom floras intermediate between the coral substrates of the other reefs and coral sand samples. It is suggested that as a result of continual replenishment of nutrients by bacterial populations associated with reef surfaces, attached diatom populations may not be nutrient limited, as are the planktonic phytoplankton populations in reef waters.

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Introduction

The association of plants and animals com-
prising coral reef communities have long intrigued
investigators. In studying the dynamics of energy
pathways within reef communities researchers have
often noted an apparent paradox between the rel-
atively low levels of primary productivity in
surface waters near reefs and the dense and com-
plex life of reef communities (1). It has been
suggested that photosynthesis within the reef
community itself provides the answer to this
seeming contradiction (2,3). Rates of net carbon
fixation from 4 to 12 g/m2/day have been reported
for many reef communities (4). Yonge (5) was the
first to suggest that corals were largely depen-
dent upon photosynthesis by their associated zoo-
xanthellae for their basic metabolic requirements,
and much work has been done demonstrating the
movement of photosynthetic materials from the zoo-
xanthellae into the coral tissues (6,7). However,
while corals often physically dominate a reef
community, Odum and Odum (2) and Webb et al. (4)
have stated that other sources of primary produc-
tivity may be much more important to the total
energy budget of the reef community. Odum and
Odum (2), in their classic study of the trophic
structure of Eniwetak Atoll, suggested that a
majority of the primary production observed was,
in fact, due to photosynthesis both by the
coenocytic, filamentous green algae Ostrobium
living within the surface layers of the coral
skeleton and by the myrid of red, brown, green,
and blue-green algae embedded in and encrusted on
living and dead reef substrates. Recent studies,
however, have tended to discount the importance
of Ostrobium as a significant source of primary
production in most coral reef systems (8).

It has become popular to view the highly
localized productivity of reef communities to be
in large part a result of the recycling of nutri-
ents in such a manner that they become "trapped"
within the community (9). DiSalvo (10) and Sorokin
(11) have stressed the important role bacterial
populations play in the production and recycling
of nutrients in such pathways. Corals are typ-
ically covered by a mucous layer which is contin-
ually sloghed off as it becomes embedded with
microorganisms and particulate matter. A number
of authors have stressed the possible importance
of mucous-orginated detrital material covered with
bacteria as an energy source for reef zooplankton
and filter feeding organisms (12,13). In addition,
a number of hermatypic corals have been shown (15)
to actively function as suspension feeders, captur-
ing fine particulate material by means of mucous
filaments. Sorokin (11) has demonstrated the abil-
ity of several scleractinian corals to filter and

utilize planktonic bacteria from the water column.
However, as important as the micro-fauna and flora
of reefs appear to be to the total energy dynamics
of these communities, very few qualitative or
quantitative studies of micro-populations have
been done (10). This is especially true for
studies of the presence and structure of non-
planktonic diatom floras associated with reef
substrates.

The low levels of phytoplankton in the waters
surrounding reef communities has been well estab-
lished (13). However, as early as 1907, Karsten
(16), as part of the "Valdivia" Expedition, noted
a sudden change in the phytoplankton composition
from oceanic to neritic species as coral atolls
were approached. Similarly, studies of the plank-
ton over Australian reefs (17,18) and at Eniwetak
Atoll (19) have shown the presence of a large num-
ber of tychopelagic pennate diatoms representing
bottom-living forms which had been churned up into
the water column. Odum and Odum (2) found that
glass slides placed in front of the reef developed,
within 21 days, communities of small diatoms (<5
microns) as high as 5.3x10^6/cm2. DiSalvo (10),
likewise, noted the presence of a great diversity
of pennate diatoms on regenerative sediments in
Kaneoke Bay, Hawaii. However, there are no com-
prehensive studies of the structure and taxonomy
of the diatom communities living upon the various
substrates within a reef community. In this paper,
the structures of such communities associated with
corals of the Florida Keys are analysed and com-
pared, with taxonomy to be discussed in a second
paper (20).

Materials and Methods

Transects

Collection stations were previously estab-
lished at Molasses, Sombrero, and Western Sambo
reefs on the open ocean side of the Florida Keys
between Key Largo and Key West (Figure 1). These
reefs were selected as representative of the var-
ying conditions of the reefs found along the
Florida Keys reef tract. At the collection site
on Molasses reef, the northernmost reef in our
study, as little as 25% of the reef structure was
comprised of living corals. At the southernmost
station, Western Sambo reef, approximately 75% of
the reef surface was covered with live corals,
while at the intermediate station, Sombrero reef,
the ratio of live to dead coral was close to 50%.

The reefs at each station were of the spur-
groove formation typical to the Florida Keys. In
August 1975, substrate samples representing the
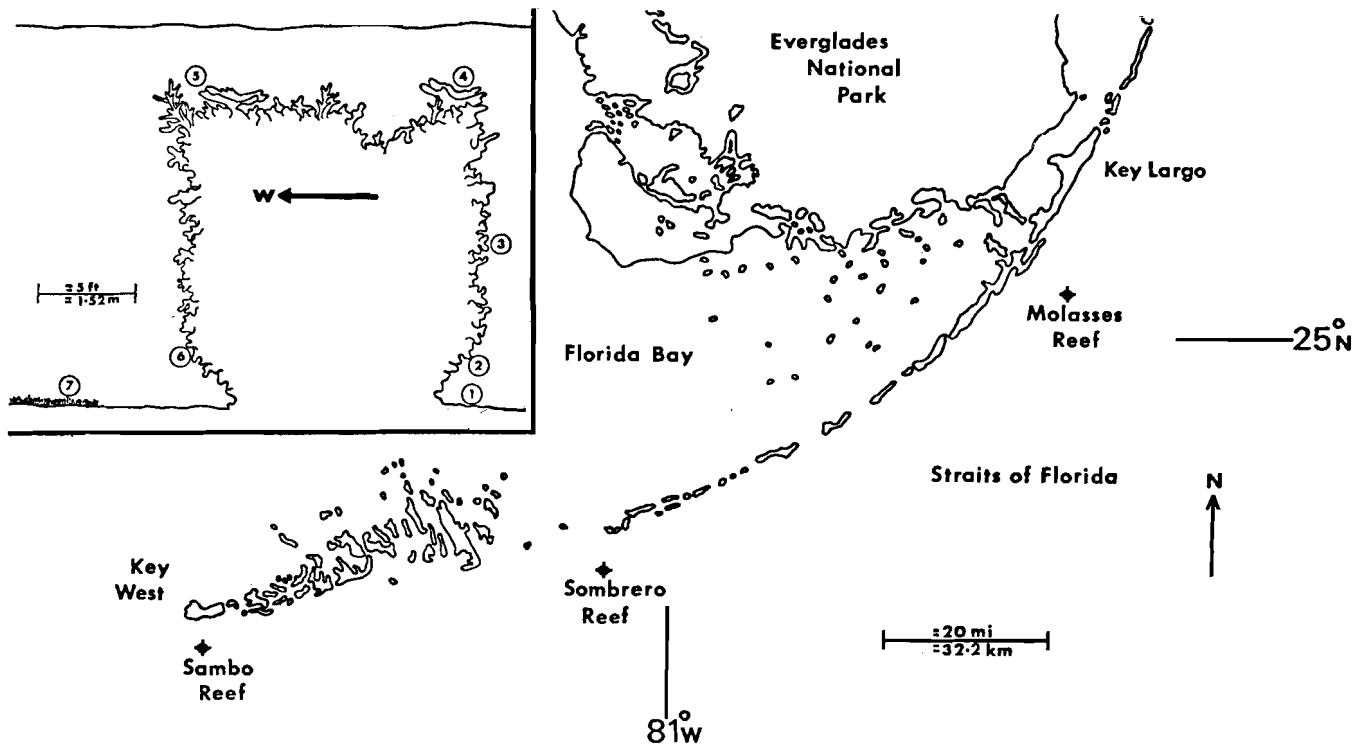coral sand bottom, the different zones of the reef

Figure 1. The map shows the locations of the reef stations at which samples were collected. The insert depicts a cross section of a reef spur formation, showing the positions of the transect sites.

structure, and the adjacent grass beds were collected at sites along similar transects at each reef station (Insert Figure 1). At the first site, 100 grams (dry wt.) of coral sand from the upper 1 cm along the base of the reef were collected. Since this study was concerned only with variations in the diatom community structure of different zones of the reef habitat, sites 2 through 6 were comprised of random coral samples rather than being of any one species. Characteristically, natural populations of attached organisms have been found to be clustered in mosaic patterns rather than randomly distributed (21). For this reason, 10 to 12 small pieces of coral with a total surface area of approximately 400 sq cm were removed from the reef substrate at each site. Of the 400 sq cm collected, 75% were comprised of living material and 25% of dead coral. Both sand and coral samples were preserved for later preparation by placing them in a drying oven at 100° C for 12 hrs. The final sample at each station was collected from the beds of Thalassia testudinum (turtle grass) located on the lee side of each reef. The encrusting material from grass blades, with a total surface area of 400 sq cm, was carefully removed using a scalpel, and preserved in 100 ml of a 10% ethyl alcohol-fresh water solution.

Preparation and Enumeration of Samples

The diatom flora was removed from the sand and coral by placing the samples from each site in a 1500 ml beaker with 200 ml of concentrated hydrochloric acid. Five ml of acetone were added to reduce foaming. When the surface layer of the coral pieces had been totally dissolved the pieces were removed from the solution, rinsed with distilled water, and discarded. For the coral samples, the sand particles were permitted to dissolve totally. In preparing material from the surface of the turtle grass, 25 ml of hydrochloric acid were added to the preserved sample. The solutions were then permitted to stand for 1 hr. to ensure that all calcium carbonate had completely dissolved. Next, 200 ml of distilled water were added to reduce viscosity, and the samples were centrifuged on a Series 1 International Centrifuge at 500 rpms for 30 minutes in a number of 15 ml tubes such that after centrifugation no more than 1 ml of organic material accumulated at the bottom of each tube. This organic material was then rinsed and recentrifuged at least six times to ensure the complete removal of all calcium in solution before the addition of sulfuric acid in the next step.

The systematic classification and identification of diatoms is based solely on the

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characteristic morphology of their siliceous cell walls. Therefore it was necessary to remove all organic material from the samples both to concentrate the diatoms and to allow for their accurate identification. Organic material was totally oxidized from the samples utilizing a modification of the sulphuric acid/potassium permanganate/oxalic acid method described by Hasle and Fryxell (22). Due to the very high levels of organic material in these samples it was necessary to repeat the first two steps of this process at least twice. Subsequent to cleaning, portions of the cleaned samples were very slowly dried on coverslips. A single drop of alchol was placed on the coverslips to reduce surface tension and promote the even distribution of diatom frustuals. Four replicate slides from each sample were prepared for counting using Hyrax (R.I. 1.71) mounting medium.

The number of diatom cells needed to be counted in order to accurately determine community structure has been widely debated (23). For these studies, curves of the diversity and total number of species observed versus the number of individuals counted were plotted to establish the sample size at which these curves became asymptotic. In addition, calculations of affinity for samples of various sizes were compared. By the use of these methods, a sample size of 1500 cells was determined sufficient for gaining maximum information of species composition and structure, while holding the sample size to a level which could reasonably be counted.

Analysis of Data

Diversity indices have been widely used to describe structural changes in phytoplankton communities (24), and the advantages of the various indices have been thoroughly reviewed (25). Recently the use of such measurements has come under increasing criticism because of the excesses to which some have gone in using them. However, if these indices are viewed merely as convenient methods for condensing complex data sets into simple descriptive statistics weighting both the number of species and the distribution of individuals among those species, then they become valuable population parameters. As with any other parameter, such indices should neither be overvalued nor used alone. For these investigations, two widely used diversity indices and their corresponding evenness measurements were calculated. The Shannon-Weiner index is generally more sensitive than the Simpson measurement, the latter giving greater weight to the more common species.

Shannon-Weiner $H' = -C \Sigma Pi \log Pi$ * (1)

Evenness $J' = H' / \log S$ * (2)

Simpson $SI = 1 / \Sigma Pi$ * (3)

Evenness $SE = SI / (1/S)$ * (4)

In addition, $C\lambda$, which is a measurement of relative similarity between samples (26) was calculated for all possible sample pairs.

$$C\lambda = 2 P1i P2i / (\Sigma P1i^2 + \Sigma P2i^2) \quad * (5)$$

$C\lambda$ is derived from Simpson's diversity index and is conceptually unambiguous in that similarly to a probability or a correlation coefficient, its value varies from 0 to 1.0; zero representing total dissimilarity and one perfect identity.

* S = total number of species
Pi = the proportion the ith species represents of the total number of individuals in the sample (P1i indicates the first sample).

Results

In Table 1 the results of the sample counts for each site are summarized. The following patterns can be observed in this table.

1. On both sand and turtle grass substrates the total number of genera and species observed are fewer than on corresponding reef substrates.
2. The proportion of species comprising greater than 1% of the population is greater on both the sand and turtle grass habitats. Thus, reef substrates are characterized by greater proportions of rare species.
3. At each station the transects show a decrease in the number of diatoms per unit area as the top of the reef is approached.
4. With the exception of the sand substrate, corresponding sites at Western Sambo reef have fewer diatoms per unit area than either of the other two reefs.
5. Both the Shannon-Weiner and the Simpson diversity indices and their evenness measurements demonstrate similar patterns.
   a) The structure of the diatom floras of the sand habitats are characterized by high diversity and evenness.
   b) Diatom communities on the surface of turtle grass show both low diversity and evenness measurements.
   c) In comparing different zones at sites on the reef stations, both diversity and evenness increase towards the top of Molasses reef, show no pattern at Sombrero, and decrease towards the top of Western Sambo.
   d) Similar substrates at different reef stations are characterized by similar indices of diversity and evenness.
   e) At each station measurements at sites (three and four) in the upper reef zone are more variable than corresponding measurements at sites (two and six) at the base of the reef.

Table 2 is an ordered matrix of the distributional similarity measurement $C\lambda$. This value corresponds to an overall estimation of the similarity between two populations. Unordered matrices of such similarity measurements, though

Reef Site	Number of Genera	No. Gen. > 1% of Population	Number of Species	No. Sp. > 1% of Population	Cell Density x10^4	H' Base e	J'	SI	SE
Molasses 1	23	12	197	30	32.80	4.49	.851	53.44	.986
Molasses 2	33	14	211	19	1.46	4.30	.803	27.77	.969
Molasses 3	41	16	237	24	3.09	4.65	.850	50.27	.984
Molasses 4	39	16	240	23	1.42	4.71	.860	62.81	.988
Molasses 5	34	11	200	22	1.10	4.28	.807	33.21	.975
Molasses 6	35	14	198	20	7.42	4.22	.798	22.96	.961
Molasses 7	28	11	140	25	2.79	3.74	.756	21.97	.961
Sombrero 1	30	10	221	30	38.10	4.59	.850	56.39	.987
Sombrero 2	39	15	226	23	5.22	4.53	.836	41.49	.980
Sombrero 3	37	13	219	25	2.28	4.57	.848	51.01	.985
Sombrero 4	39	13	213	22	.98	4.39	.819	38.41	.979
Sombrero 5	38	14	217	26	1.43	4.49	.835	45.07	.982
Sombrero 6	36	11	217	21	7.61	4.45	.827	41.65	.981
Sombrero 7	24	10	138	24	1.25	3.73	.757	21.74	.961
Sambo 1	23	9	174	33	76.77	4.39	.850	50.05	.986
Sambo 2	37	12	220	22	2.02	4.43	.821	35.47	.976
Sambo 3	32	11	206	27	.40	4.19	.787	30.19	.972
Sambo 4	33	11	196	31	.55	4.30	.815	29.71	.971
Sambo 5	31	12	178	18	.13	3.90	.753	18.82	.952
Sambo 6	35	15	228	26	2.80	4.60	.848	52.72	.985
Sambo 7	23	11	125	21	.78	3.62	.751	19.81	.957

Table 1. Summary of diatom counts for all stations. For each reef station, transect sites are coded by number as in the insert Figure 1. Cell densities are per cm^2 of surface area, with the exception of the three sand samples for which density values are per gram dry wt. H' = Shannon-Weiner diversity index, J' = evenness Shannon-Weiner, SI = Simpson diversity index, SE = evenness Simpson

useful, are often confusing. In developing this table an unordered matrix was rearranged by the following criteria. The site with the lowest affinity (Cλ value) to all other sites was placed at the top of the Y axis. Below this was placed the site with the highest affinity to the first site. Ranked in the third position was the site with the highest affinity to the second site, not already listed. This proceedure was followed until all sites had been listed in a comparative ordination along the Y axis. The matrix was completed by reversing the ranked order along the X axis. By this method table 2 was constructed in such a manner that affinity values generally increase both from top to bottom and from left to right. This results in the clustering of areas of high similarity separated by areas of low affinity along the diagonal margin. The following groupings of diatom communities are, thus, depicted in Table 2.

I. Group one shows the high similarity of species composition among all the sand samples.
II. The second group indicates that sites comprised of coral substrate from Molasses reef are unique and characterized by diatom floras intermediate between the coral substrates of the other reefs and the coral sands.
III. The third large group reveals a high degree of similarity between the diatom communities

on coral substrate samples at Sombrero and Western Sambo reefs.
IV. This last group indicates that diatoms living on the surface of turtle grass are generally unique and characteristic of that habitat.

Discussion

Pomeroy (27) has suggested that many of the major pathways of energy flow in marine systems may be directed by the little studied populations of micro-organisms. Organic aggregates derived from muscus, bacteria and diatoms are possibly a major nutritional source for the zooplankton and the filter feeding fauna of reef communities (1, 13). In addition many conspicuous reef species are herbivors adapted to scraping encrusting material from reef substrates. In this study, dense populations of diatoms were found to be attached to these substrates. It is possible that, as a result of the continual replenishment of nutrients by bacterial populations associated with such surfaces (10,11), attached diatom populations may not be nutrient limited, as are the planktonic phytoplankton in reef waters. In addition, actively growing diatoms have been shown to release a significant proportion of their photoassimilated carbon as disolved organics (28). Such compounds may be of significant importance when considering the energy pathways of reef communities (11).

	Sa7	So7	Mo7	Sa5	So5	Sa3	So3	So6	Sa4	So4	Sa6	Sa2	So2	Mo4	Mo3	Mo5	Mo6	Mo2	Sa1	So1
Mo1	.04	.04	.05	.11	.17	.23	.23	.23	.19	.16	.24	.20	.22	.18	.31	.26	.38	.38	.80	.91
So1	.08	.08	.10	.19	.23	.26	.28	.29	.23	.22	.30	.27	.28	.25	.38	.31	.39	.43	.82	
Sa1	.05	.06	.06	.11	.16	.21	.20	.22	.17	.15	.21	.18	.19	.17	.27	.23	.33	.34		
Mo2	.31	.30	.37	.51	.52	.46	.50	.50	.51	.54	.52	.62	.61	.52	.74	.82	.92			
Mo6	.24	.24	.30	.35	.40	.41	.45	.45	.45	.43	.43	.50	.52	.40	.64	.77				
Mo5	.45	.45	.55	.57	.68	.67	.70	.73	.79	.76	.71	.80	.82	.63	.79					
Mo3	.30	.33	.43	.56	.73	.61	.79	.78	.70	.74	.77	.80	.88	.80						
Mo4	.27	.30	.37	.53	.72	.51	.69	.64	.56	.70	.72	.73	.77							
So2	.34	.39	.50	.61	.76	.73	.86	.90	.84	.86	.85	.90								
Sa2	.32	.38	.48	.73	.78	.76	.81	.78	.86	.87	.87									
Sa6	.32	.34	.42	.58	.78	.69	.79	.79	.79	.81										
So4	.35	.39	.49	.67	.73	.69	.76	.80	.81											
Sa4	.36	.45	.55	.53	.67	.76	.79	.83												
So6	.36	.41	.49	.45	.71	.76	.88													
So3	.31	.41	.46	.44	.72	.81														
Sa3	.27	.35	.42	.56	.61															
So5	.44	.44	.48	.55																
Sa5	.28	.23	.31																	
Mo7	.72	.95																		
So7	.73																			
Sa7																				

Table 2. Ordered matrix of Cλ values. Matrix values are coded by reef station (Mo = Molasses, So = Sombrero, Sa = Sambo) and site (1 - 7 as depicted in the insert Figure 1). Groupings of affinities of substrates with similar diatom floras are apparent along the diagnal margin.

The results demonstrate marked differences in species composition and community structure of attached diatom populations on various reef substrates. Among the types of substrates studied the number of species observed increased with the degree of structural complexity of the habitat. The opposite patterns of zonation shown by the population diversity indices at Molasses Reef (25% living coral) and Western Sambo Reef (75% living coral) seem to indicate that zonation of diatom communities over these reefs is dependent not only on physical gradients but also upon biological interaction with the coral substrate. This conclusion is further supported by the measurements of affinity. These indicate that the species composition of the diatom flora of Molasses Reef is characteristically different from that of either of the other two reefs, which are covered by greater percentages of living coral substrate.

The structure and species composition of these attached diatom communities can be viewed at any given time as a reflection of both favorable and unfavorable biotic and abiotic conditions. For these reasons, measurements of diatom flora have long been used as indicators of pollution in freshwater systems (29). It is suggested that this may also be possible for reef communities. Continued studies of seasonal variations of diatom floras along these reefs and investigations into the effects of area of substrate, complexity of substrate, period of colonization, and coral species upon diatom community structure are presently being completed.

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