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Morphological Characterization of *Halimeda* (Lamouroux) from Different Biotopes on the Red Sea Coral Reefs of Egypt

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Abstract: The genus *Halimeda* (Lamouroux) was examined on the basis of collection made from nine locations during 1999-2008 along the Egyptian Red Sea coral reefs with emphasis on the effect of illumination at different biotopes on the morphology of the species. Five species were identified: *Halimeda monile* (Ellis and Solander) Lamouroux, *H. macroloba* Decaisne, *H. opuntia* (L) Lamouroux, *H. discoidea* Decaisne and *H. tuna* (Ellis and Solander) Lamouroux. These species are described in detail with original illustrations and an identification key. As for *Halimeda monile* and *H. macroloba* showed no marked changes in relation to light conditions; *H. opuntia* showed marked difference in branching pattern and segment shape; whereas *H. discoidea* and *H. tuna* exhibited very similar gross morphology that makes difficulties in distinguishing some specimens. Nodal structure seems to vary less at different biotopes and, therefore, is more useful in species identification. Southern area of the Egyptian Red Sea coast favored the growth of all species while, only *H. opuntia* and *H. tuna* were found in the northern area and this may point out for conservation of the reef communities.

Key words: Egypt · red sea · caulerpales · *Halimeda*

INTRODUCTION

The genus *Halimeda* Lamouroux is widespread in tropical and subtropical waters and is often well developed on coral reefs [1]. Their thalli are characterized by a series of green segments impregnated with calcium carbonate as aragonite [2] and are joined together by small un-calcified nodes into variously branching chains. Unconsolidated carbonate sediments provided by *Halimeda* at present and also have been in the geological past, quantitatively more important to building reefs than is the carbonate incorporated into the framework organisms such as corals and crustose coralline red algae [3].

Halimeda is being represented by 33 species and currently placed, according to [1], in order Caulerpales of Chlorophyceae. Many taxonomical studies have been dealt with the distinctive characters of species. Hillis-Colinvaux [1] divided the genus into five sections based on the patterns of nodal structure. Hillis *et al.* [4] and Kooistra *et al*. [5]; based on a partial sequence of the rDNA, found a good correlation between several morphological characters and the molecular data.

Seven species of *Halimeda* were reported by [6] from the whole area of Red Sea. A general knowledge of the habitats of only three of these species can be derived from the works of [7-10]. However their circumscription is not always clear and varies with the author.

This study deals with the morphological characterization and distribution of the genus *Halimeda* in the Egyptian Red Sea coasts. Such information is required for conservation of reef communities and resolving the confusion in identification in local publications. Particular emphasis has been placed on determining the effects of habitat conditions on the morphology of the species. An identification key, original descriptions and illustrations have been also considered.

MATERIALS AND METHODS

Halimeda was collected from nine major sites on the reefs situated along the Egyptian coast of Red Sea from (30.00 N, 32.25 E) to (22.39 N, 36.51 E). The individual sites were Suez, Ein el-sukhna (60 Km south of Suez), Ras Gharib, Ghardaqa, Safaga, Koseir, Zabargad and Shalateen-Halaib coast with it's off shore islets.

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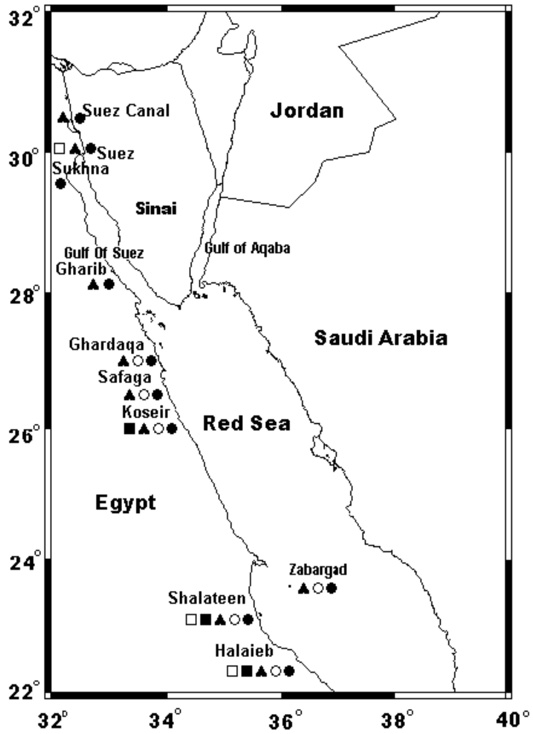

Fig. 1: Geographic distribution of *Halimeda* species on the Egyptian coasts of Red Sea. Legends: *H. monile* (open quadrate □); *H. macroloba* (blind quadrate ■); *H. opuntia* (blind triangle ▲); *H. discoidea* (open circle ○); *H. tuna* (blind circle ●)

Their locations are shown in Fig. 1. Specimens from the Suez Canal were also included. Collections were made from shallow to 25 m depth of subtidal habitats on reef flat, lagoon, windward and leeward reef slope within each site.

Botanical and ecological information had been accumulated in various sites during the period of 1999-2008. Algal materials were preserved as herbaria. Records of site, depth, habitat types and the general ecological conditions are included with all voucher specimens. For light microscopy, specimens were preserved in 4% Formalin and decalcified in 20% HCl. Dried herbarium specimens were immersed in 50% glycerol for 24 hours and then processed as described for liquid preserved specimens. Longitudinal and cross sections were made with steel blades. Nodal regions were dissected with needles and the preparations were mounted in a 50% glycerol solution. All the drawings are original and made by the author using Camera Lucida and an Olympus 1X70 microscope (Olympus, Tokyo, Japan).

RESULTS

Thallus of *Halimeda* consists of calcified segments separated by more or less flexible little calcified nodes. In Red Sea, its growth form is erect or pendant, achieving a height of 6-33 cm and anchoring to unconsolidated substrata by a single small or large holdfast of matted filaments. Coenocytic filaments, called siphons, make up the thallus. In each segment, siphons branch, cohere, or fuse to form a multiaxial core known as medulla and, then branched and inflated near the surface of segment to form a cortex of 2-5 layers of utricles. The utricles stick together to form unbroken surface. Their cell walls are calcified. Nodes consist only of medullary siphons, which may fuse in pairs, in small units, or into single unit.

Five species of *Halimeda* were found from shallow to 25 m below low water on the coral reefs of Red Sea. Their distribution is shown in Fig. 1 and their morphological and anatomical characteristics are assembled in Table 1 and illustrated in Fig. 2-7. The following is the key that could be used for identification of these species.

Key for the identification of *Halimeda* species reported from Red Sea

1. Medullary siphons at node thick-walled, fused into a single unit with a series of pores; holdfast large and bulbous 2

1. Medullary siphons at node thin walled, fused in pairs or three for a distance of node; holdfast small and felt-like 3

2. Segments mainly cylindrical, moderately calcified; nodal pores more than 20 µm; secondary utricles moderately inflated *H. monile*

2. Segments mainly flat-fans, moderately calcified; nodal pores less than 20 µm; secondary utricles moderately inflated *H. macroloba*

3. Nodal fusion more than 100 µm; segments large, light to moderately calcified, discoid or reniform; utricles hexagonal or polygonal in surface view 4

3. Nodal fusion less than 100 µm; segments small, substantially calcified, mainly heart-shaped with prominent central rib *H. opuntia*

4. Segments mainly discoid, lightly calcified, green when dried, up to 33 mm broad; secondary utricles swollen *H. discoidea*

Table1: Morphological and anatomical characteristics of species. Note the variability of gross morphology between the specimens from shallow and deep water

Character	Note	*H.monile*	*H.macroloba*	*H.opuntia*	*H.discoidea*	*H.Tuna*
Calcification	Shallow	Moderate	Moderate	Substantial	Light	Moderate
	Deep	Light	Light	Less	Light	Light
Color	Shallow	Whitish	Green	Whitish	Green	Light green
	Deep	Green	Light green	Light green	Light green	Whitish
Branching	Shallow	Dense	Dense	Dense	Dense	Dense
	Deep	Loose	Loose	Loose	Loose	Loose
Holdfast		Large bulbous	Large Bulbous	Small felt-like	Small felt-like	Small felt-like
Basal segment		Cylindrical	Cylindrical	Cylindrical	Cylindrical	Cylindrical
Segment shape	Shallow	Cylindrical	Fan	Heart	Discoid	Reniform
	Deep	Fan	Reniform	Reniform	Reniform	Discoid
Segment size (in mm)	Shallow	7W, 8(13)L	24 W, 18 L	12 W, 7 L	18 W, 15 L	14 W, 10 L
	Deep	6 W, 8 L	14 W, 10 L	9 W, 7 L	33 W, 21 L	23 W, 13 L
Surface view of segment		Large hexagonal	Small hexagonal	Small irregular	Large hexagonal	Large polygonal
2ry utricles	Shallow	Elongated	Elongated	Elongated	Subglobular	Elongated
Nodal siphons		Thick walled, fuse into single set open to neighbors by large pores more than 20 µm	Thick walled, fuse into single set open to neighbors by small pores less than 20 µm	Thin walled with short nodal fusions of 2 rarely 3 siphons for less than 100 µm	Thin walled with short nodal fusions of 3 rarely 2 siphons for more than 100 µm	Thin walled with short nodal fusion of 3 rarely 2 siphons for more than 100 µm

4. Segments mainly reniform, moderately calcified, white when dried, up to 23 mm broad; secondary utricles elongated *H. tuna*

***Halimeda monile* (Ellis and Solander) lamouroux:** The thallus is erect, light-green in color, generally slender and moderately calcified, measuring up to 13 cm tall, anchoring in sand by a large bulbous holdfast [11]. Its branching is dichotomous to tetrachotomous with cylindrical compressed basal segments. Upper segments are primarily cylindrical to slightly compressed, 7 mm broad, 8 - 13 mm long, some are fan-shaped of 9 mm wide and 6 mm long with three upper appendages (Fig. 2). Cortex of different segments is dense, composed of up to five layers of utricles. Surface utricles (the primary ones) are elongated, up to100 µm long, with large hexagonal outlines in surface view (Fig. 7); secondary utricles are elongated, measuring 44 to 74 µm in length. Medullary siphons are interwoven and fused at node into a single unit with a series of pores that more than 20 µm in diameter (Fig. 7).

The species was found from the intertidal up to a depth of 13 m. Populations from North Halaib were found partially buried in sand among beds of the angiosperm in shallow waters. Specimens from the shallow depths are robust and calcified with cylindrical to sub-cylindrical segments, whereas specimens from deep water are less calcified, loose in branching and with more fan-shaped segments.

Locality: this species was found in Red Sea (Fig. 1) at Koseir, Shalateen to North Halaib coast and their off shore islets.

***Halimeda macroloba* decaisne:** The thallus is erect and flat, moderately to lightly calcified, dark to light green in color, up to 10 cm long and anchored in sand by a large bulbous holdfast. Branching is mainly dichotomous with rare trichotomous [12]. The first two basal segments are rectangular with slightly expanded anterior portion. Upper segments are generally fan-shaped with few reniform, measuring 12-24 mm wide and 15-18 mm long (Fig. 3). Upper margins are undulate and thick. Cortex of different segments is dense and up to four layers of utricles. Surface utricles are similar to *H. monile* in surface view, elongated and hexagonal, but smaller with up to 54 µm long (Fig. 7). Secondary utricles are elongated, up to 92 µm long and every utricle supports 2-4 primary (surface) ones. Medullary siphons are interwoven and fused at node into a single unit with a series of pores that less than 20 µm in diameter (Fig. 7).

This species was collected from both the shallow and deep subtidal habitats. In few locations this species occurred in dense intertidal populations among species of *Caulerpa*, *Udotea* and other *Halimeda* species. The specimens are more vigorous and calcified in shallow than the deep subtidal. Specimens from the shallow depths showed some differences in gross morphology from those growing in much deep water (Table 1).

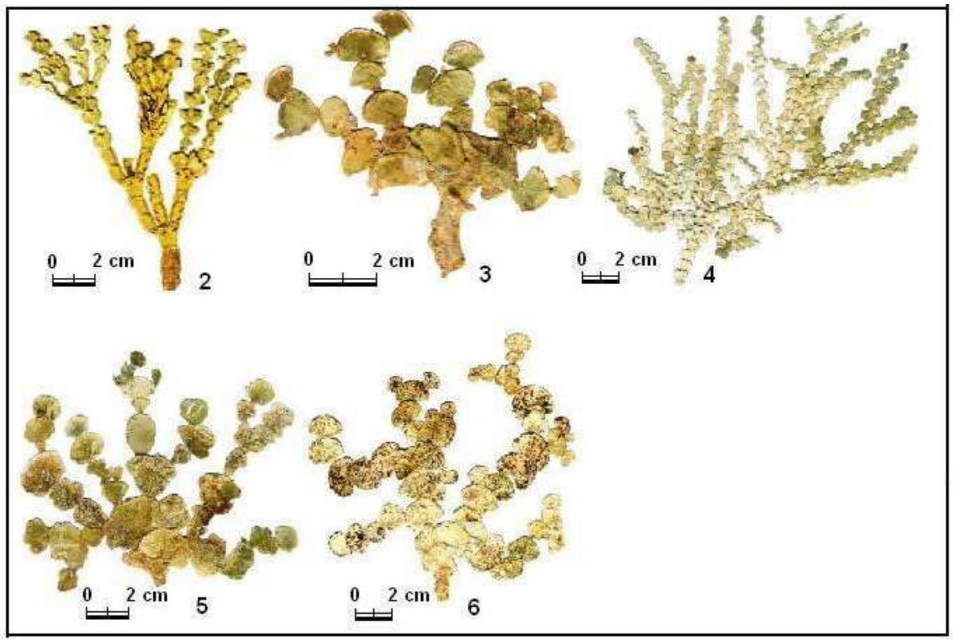

Fig. 2-6: General morphology of *Halimeda* species, 2; *H. monile*, 3; *H. macroloba*, 4; H. *opuntia*, 5; H. *discoidea*, 6; *H. tuna*

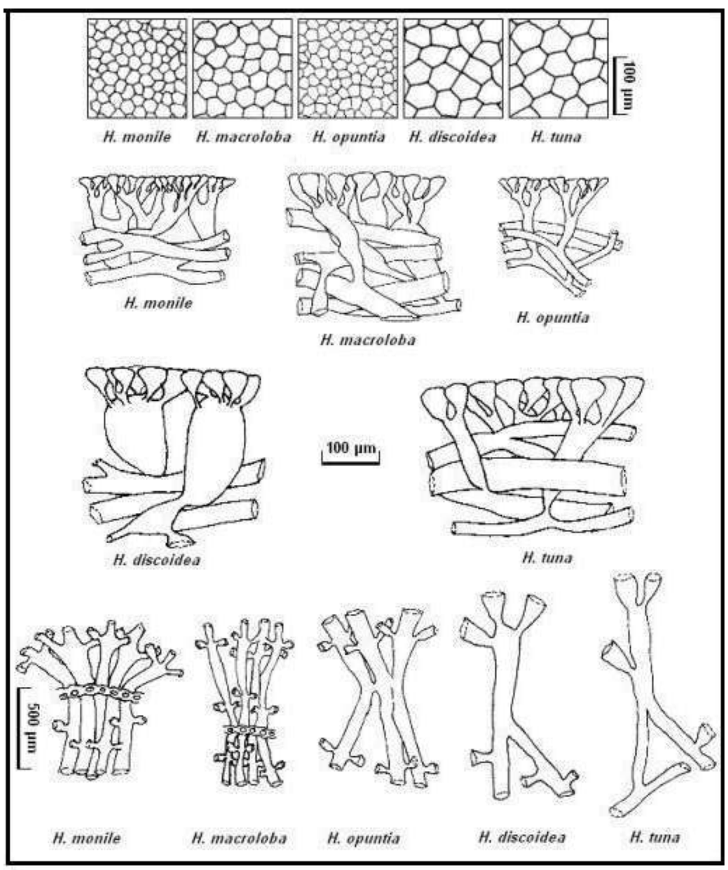

Fig. 7: Surface views of segments (upper), shapes of utricles (middle) and details of nodal fusion (lower) of different species of *Halimeda*

Locality: this species was found in Red Sea (Fig. 1) at Suez, Shalateen to North Halaib coast and their off shore islets.

Halimeda opuntia **(Linnaeus) lamouroux:** The thallus forms substantially calcified, white-green thick prostrate clumps, up to 20 cm in length, attaching to sandy-reef substrata by multiple felt-like holdfasts from any point at its lower portion [11]. It becomes greenish to cream when dried. Branching is dense, irregular and overlapping. Few basal segments are cylindrical. Upper segments are heart to reniform, up to 12 mm wide and 7 mm long, with prominent central rib often with two other lateral ribs (Fig. 4). Their distal margin is sinuating to deeply lob. Cortex appeared thin as it has 3-5 layers of small utricles, which are irregular in surface view. Secondary utricles are elongated, up to 65 µm long and every utricle supports 2 peripheral ones. Medullary siphons are interwoven and fused at node into small groups of 2 or 3 siphons. Fusions occur over a distance of less than 100 µm (Fig. 7).

The species is collected from various reef habitats in both shallow and subtidal habitats of the Red Sea. In some places it forms dense beds and their dead segments accumulated in relatively large amounts. On reef flats clumps are more compact and imbricate with twisted segments, whereas clumps are looser, branching tends to be distichous and segments become reniform on vertical reef. Less calcified specimens were also collected from shaded sheltered lagoons.

Locality: this species was found in Red Sea (Fig. 1) at Suez Canal, Suez, Gharib, Ghardaqa, Safaga, Koseir, Zabargad, Shalateen to North Halaib coast and their off shore islets.

Halimeda discoidea **decaisne:** The thallus forms erect yellowish green clump with single small holdfast, lightly calcified, reaching 12 cm in length. It becomes white on drying on herbarium sheets. It attached to consolidated substrata by a felt-like holdfast [13]. The holdfast showed to be a minute bulbous holdfast when the alga anchored to unconsolidated substrata. Branching is dense to sparse, di- to trichotomous. Segments are generally discoid, but few are reniform, with a smooth surface, up to 33 mm wide and 21 mm long (Fig. 5). Cortex has 2-3 layers of utricles. Surface utricles are large, sub-globular, hexagonal to pentagonal in surface view, holding together strongly even after decalcification. Secondary utricles are sub-globular, up to 92-210 µm wide and every utricle supports many primary (surface) ones. Medullary siphons are interwoven and fused at node into small groups of 2 to 3 siphons. Fusions occur over a distance of more than 100 µm (Fig. 7).

The species was found in sheltered to exposed biotopes. Specimens of this species were generally gathered from subtidal habitats and often occurred together with *H. opuntia*. When grow in shallow habitats, specimens tend to be darker in color, more calcified and densely branched, with smaller and thicker segments.

Locality: this species was found in Red Sea (Fig. 1) at Ghardaqa, Safaga, Koseir, Zabargad, Shalateen to North Halaib coast and their islets.

Halimeda tuna **(Ellis and Solander) lamouroux:** The thallus is erect, up to 15 cm in length, light green in color when alive, greenish to cream when dried, moderately calcified, anchoring with inconspicuous rhizoidal holdfast. Basal portion consists of one or two segments which may be fused, forming a barrel shaped stipe. Branching is sparse to dense, dichotomous to tetrachotomous [11]. Upper segments are broadly reniform, overlapping, with dull glossy surface, up to 23 mm wide and 13 mm long (Fig. 6). The distal margins are significantly thicker appearing wavy and folded when pressed. Cortex has 3-4 layers of utricles. Surface utricles are large, elongated, hexagonal in surface view, lightly holding together even after decalcification. Secondary utricles are also elongated, up to 140 µm long. Similar to *H. discoidea*, medullary siphons are interwoven and fused at node into small groups of 2 to 3 siphons. Fusions occur over a distance of more than 100 µm (Fig. 7).

The species inhabited both shallow and deep subtidal region. In shallow water, specimens were dark green, more calcified, thicker, smaller and rougher segments and with a smaller holdfast.

Locality: this species was found in Red Sea (Fig. 1) at Suez Canal, Sukhna, Gharib, Ghardaqa, Safaga, Koseir, Zabargad, Shalateen to North Halaib coast and their off shore islets.

DISCUSSION

The distribution of the five reported species of *Halimeda* along the Egyptian coast of Red Sea was concentrated in the southern part, decreasing northward. The frequency of each species at the individual site was also the same. Thick clumps of all species, especially of *H. opuntia*, were frequently found at the southern area from Shalateen to North Halaib coast and their off shore islets. This fits well with the distribution of other species of macroalgae on southern reefs of Red Sea [8] where the area is far from the sources of pollution. The northern distribution of the genus is limited by only the *Halimeda tuna* and *H. opuntia*. The latter is limited to the southern part of Suez Canal, while the former extends its distribution into the Mediterranean. As mentioned by [14], the type locality of the *H. tuna* is the Mediterranean Sea and is widely distributed in both tropical and subtropical regions such as Atlantic [15], Pacific [16] and Indian Ocean [17].

Halimeda opuntia is the most common and widespread species of *Halimeda* worldwide [17-20]. This species could be of greatest importance as sediment producer in many reef regions in the Red Sea. It was often occurred together with *H. discoidea* at various reef habitats in both shallow and subtidal habitats. In some places it forms dense clumps and their dead segments accumulated in relatively large amounts. It is well known that *Halimeda* species produce aragonite form of calcium carbonate and can double their size in 15 days and produce 7 g dwt m^{-2} d^{-1} [1]. Other reported species of *Halimeda* were also known to be tropical and subtropical in distribution [17-19, 21-23].

As to vertical distribution, the species of *Halimeda* in the Red Sea occupied different photophil and sciaphil biotopes from shallow to deep water (25 m) and from exposed to sheltered situations. The genus is well known for having species living deeper in the sublittoral zone [18]. In Brazil, *Halimeda discoidea* was found at depths of over 160 m, whereas *H. tuna* and *H. opuntia* were found up to 60 m [24]. In Red Sea, the reported species were found in shaded areas such as crevices, cliffs, fissures, gullies, overhangings and ledges, in association with the other sciaphilic species of seaweeds such as *Caulerpa racemosa*, *C. serrulata* and *Udotea argentea*. In shallow waters, *Halimeda* may hideaway from intense solar radiation by occupying the areas between other larger algae such as *Laurencia papillosa* and *Sargassum ilicifolium*. *H. opuntia* and *H. tuna* were only the species that dominate both highly illuminated and shaded habitats.

Variations in species morphologies that occurred between locations usually reflect differences in light intensity; and this is in close to that previously noticed by [25] for the genus *Caulerpa*. For instance, *Halimeda opuntia* showed marked difference in branching pattern and segment morphology when grows under different light intensity. In photophilic biotopes, this species forms extensive clumps with entangled branches. In sciaphilic biotopes, it forms loose tufts with distichous branching. Its segments have different degrees of calcification and may be flat, sub-cylindrical, contorted, auriculate, trilobate or crenulate depending on where it was growing. Morphological variability as this found in the present study has been led [26] to divide *H. opuntia* into three forms: f. *cordata*, f. *triloba* and f. *minor*. This division is not supported by the observations found in the present study, as the fusion pattern of nodal medullary siphons, in addition to the size, shape and distribution of the utricles seem to vary less among the specimens collected from different biotopes.

H. monile and *H. macroloba* were the less frequent species during this study. Their gross morphology in relation to light conditions showed no marked changes. Specimens from the shallow lightened depths were robust and more calcified; whereas specimens from deeper and less lightened areas were slightly smaller and less calcified. Segment shape showed small morphological differences.

Halimeda discoidea and *H. tuna* exhibited very similar gross morphology that makes difficulties in distinguishing some specimens from different biotopes. In regarding this problem, [27] put together these two species with other closely related ones under the *H. tuna* complex. Later on, [1] divided the genus into five sections, based on the patterns of nodal structure, putting this two species in section *Halimeda*. In the present study, *H. discoidea* can be distinguished microscopically by the presence of inflated or sub-globular secondary utricles. On the other hand, the secondary utricles of *H. tuna* are elongated. The use of gene sequencing in the classification of 33 species of *Halimeda* [5] supports in a great part the classification based on morphological characters [1], but not always. Molecular data of [5] showed that *H. tuna* from the Atlantic and from the Mediterranean were not nearest neighbor.

In conclusion, the identification of *Halimeda* at species level is possible only by combination of several criteria including biotopes, gross morphology and anatomy of mature segments and nodes. Nodal structure seems to be less varied at different biotopes and, therefore, is useful criterion. The southern area of the Egyptian Red Sea coast favors the growth of all study species of *Halimeda*, where two species were found in the northern area and this may point out for conservation of the reef communities.

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