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**EXTRAORDINARY MOUND-BUILDING FORMS OF *AVRAINVILLEA*
(BRYOPSIDALES, CHLOROPHYTA): THEIR EXPERIMENTAL TAXONOMY,
COMPARATIVE FUNCTIONAL MORPHOLOGY AND ECOLOGICAL
STRATEGIES**

BY

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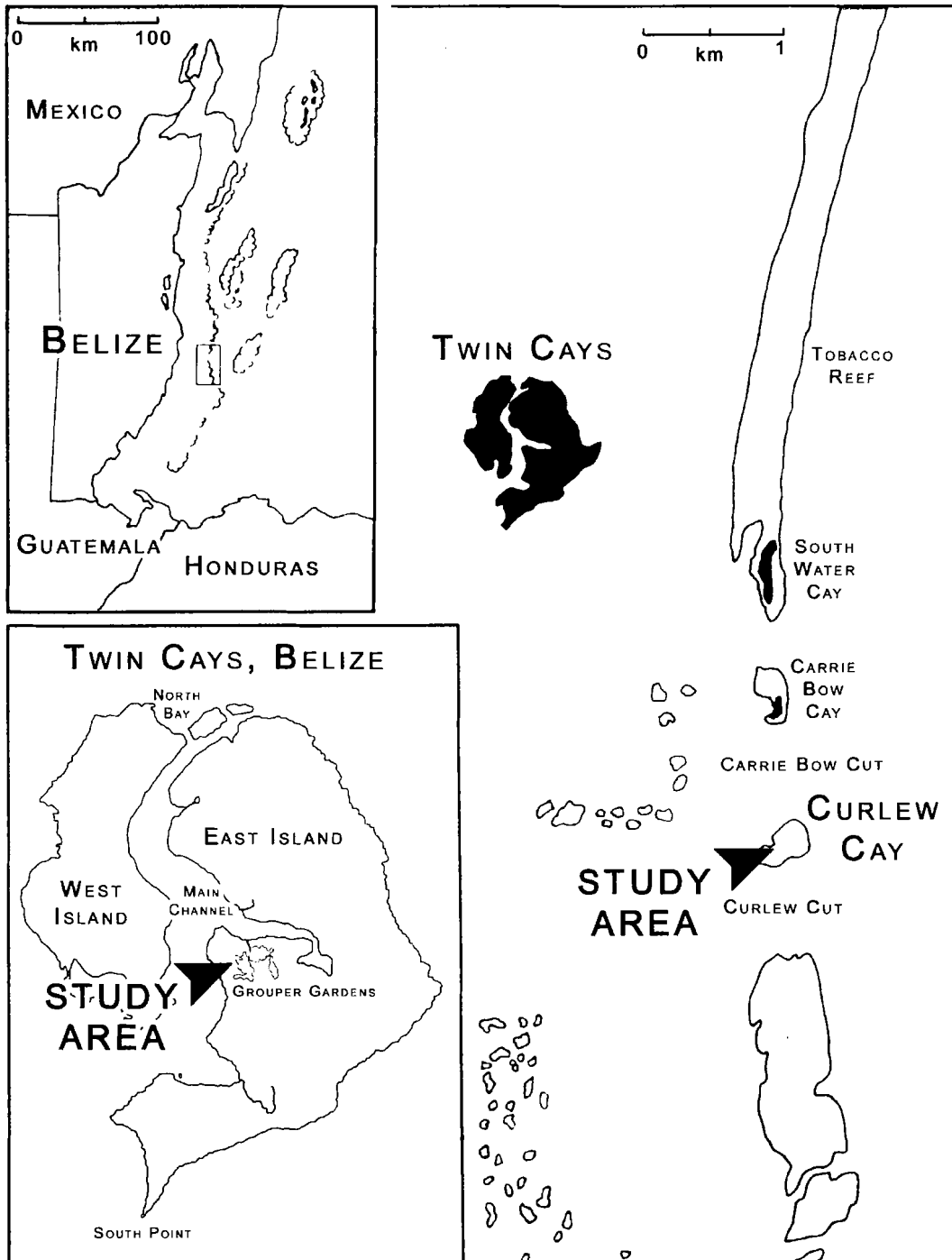


Figure 1. The Central Province of the Belize Barrier Reef showing the study sites on Twin Cays and Curlew Cay.

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ABSTRACT

The discovery of astounding mound-building forms of *Avrainvillea* (to 30 m diam.) catalyzed this study. These colonial (possibly clonal) mounds dominate the standing stocks and productivity of protected, shallow, eutrophic interiors of Belizean mangrove islands. A common-garden reciprocal-transplant experiment showed that the mound formers (*A. longicaulis* f. *laxa* and *A. asarifolia* f. *olivacea* from Twin Cays), which we initially hypothesized to be undescribed species, readily acquired the morphological features consistent with the taxa characteristic of open-water habitats (*A. longicaulis* f. *longicaulis* and *A. asarifolia* f. *asarifolia* from Curlew Cay), thereby falsifying the hypothesis that the mound formers are distinct species.

In support of the coloniality hypothesis, the Twin Cays f. *laxa* and f. *olivacea* morphs were uniquely adapted to produce flabellar stipes that serve as shallow subterranean rhizomes which spread laterally to overgrow rich organic peat bottoms. The massive columnar rhizoidal holdfasts found in the Curlew Cay f. *longicaulis* and f. *asarifolia* morphs were adaptive for both anchoring and obtaining pore-water nutrients, but proved to be superfluous under placid enriched water-column nutrient conditions and were incapable of surviving the deeper anoxic conditions of the composting peat deposits. The large colonial mangrove morphs (i.e., f. *laxa* and f. *olivacea*) were not physically resistant to even the moderate current levels (3.6 ± 0.5 cm per sec) encountered in the back-reef lagoon habitats of the deeply anchored morphs (i.e., f. *longicaulis* and f. *asarifolia*). However, smaller 2-3 blade clumps, with their stipes deeply buried, survived and grew.

Consistent with the perennation hypothesis, only the experimentally amputated Curlew Cay morphs (both f. *longicaulis* and f. *asarifolia*) showed significantly more proliferations (100 %) than either the amputated Twin Cays morphs (both f. *laxa* and f. *olivacea*) or the uncut Curlew and Twin Cays control plants. The stipes and blades of the open-water morphs (f. *longicaulis* and f. *asarifolia*) serve as expendable assimilators with a major function of building a massive perennating/storage organ, the columnar holdfast, which comprises the bulk of the plant. Physical disturbances (such as storms and herbivory), as well as physiological stresses (such as epiphyte loading), can cause disproportionate losses of the relatively delicate expendable assimilators which are replaced subsequently by perennation from the long-lived subterranean holdfast during more favorable conditions.

INTRODUCTION

Overall, the ecology of siphonaceous green algae (Bryopsidales) is not well known even though members of this group occur abundantly in virtually all tropical open-water reef and lagoon habitats. The discovery of three incredible mound-building colonial forms [=morphs or forma (f.)] of *Avrainvillea*, dominating the standing stocks and productivity of submerged habitats within Belizean mangrove island interior creeks, ponds and lakes, literally demanded this study. These persistent mound-formers are restricted to shallow (<3 m), calm, peat-bottom, high-nutrient waters in the protected interiors of mangrove islands.

Lagoons of the Belize Barrier Reef Central Province, such as those westward of Carrie Bow Cay and Curlew Cay (Fig. 1), are the most extensive of the entire reef tract including diverse and abundant populations of sand-dwelling macroalgae and seagrasses. These back-reef environs comprise a well-developed lagoonal system remote from major human pollutants. Organic detritus rarely accumulates on coral-dominated reefs because of intense herbivory and export processes. However, the characteristic nutrient limitation patterns typically observed in tropical reef systems are not applicable to the detritus-rich mangrove-peat systems of Twin Cays (Lapointe et al., 1987), which are characterized by elevated nutrient availability.

In general, mangrove ecosystems are well-known for their high levels of marine compost (Fell et al., 1980; Newell et al., 1984) that release relatively high concentrations of dissolved inorganic nitrogen and phosphates (Snedaker and Brown, 1981) into the adjacent water column. Considering that nutrient uptake kinetics in macroalgae are highly concentration dependent, mangrove macroalgae have been shown (Lapointe et al., 1987) to be far less nutrient-limited compared to macroalgae on coral reefs, based on seawater and tissue analyses as well as nutrient limitation/bioassays performed at the identical sites investigated here (i.e., Curlew Cay and Twin Cays, Fig. 1). Also, the geology, natural history and biology of these systems are comparatively well-known as a result of over three decades of multidisciplinary investigations (see Rützler and Macintyre 1982, Rützler and Macintyre this volume).

The rhizomatous ("rooted") Bryopsidales are considered to be important stabilizers of both organic and carbonate sediments. It also has been documented (Williams and Fisher, 1985; Littler et al., 1988; Littler and Littler, 1990) that the rhizoidal sand-dwelling forms of the open lagoon play a significant role in cycling nutrients from sediment pore waters. Rhizophytes such as *Avrainvillea*, *Udotea*, *Halimeda*, *Penicillus*, *Rhipocephalus*, *Cladocephalus* and *Caulerpa* (Chlorophyta, Bryopsidales) are among the predominant contributors to macroalgal cover and primary productivity within the vast seagrass meadows throughout the tropical western Atlantic. Such "rooted" plants, by tapping into the nutrient-rich interstitial pore waters (Littler and Littler, 1988), can avoid many of the nutrient-limitation problems experienced by their rock-dwelling counterparts.

While seagrasses and diverse macroalgal phyla are abundant on the outer perimeters of Twin Cays (Fig. 2, Littler et al., 1985), it is the siphonaceous Chlorophyta that dominate the standing stocks and productivity of submerged interior habitats within the mangrove island proper. In particular, the genus *Avrainvillea* is conspicuous among the predominant contributors to biomass and primary productivity within the vast array of creeks, ponds, lakes and borders of Twin Cays. Some members of the siphonaceous green algae characteristic of Twin Cays contain unique and interesting secondary chemical compounds (Sun et al., 1983;

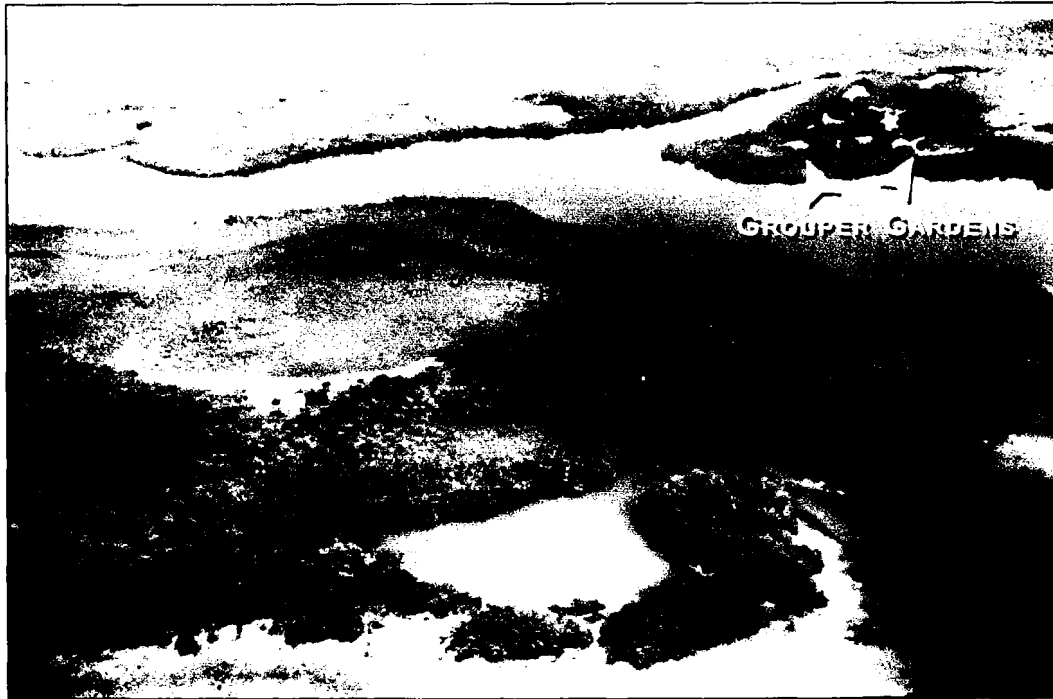


Figure 2. Oblique aerial view of Twin Cays (looking east) showing the many hidden lakes and ponds. The Grouper Gardens study site is labeled on the upper right.

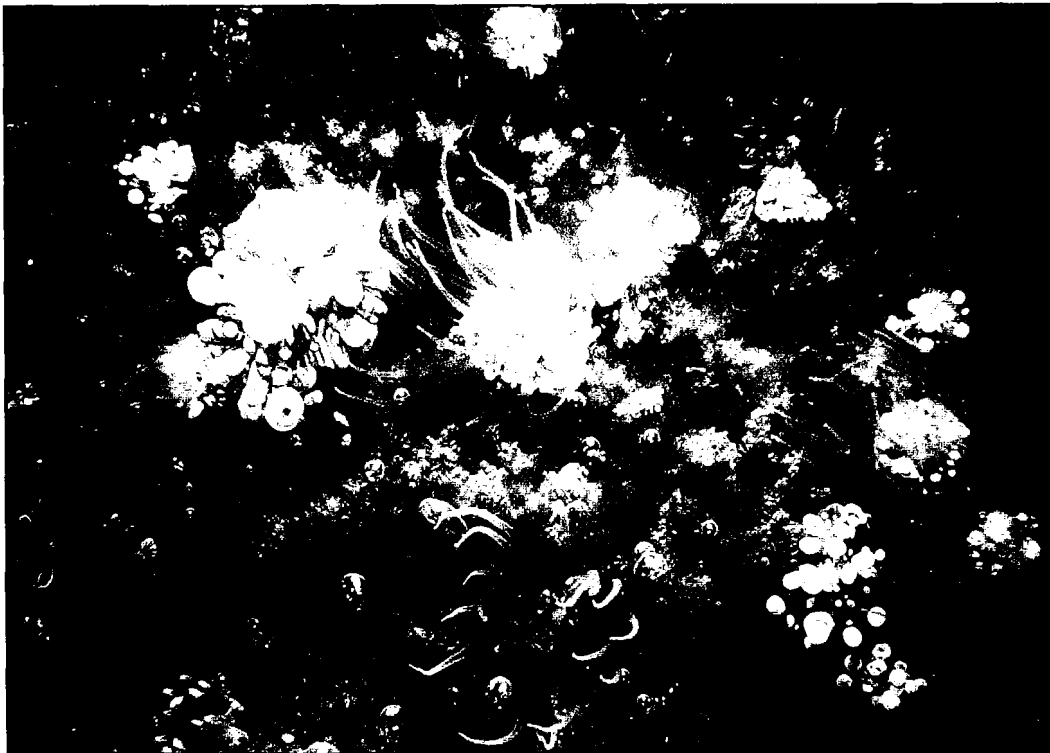
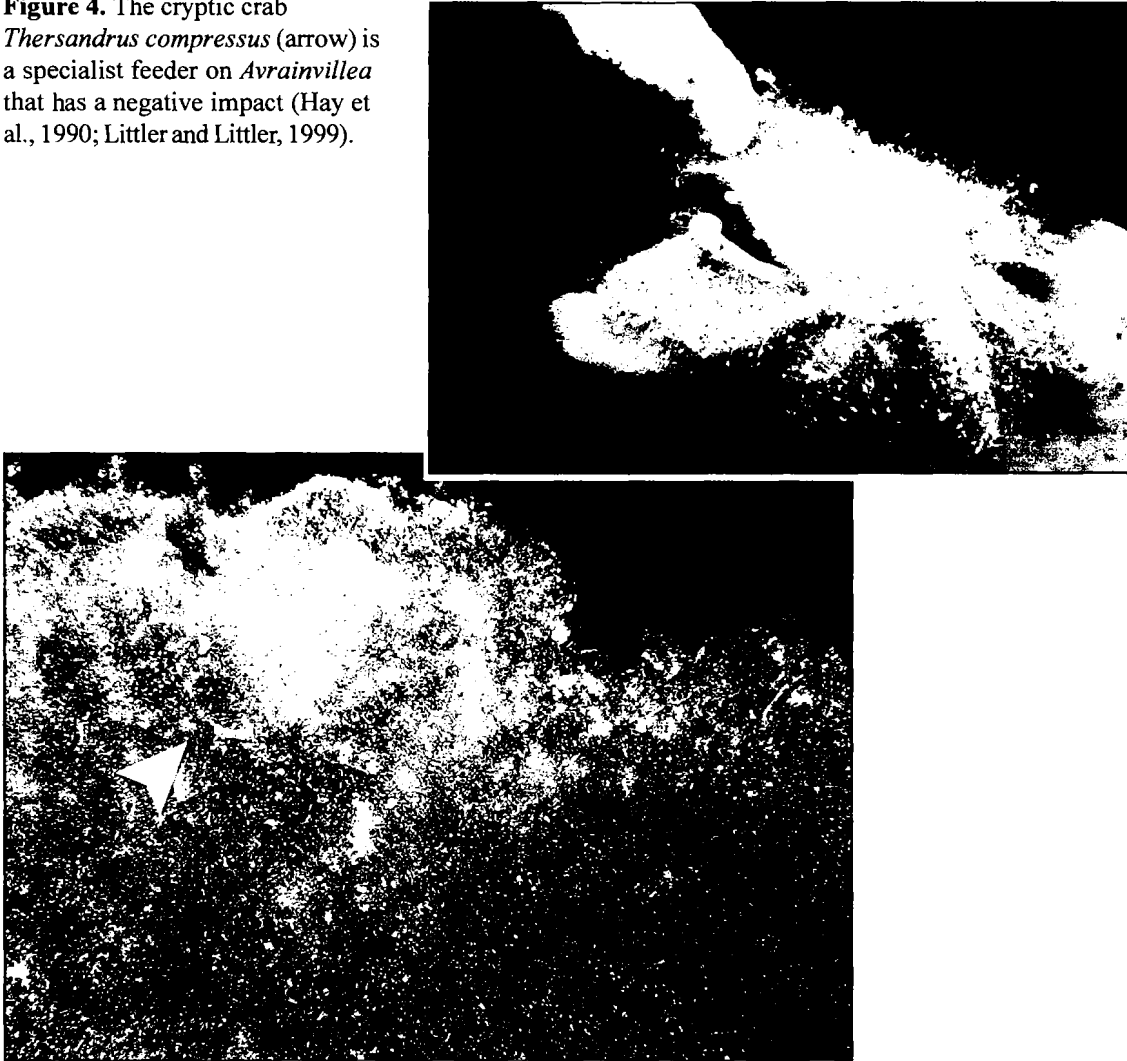


Figure 3. Colonial sea anemones using *Avrainvillea* blades as an attachment substrate.

Figure 4. The cryptic crab *Thersandrus compressus* (arrow) is a specialist feeder on *Avrainvillea* that has a negative impact (Hay et al., 1990; Littler and Littler, 1999).



Hay and Fenical, 1988) and highly specialized interactions between the larger forms (e.g., *Udotea*, *Avrainvillea*, *Caulerpa*, *Penicillus*) and such herbivorous invertebrates as crabs and molluscs have been observed (Hay et al., 1990; Littler and Littler, 1999). *Avrainvillea* provides microhabitats (Fig. 3), as well as food and shelter (Fig. 4), for many meso- and micro-invertebrates. These one-sided associations have proven (Hay et al., 1990) to be primarily beneficial to the invertebrates and detrimental to the algal host.

The mucilage-free spongy textures of *Avrainvillea* would seem to make them susceptible to epiphytic plant/animal loading. However, we showed in an earlier study (Fig. 5, Littler and Littler, 1999) that the solitary lagoon morphs are able to rapidly produce new fronds by cytoplasmic streaming and translocation through their siphons, a process that is not impaired by cross walls (as is the case of cellular plants). This represents a unique antifouling mechanism (Littler and Littler, 1999) whereby old assimilators and their inhibitory epiphytes can be shed by “blade abandonment/proliferation” at relatively low cost to the plant.

A major obstacle to understanding the ecological role of siphonaceous algae at Twin Cays has been the high biodiversity of the taxonomically problematical genera named above. Six distinct species of *Avrainvillea* co-occur in the creeks, ponds and lakes of Twin Cays (Figs. 6, 7). Using treatments prior to the beginning of this investigation (Taylor, 1960; Norris and Bucher, 1982), it would have been possible to discern only a small fraction of the taxa that are actually present. As one example, *Avrainvillea longicaulis* f. *longicaulis* (Fig. 6) and the similar appearing *A. mazei* (Fig. 7) co-occur at Twin Cays as well as throughout lagoonal grass bed habitats and require precise discrimination (see misidentification of *A. mazei*, as *A. longicaulis*, on page 225 of Humann and DeLoach, 2002). In fact, there had been no serious systematic work on the group since the turn-of-the-century (Gepp and Gepp, 1911) with the



Figure 5. The paddle-like blades of the lagoon forms of *Avrainvillea* (A) can rapidly translocate protoplasm to proliferate new epiphyte-free blades (B-natural epiphytes, C-mesh bag).

exceptions of herbarium-based treatments of *Halimeda* (Hillis-Colinvaux, 1980) and Pacific *Avrainvillea* (Olsen-Stojkovich, 1985). The systematic monograph on tropical western Atlantic *Avrainvillea* (Littler and Littler, 1992), as well as the floristic field guide for the nearby Pelican Cays (Littler and Littler, 1997), alleviated the major taxonomic stumbling blocks and enabled this study.

Experimental Organisms

As mentioned, the siphonaceous green algal genus *Avrainvillea* often dominates the standing stocks and productivity of submerged habitats within mangrove island creeks, ponds and lakes as well as occurring abundantly throughout virtually all calm-water reef systems.

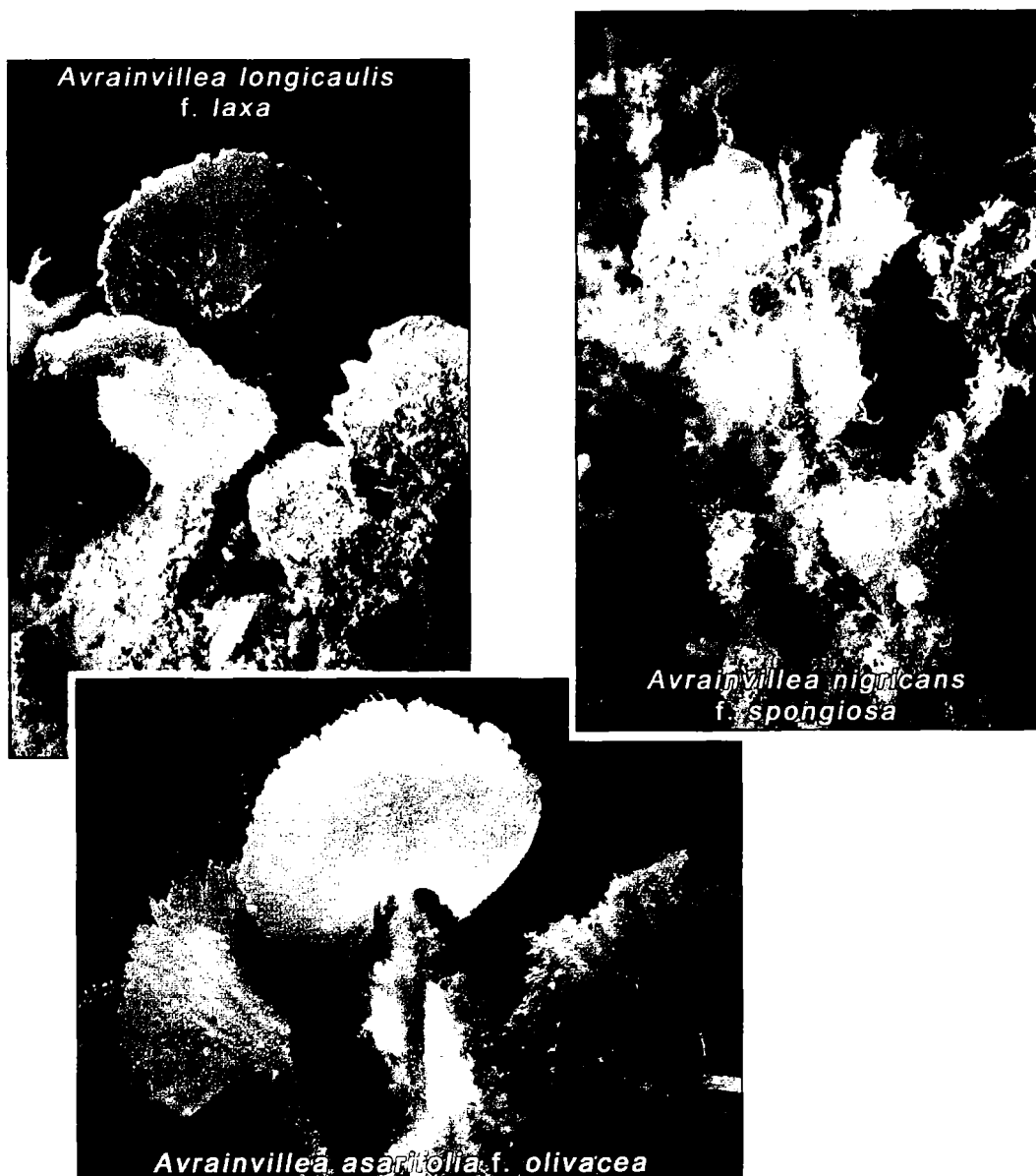


Figure 6. The three species shown here (*Avrainvillea nigricans* f. *spongiosa*, *A. asarifolia* f. *olivacea* and *A. longicaulis* f. *laxa*) create large mound-like colonies in mangrove lakes and ponds.

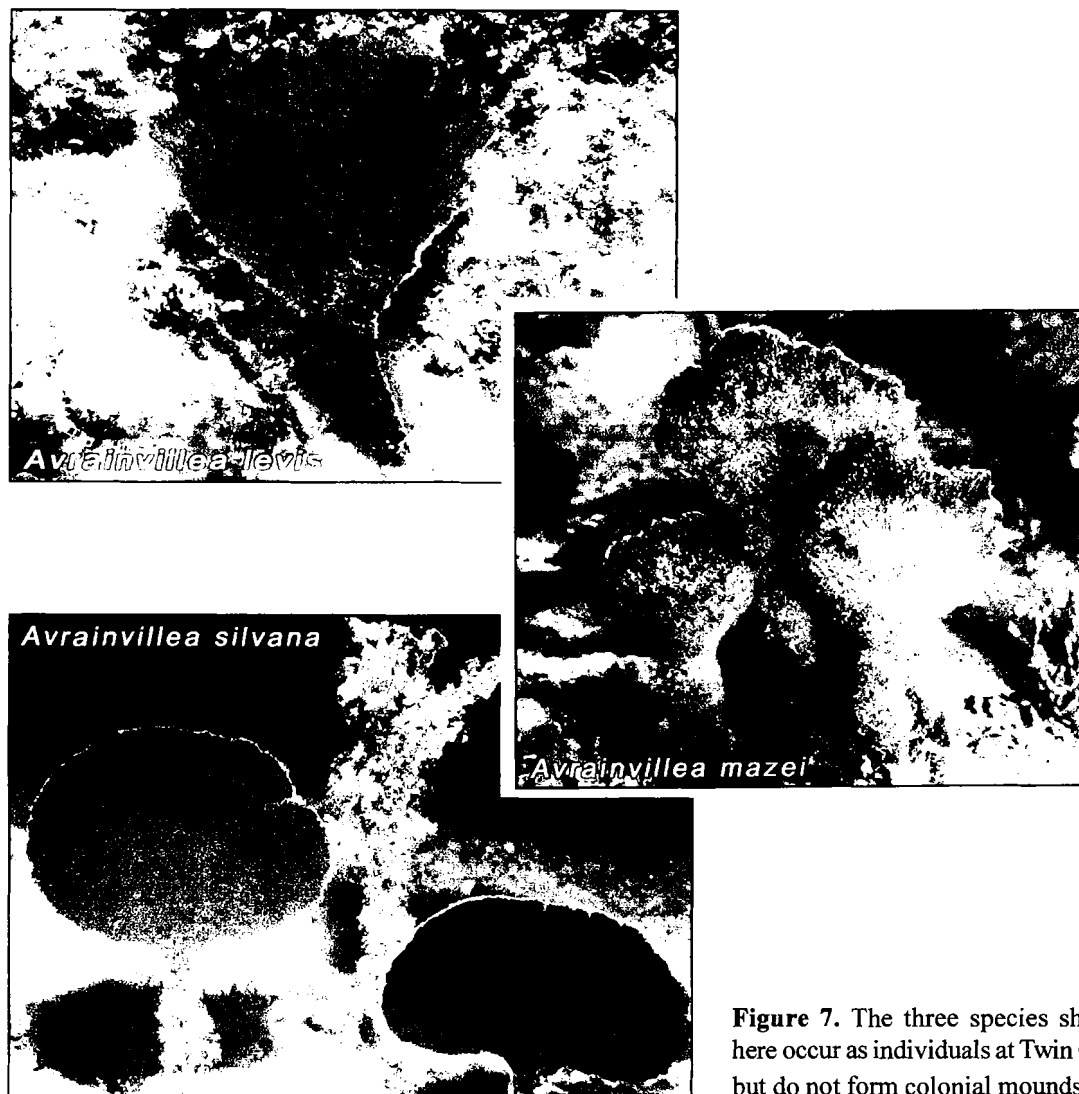


Figure 7. The three species shown here occur as individuals at Twin Cays but do not form colonial mounds.

Although sporogenic reproduction has never been reported for Belizean *Avrainvillea*, rare club-shaped release structures produced at the tips of individual blade siphons have been observed elsewhere (Littler and Littler, 1992). Unlike other Bryopsidales, species of *Avrainvillea* are long-lived (see Littler and Littler, 1992) and do not undergo holocarpic [i.e., mass synchronous sporogenesis (Clifton, 1997) followed by death and disintegration of the entire thallus].

The experimental macroalgae *Avrainvillea longicaulis* f. *longicaulis*, *A. longicaulis* f. *laxa* (Fig. 8), *A. asarifolia* f. *asarifolia* and *A. asarifolia* f. *olivacea* (Fig. 9) are particularly abundant but mostly unstudied in the Belize Barrier Reef lagoon and mangrove islands. The paddle-shaped blades (=flabella, caps or assimilators) of *Avrainvillea* number from one to many and are broadly oval (to 24 cm high, to 29 cm wide) with truncated lower margins. They are thick (> 4 mm) and spongy (lacking a mucilaginous coating) with cylindrical or flattened stipes (to 12 cm long, 13 mm diam.). The blades, stipes and holdfasts are composed of dichotomously branched interconnected siphons entirely lacking cross walls. The thalli of *A.*

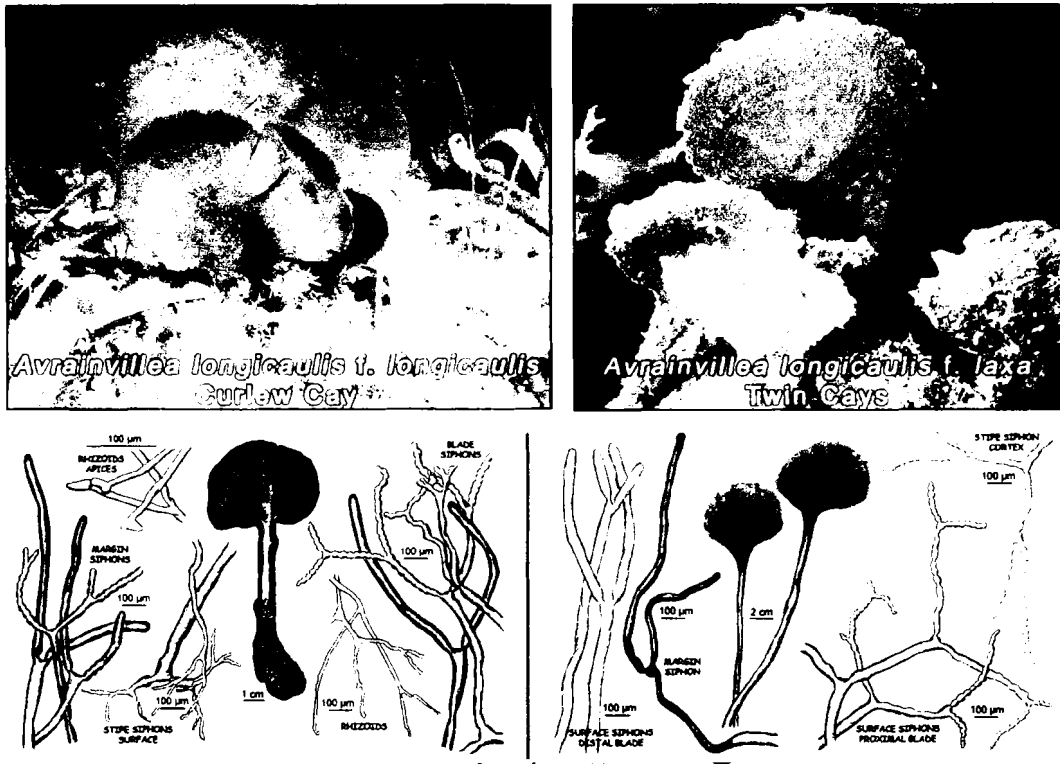


Figure 8. The two dramatically different morphological forms (morphs) of *Avrainvillea longicaulis* (f. *longicaulis* & f. *laxa*). However, note the anatomical (siphons) similarities.

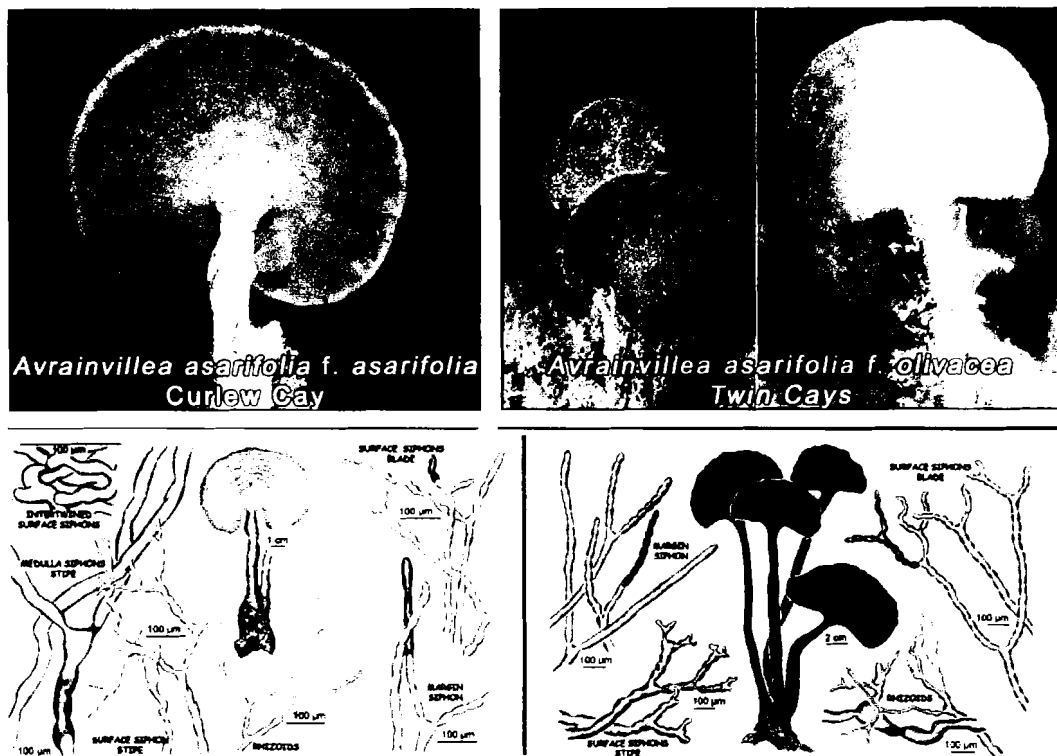


Figure 9. The two dramatically different morphological forms of *Avrainvillea asarifolia* (f. *asarifolia* & f. *olivacea*). However, note the anatomical (siphons) similarities.

longicaulis f. *longicaulis*, *A. asarifolia* f. *asarifolia* and *A. nigricans* f. *nigricans* (Figs. 8, 9, 10) are typically anchored by a massive, perennating, bulbous, rhizoidal holdfast (Fig. 11) in open sandy or seagrass areas of shallow (to 30 m) pristine waters.

As emphasized above, the discovery of incredible mound-building colonial morphs of *Avrainvillea* [*A. longicaulis* f. *laxa* (Fig. 8), *A. asarifolia* f. *olivacea* (Fig. 9) and *A. nigricans* f. *spongiosa* (Fig. 10)] catalyzed this study. These three colossal mound-formers are restricted to shallow (<3 m), placid, peat-bottom, high-nutrient waters in the protected interiors of mangrove islands.

HYPOTHESES TESTED

Coloniality Hypothesis

To reiterate, *Avrainvillea longicaulis* f. *longicaulis* and *A. asarifolia* f. *asarifolia* (Figs. 8, 9) are solitary in open lagoonal sandy environments with consistent wave action but can form extraordinary decades-old colonial (possibly clonal?) mounds (Fig. 12). The taxa, described (Littler and Littler, 1992) as *A. longicaulis* f. *laxa* (Fig. 8) and *A. asarifolia* f. *olivacea* (Fig. 9), are persistent in peaty, highly eutrophic, placid, interior mangrove habitats. The f. *laxa* and f. *olivacea* morphs hypothetically (i.e.,

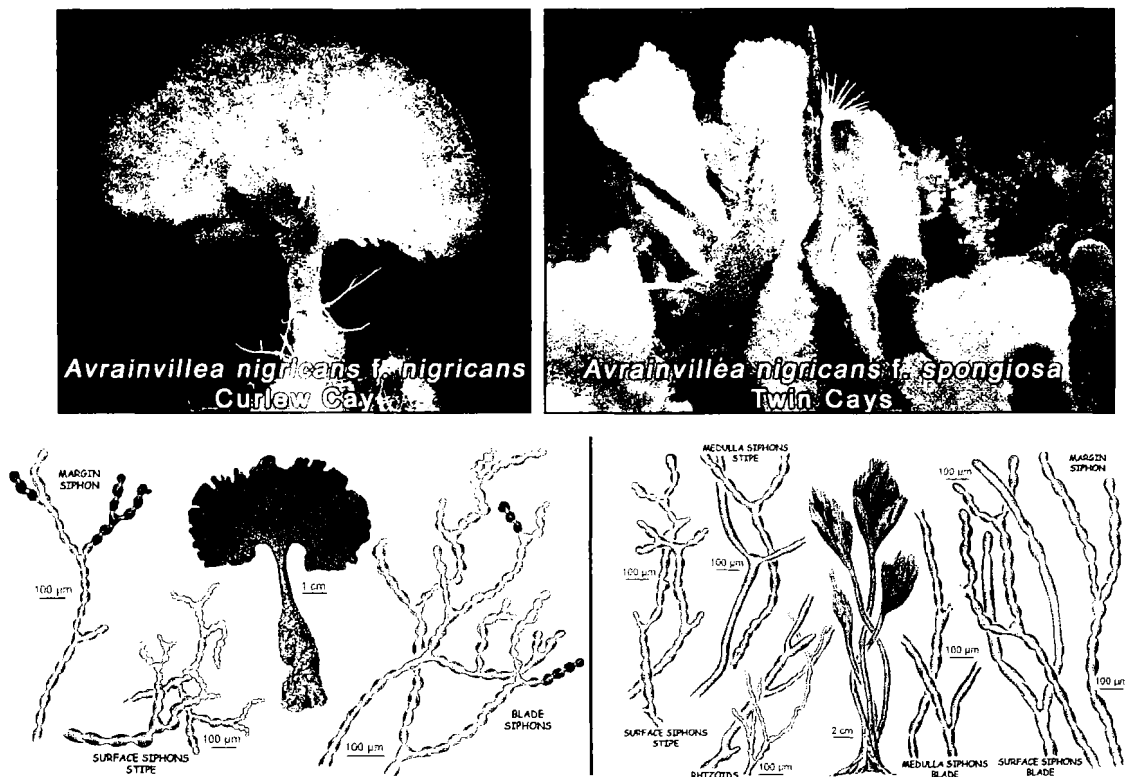


Figure 10. The two dramatically different morphological forms of *Avrainvillea nigricans* (f. *nigricans* & f. *spongiosa*). However, note the anatomical (siphons) similarities.



Figure 11. The massive perrenating, bulbous, rhizoidal holdfast of *Avrainvillea longicaulis* f. *longicaulis* characteristic of open sandy lagoonal areas.



Figure 12. Portion of a colossal colonial mound of *Avrainvillea longicaulis* f. *laxa* supporting diverse epiphytes at Twin Cays.

“coloniality hypothesis”) are uniquely adapted to utilizing flabellar stipes as shallow subterranean rhizomes that spread laterally to produce enormous (several meters-thick, to 30 m diameter, Fig. 12) mound-like colonies that overgrow rich organic peat bottoms. Massive columnar rhizoidal holdfasts, such as those found in the f. *longicaulis* (Fig. 11) and f. *asarifolia* (Fig. 9) morphs, hypothetically would be superfluous under placid enriched water-column nutrient conditions as well as incapable of surviving the deeper anoxic conditions of the composting peat deposits. Conversely, open-water wave surge and current drag on the huge colonial morphs (lacking a strong anchoring rhizoidal holdfast that could augment the low nutrient conditions in open lagoonal waters) should result in uprooting, wave-shearing damage and general attrition.

Perennation Hypothesis

In the open-water morphs of *Avrainvillea longicaulis* f. *longicaulis* and *A. asarifolia* f. *asarifolia* (Littler and Littler, 1992), we had observed what appeared to be perennation; where the remains of lost blades were indicated by breakage points and scars with newly forming flabella arising from either the former stipes or columnar holdfasts. We postulated (i.e., “perennation hypothesis”) that the stipes and blades of *Avrainvillea longicaulis* f. *longicaulis* and *A. asarifolia* f. *asarifolia* serve as expendable photosynthetic assimilators with a major function of building a massive perennial storage organ, the columnar rhizoidal holdfast. This structure (Fig. 11) can comprise up to 90% of the total thallus (Olsen-Stojkovich, 1985; Littler and Littler, 1999). In other words, physical disturbances (such as storms and herbivory) as well



Figure 13. Conducting primary productivity experiments on *Avrainvillea longicaulis* f. *laxa* at Twin Cays using oxygen electrode methods (Littler and Littler, 1987).



as physiological stresses (such as epiphyte loading) should result in disproportionate losses of the relatively delicate above-ground assimilators, which can be replaced by perennation from the massive subterranean holdfasts (Fig. 11) during more favorable conditions.

METHODS AND MATERIALS

Experimental Taxonomy

The critical initial phase of this research included completion of a systematic and phylogenetic monograph of Caribbean *Avrainvillea* based on intensive and extensive collections throughout Twin Cays and surrounding environments (Littler and Littler, 1992). As emphasized, the discovery of astounding mound-building persistent colonial morphs of *Avrainvillea* motivated this study. However, taxonomic issues still prevailed regarding these mound formers, which were initially thought by us to be distinct species although the internal anatomical data suggested (Littler and Littler, 1992) otherwise. The experimental “common garden” reciprocal transplant approach used here (see below) provided quantitative resolution of these issues.

Coloniality Hypothesis

We used a costs vs. benefits approach to test the coloniality hypothesis (N=10/ treatment). We posited that the two sets of remarkably different morphs (i.e., f. *laxa* vs. f. *longicaulis* and f. *olivacea* vs. f. *asarifolia*) are adaptive for their respective habitats. We attempted to experimentally induce colony formation in the f. *longicaulis* and f. *asarifolia* morphs by burial of the flabellar stipes as well as by conducting reciprocal transplant

experiments with appropriate control (C= tagged only) and transplant controls for both morphs. In each habitat (i.e., Curlew Cay and Twin Cays, Grouper Gardens, Figs. 1, 2), the transplant controls (TC) were completely removed by careful digging and gently replanted in holes wedged open by titanium crowbars in the nearby general area. The experimental transplants (T) were carefully removed, floated into a 100-liter cooler of seawater and transferred to the reciprocal field sites where they were carefully replanted as above.

As described earlier, this “common garden” approach also led to an experimental taxonomic analysis of whether or not the various morphs might represent distinct species. All replicates were tagged by nearby surveyors’ flags. After one year of growth, the plants were returned to the laboratory for final photography and morphometric documentation. If we could (1) induce coloniality in the individuals of *f. longicaulis* and *f. asarifolia* (under the presence of high nutrients and benign physical conditions) within calm interior mangrove ponds and (2) show that large colonies of *f. laxa* and *f. olivacea* are susceptible to removal by natural levels of current and wave surge, then the coloniality hypothesis would be deemed to be supported. A further bonus in support of the hypothesis would be (3) any sign of long-term induction of columnar holdfasts in the *f. laxa* and *f. olivacea* transplants moved from mangrove island pools into open water habitats.

Perennation Hypothesis

In addition, we concurrently tested the perennation hypothesis as follows: (1) in experimental lagoon thalli with blades physically amputated, proliferation of new blades should be stimulated and (2) moderate losses of blades should not lead to high levels of mortality relative to control plants not subjected to such mutilation. Twenty separate plants of *Avrainvillea longicaulis* *f. longicaulis* and another 20 of *A. asarifolia* *f. asarifolia* were assessed in the lagoon behind Curlew Cay. Both sets were divided into the following two replicate groups (N=10) by double randomization to provide: controls (CO, to correct for natural changes and possible stochastic events) and cut plants (C, to simulate natural physical damage). All were marked by surveyors’ flags.

The same procedure was repeated at Grouper Gardens for *Avrainvillea longicaulis* *f. laxa* and *A. asarifolia* *f. olivacea* (N=10/treatment). After one year, the controls (CO, which had been left intact) and the amputated/cut plants (C, which were trimmed with scissors, leaving the intact holdfast and 2-cm stipe lengths) were assessed for blade numbers and new proliferations. Data analysis employed ANOVA and the Bonferroni Test for significant differences.

Ecological Role

To assess the ecological importance of the *Avrainvillea longicaulis* *f. laxa* mounds, quantitative transect surveys of biotic cover were conducted using the nondestructive photogrammetric techniques developed by Littler and Littler (1985). This entailed video transects at right angles to the substrata that were then scored in stop action on a high-resolution video monitor. Cover was determined by recording the percentages of point intercepts from a randomized array superimposed over the video images. Two randomly selected 0.25 m sections at the edges of two separate mounds were harvested for biomass determinations.

These were cleaned of peat deposits and epiphytes, photographed and weighed wet. A set of subsamples from these were rinsed in freshwater, weighed, dried and reweighed to determine wet-to-dry weight relationships. Organic dry-weight (ODW) was determined by igniting the dried samples in a muffle furnace to constant weight at 500° C.

Primary productivity measurements were made for the dominant *Avrainvillea longicaulis* f. *laxa* using traditional light-dark bottle oxygen electrode techniques (Fig. 13). This, and the transect data, were used to ascertain an average mound's contribution to primary production at Twin Cays. We measured photosynthetic rates of the assimilators during early summer under ambient environmental conditions (30–31° C, 36 ppt salinity, 1500–2100 $\mu\text{mol photons m}^{-2}$ per sec) using the same methods detailed in Littler and Littler (1990). We incubated healthy assimilators containing natural levels of epiphytes and replicate blades with the epiphytes carefully removed by pinching (Fig. 13), as well as incubating the epiphytes separately (N=6 for all treatments), to ascertain the primary productivity contributions of the epiphytes. We chose photosynthesis as an indicator of physiological production since growth is relatively intractable due to the continual translocation processes and the inaccessibility of the stipe/holdfast system. Data analysis employed ANOVA and the Bonferroni Test for significant differences.

RESULTS

Experimental Taxonomy

The mound formers (*f. laxa* and *f. olivacea*), which were initially hypothesized to be putative species (Littler and Littler, 1992), presented unresolved taxonomic questions. The common-garden reciprocal-transplant approach provided definitive resolution of these issues. Following one year of transplantation, all experimental transplants had acquired the morphological features consistent with the taxa characteristic of the new habitats (Fig. 14, 15, 16, 17), thereby falsifying the hypothesis that the mound formers were distinct species.

Coloniality Hypothesis

The large colonial mangrove morphs (i.e., *f. laxa* and *f. olivacea*) were not resistant to even the moderate current levels (3.6 ± 0.5 cm per sec) encountered in the back-reef lagoon habitats of the deeply anchored morphs (i.e., *f. longicaulis* and *f. asarifolia*). When buried to normal depths, the massive natural colonial morphs would pull free and begin to drift downstream (Figs. 18, 21). However, the majority of the 2- to 3-blade clumps (Fig. 15), with their stipes deeply buried in the sandy sediments, were able to survive and grow.

After one year, the surviving 2- to 3-blade clumps transplanted from Twin Cays to Curlew Cay had developed rudimentary stages of the massive holdfast with the adherent sand grains (Fig. 15) characteristic of *f. longicaulis* and *f. asarifolia*. Conversely, all of the surviving morphs with normally massive holdfasts that were transplanted to the mangrove pools at Grouper Gardens showed degeneration of the columnar holdfasts (Fig. 17), with only remnant rhizoids containing clumped sand grains present at the end of one year. Concurrently, they had developed new assimilators/stipes characteristic of the colonial morphs (*f. laxa* and *f. olivacea*). Both sets of surviving controls and transplant controls were 100% uniform in retaining morphs consistent with their original habitats.

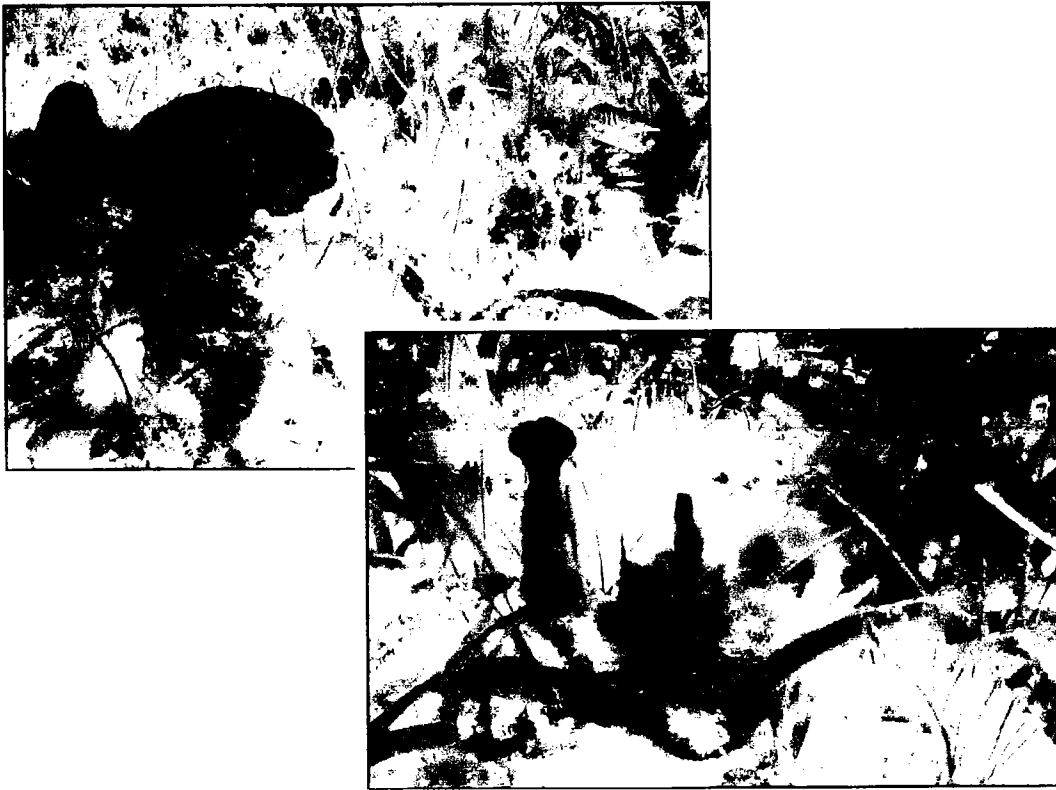


Figure 14. Examples of *Avrainvillea longicaulis* f. *laxa* transplanted from twin Cays to Curlew Cay after 12 months. Blades now are consistent with the f. *longicaulis* morph.



Figure 15. Examples of *Avrainvillea longicaulis* f. *laxa* transplanted from twin Cays to Curlew Cay and harvested after 12 months. Holdfasts now are consistent with the f. *longicaulis* morph.

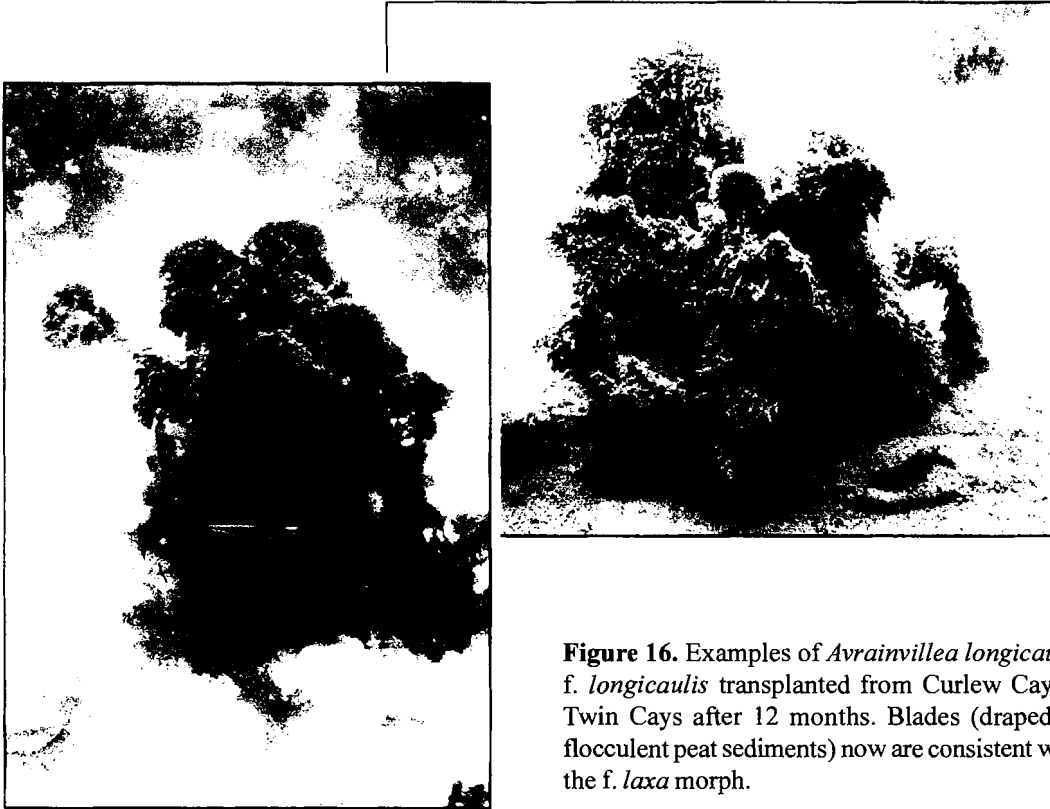


Figure 16. Examples of *Avrainvillea longicaulis* f. *longicaulis* transplanted from Curlew Cay to Twin Cays after 12 months. Blades (draped in flocculent peat sediments) now are consistent with the f. *laxa* morph.



Figure 17. Examples of *Avrainvillea longicaulis* f. *longicaulis* transplanted from Curlew Cay to Twin Cays and harvested after 12 months. Pseudo-rhizomatous holdfasts and stipes now are consistent with the f. *laxa* morph.

We also discovered unexpected evidence in further support of the coloniality hypothesis in the case of *Avrainvillea longicaulis* f. *laxa*. We found that the colonial morphology is uniquely reinforced by the intermingling of blade and stipe siphons at areas of contact (Figs. 19, 25). Contact frequently occurs for prolonged periods in such calm habitats, leading to abundant anastomosing points of fusion/adhesion.

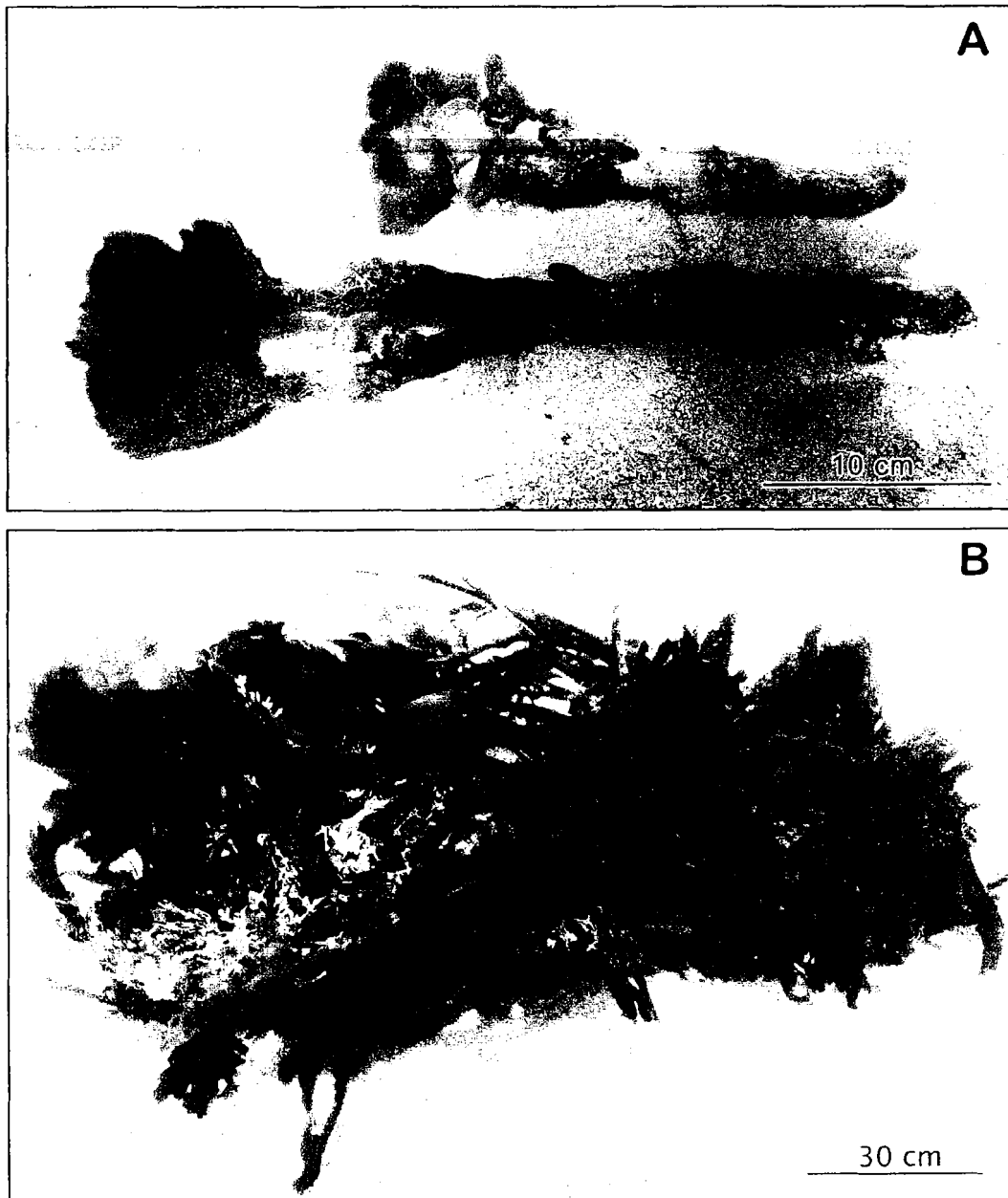


Figure 18. A – Two individuals of *Avrainvillea asarifolia* f. *asarifolia* from Curlew Cay. B – Colony of *f. olivacea* from Twin Cays. When both forms were transplanted to the back-reef sandy habitat, the *f. olivacea* colony was uprooted by current within hours, whereas the *f. asarifolia* thalli remained indefinitely.



Figure 19. Typical inter-thallus fusions characteristic of the colonial mound-forming species.

Perennation Hypothesis

In support of the hypothesis (Fig. 20), only the experimentally amputated Curlew Cay morphs (both *f. longicaulis* and *f. asarifolia*) showed significantly more proliferations (100 %) than either the experimentally amputated Twin Cays morphs (both *f. laxa* and *f. olivacea*) or the uncut Curlew or Twin Cays control plants. In particular, the amputated Curlew Cay *Avrainvillea longicaulis f. longicaulis* showed 100 % new proliferations, a significant fivefold increase relative to the Twin Cays *f. laxa* (20 %). The uncut controls from Curlew Cay *f. longicaulis* showed significantly fewer (70 %) new proliferations, whereas the Twin Cays experimental *f. laxa* plants also had significantly fewer (50 %) new proliferations.

In the case of *Avrainvillea asarifolia f. asarifolia* from Curlew Cay (Fig. 20), the experimentally amputated plants also had 100 % new proliferations paralleling the results for *A. longicaulis f. longicaulis*. The experimentally amputated *f. olivacea* also showed significantly fewer proliferations comparable to those for *f. laxa* (only 20 % new proliferations, significantly less at $P < 0.05$). The uncut control morphs of *A. asarifolia* from both Curlew Cay (*f. asarifolia*) and Twin Cays (*f. olivacea*) produced comparably low results as well, with significantly fewer (40 %) new proliferations (Fig. 20).

Ecological Role

One of the smaller *Avrainvillea longicaulis* f. *laxa* colonies measuring 0.6 X 1.1 m in diam. (Fig. 21) and hand-cleaned of debris and epiphytes (mostly unusual forms of *Laurencia intricata*, *Cladophoropsis membranacea* and *Polysiphonia flaccidissima*, Fig. 22) weighed 19 kg. Similar weights were recorded for comparable colonies of *A. asarifolia* f. *olivacea* (Fig. 18). Given that transect studies documented that both *A. longicaulis* f. *laxa* and *A. asarifolia* f. *olivacea* form colonies in excess of 30 m diam. (Fig. 12), their contribution to biomass in Twin Cays ponds is enormous.

Epiphyte-free *Avrainvillea longicaulis* f. *laxa* blades showed a net photosynthetic rate of about two-and-a-half mg C fixed per gram of organic dry mass (ODM) per h, with a dark respiration rate of about half a mg C consumed per g ODM per h (Fig. 23). Twenty newly formed blades (mean area = 50 cm²) contained an average of 12 mg ODM per cm² (28 % C), which converts to about four mg C per cm² of proliferating blade. Given the net photosynthetic production determined above (with the normal inhibitory effects of natural levels of epiphytes) and assuming that this rate could be sustained throughout a 10 h-day, with dark respiration at half a mg C consumed per g ODM per h for 14 h (not including respiration of the pseudo-

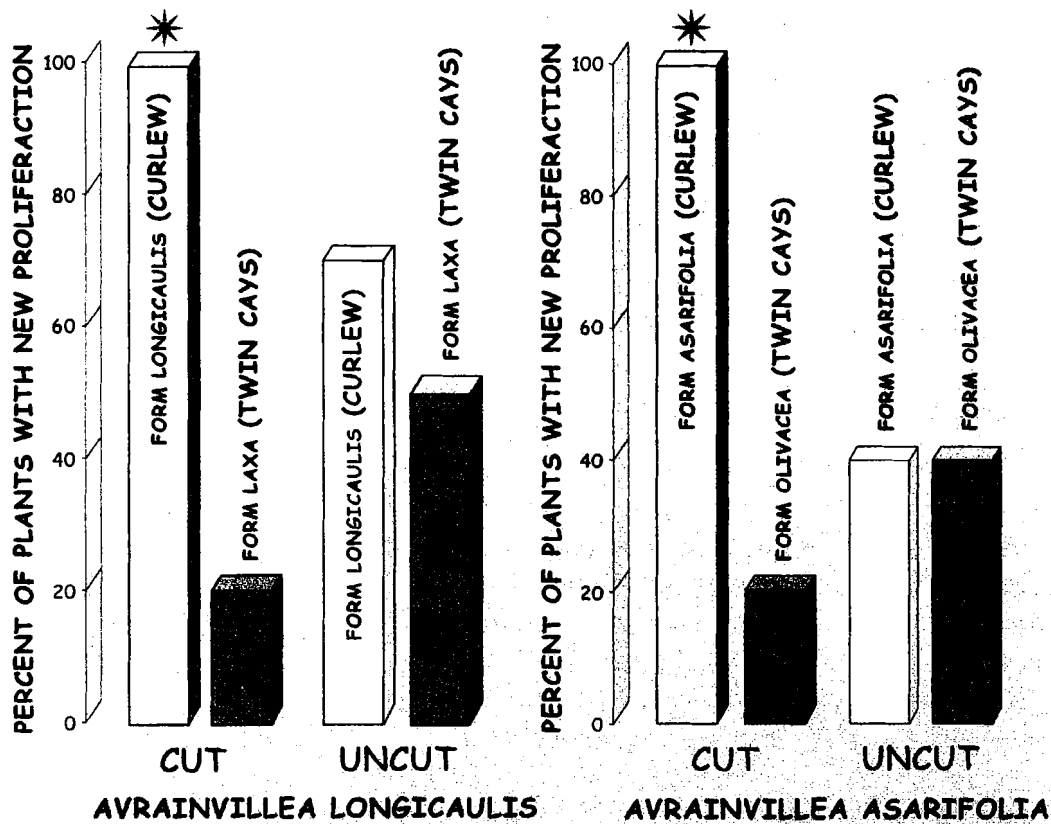


Figure 20. The percent of plants with new proliferations following mutilation (blade decapitation by cutting) after 12 mos. The massive holdfast morphs f. *longicaulis* and f. *asarifolia* from Curlew Cay showed significantly greater proliferation following cutting than the uncut treatments or the uncut and cut colonial mangrove morphs, f. *laxa* and f. *olivacea*. (* indicates significant differences at $P < 0.05$)



Figure 21. This small colony of *Avrainvillea longicaulis* f. *laxa* from Twin Cays weighed 19 kilograms (spun wet weight).

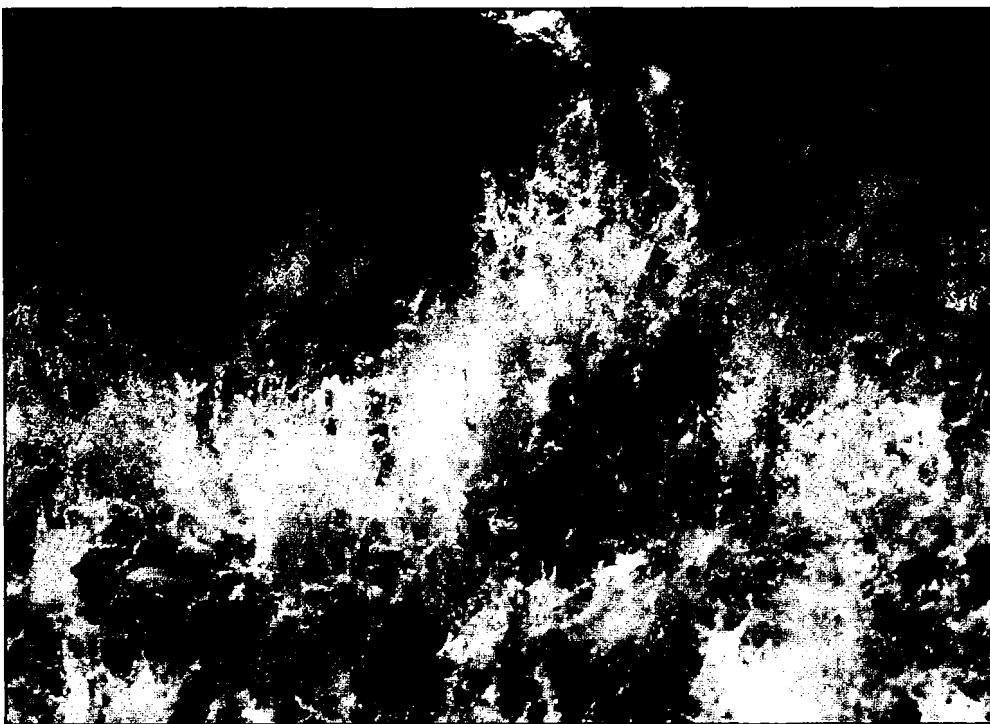


Figure 22. Twenty meter diameter colony of *Avrainvillea asarifolia* f. *olivacea* at Twin Cays showing the extensive coverage of epiphytes (predominantly unusual forms of *Laurencia intricata*, *Cladophoropsis membranacea* and *Polysiphonia flaccidissima*).

rhizomatous mass), we arrived at a net rate of about four mg C per g ODM per day. The wet samples yielded an average dry weight (DW) of about 7% of the WW. When ignited to constant ash weight at 500° C, the organic dry mass averaged 79 % of the DW. Mature assimilators produced about two mg C per plant (single stipe with blade) per day. Based on these calculations, the productivity of an average single mound of *Avrainvillea* at Twin Cays would conservatively yield an astounding 4 kg of carbon fixed per day. The productivity of a square meter of an average mound calculates at 6.2 g of C fixed per day.

DISCUSSION

Taxonomy

Experimental field approaches to macroalgal taxonomic questions are seldom utilized even though the rapid growth of most seaweeds makes them amenable to manipulative techniques. The “common garden” reciprocal transplant experiment provided definitive resolution of the hypothesis that the mound formers (*f. laxa* and *f. olivacea*) were discrete species. Following one year of transplantation, all experimental transplants had acquired morphological features that were consistent with the morphs characteristic of their new habitats (Fig. 24). This result, and the internal anatomical data (Littler and Littler, 1992), supports the hypothesis that the mound forms are not distinct from the solitary forms (*f. longicaulis* and *f. asarifolia*) and, therefore, falsifies the hypothesis that mound-forming colonial taxa are separate species.

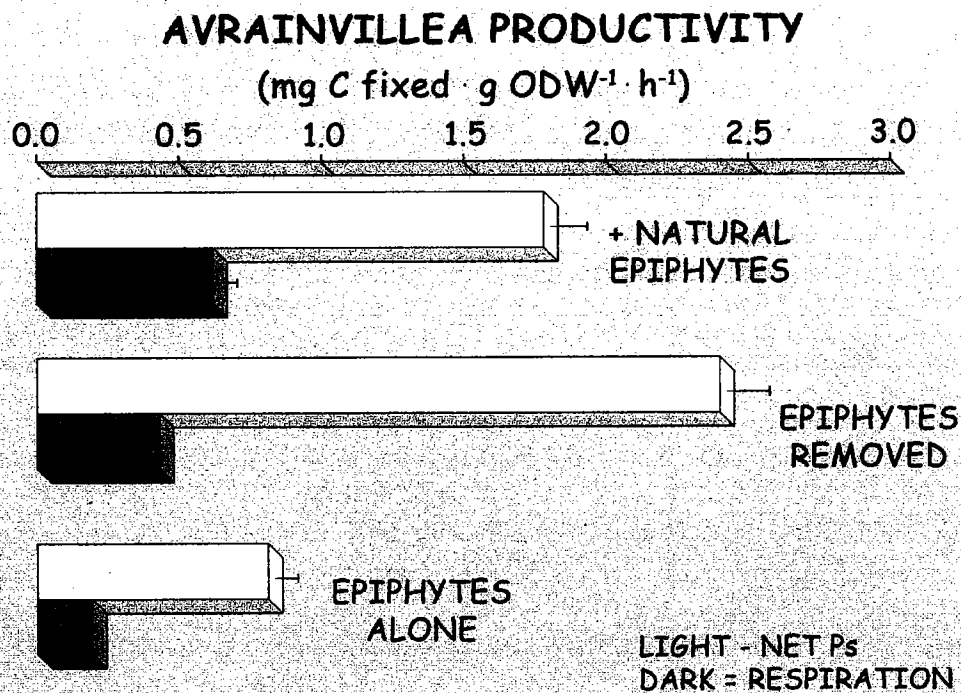


Figure 23. The net primary productivity (light histograms) and respiration (dark) of *Avrainvillea longicaulis* f. *laxa* blades with natural epiphytes, epiphytes removed and epiphytes alone.

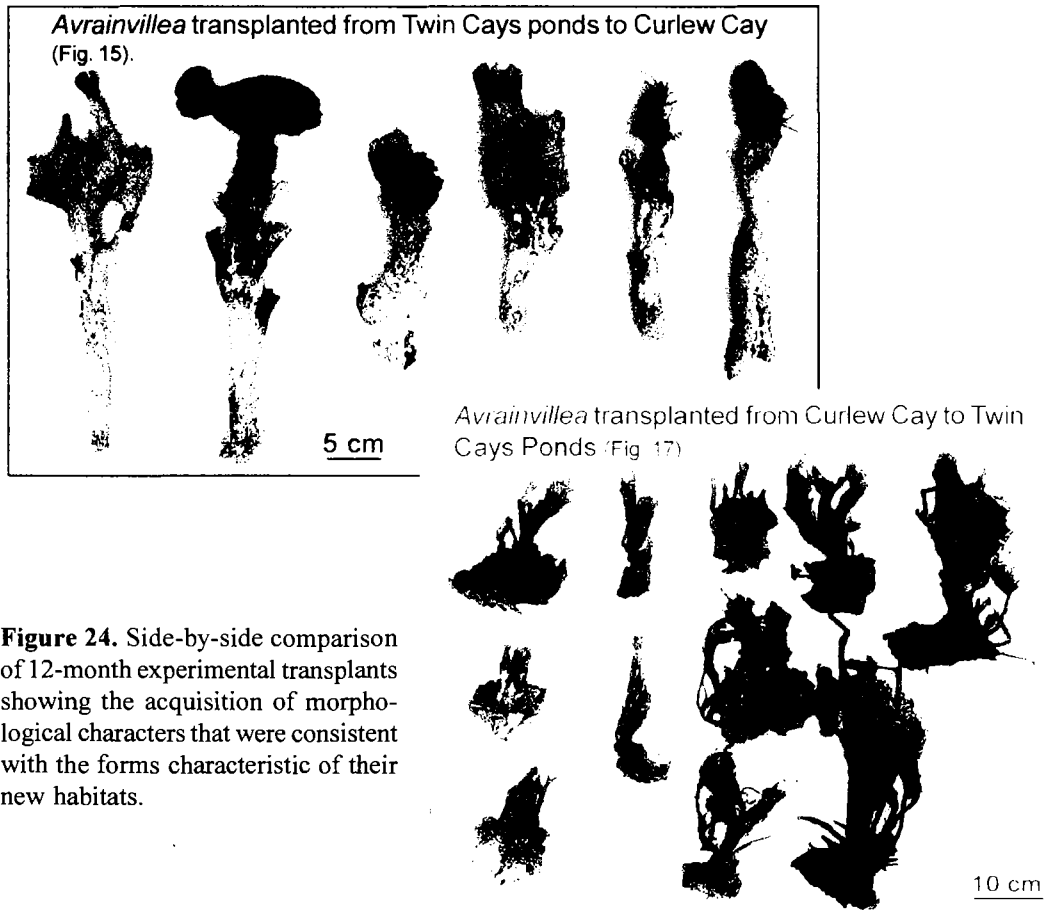


Figure 24. Side-by-side comparison of 12-month experimental transplants showing the acquisition of morphological characters that were consistent with the forms characteristic of their new habitats.



Figure 25. *Avrainvillea longicaulis* f. *laxa* showing tangled jumble of stipes and blades forming extensive mounds in Twin Cays ponds. Note the tangled fused pseudo-rhizomatous stipe structure adaptive for the flocculent anoxic peat substrate.

Coloniality

This study suggests that the ecological attributes of mangrove interior ponds, lakes and creek areas select for the colonial morphs of *Avrainvillea*, not only by providing refuge habitats from the intense fish-and-sea-urchin herbivory (Taylor et al., 1986) that is associated with open-water systems (e.g., Littler et al., 1983; Lewis, 1986) but also by ameliorating the nutrient stresses that frequently occur in such reef- and lagoon-ecosystems. This enables the more delicate colonial morphology to prevail, spreading by means of the unique pseudo-rhizomatous stipe structure (Fig. 25) to cover the otherwise unavailable flocculent anoxic peat substrate.

It is interesting to note that the mound formers, while capable of overtopping other psammophytic (sediment dwelling) organisms, tend to bear prodigious quantities of epiphytes such as *Laurencia intricata*, *Cladophoropsis membranacea* and *Polysiphonia flaccidissima* (Figs. 12, 22), which, given sufficient light, would add about 30% to overall colony productivity in this shallow light- and nutrient-rich environment (Fig. 23). An earlier study at Twin Cays (Littler and Littler, 1985) recorded 17.2 and 13.4 grams of carbon fixed per square meter per day at outer fringe, dense seagrass/algal, bay- and channel-sites, respectively. These values rank among the higher productivity rates recorded and were two-to-three times the production rates of the mangrove pond *Avrainvillea* colonies calculated in the present study. In contrast to the epiphytized colonial morphs, the deeper occurring lagoon morphs have been shown (Littler and Littler, 1999) to actively expel their more harmful epiphyte loads by translocation followed by senescence and shedding (i.e., blade proliferation/ abandonment, Fig. 5).

The theoretical costs vs. benefits of coloniality in terrestrial plants and marine animals (e.g., see review by Jackson, 1977) have received substantial attention. However, consideration of this phenomenon for marine plants previously had been limited to the advantages/disadvantages of the algal-turf morphology (Hay, 1981). In comparison to the extraordinary mound-forming species of *Avrainvillea*, it should be noted that two other genera of Bryopsidales also form knoll-like colonies. *Caulerpa* species, particularly the various forms of *C. racemosa*, can overgrow reef habitats to create small (tens-of-centimeters high), but often extensively spreading, humps. *Halimeda* is unique for the massive (tens-of-meters high) fossil bioherms recorded (Drew 1997) from the Great Barrier Reef lagoon. Although time and resources did not allow us to do comparative functional morphology studies on the two morphs of *A. nigricans* (i.e., f. *nigricans* and f. *spongiosa*), we predict that the findings would have closely paralleled those for the morphs of *A. longicaulis* and *A. asarifolia*.

Perennation

We also showed that the stipes and blades of the open-water morphs (*Avrainvillea longicaulis* f. *longicaulis* and *A. asarifolia* f. *asarifolia*) indeed serve as expendable assimilators, with a major function of building a massive perennating/storage organ, the columnar holdfast (Fig. 11), which comprises the bulk of the thallus biomass (Olsen-Stojkovich, 1985). Among all the other Bryopsidales, *Avrainvillea* is uniquely long-lived (see Littler and Littler, 1992) and does not undergo holocarpic reproduction (Clifton, 1997) leading to death. Physical disturbances (such as storms and herbivory), as well as physiological stresses (such as epiphyte loading), result in disproportionate losses of the relatively delicate expendable assimilators, which can be readily replaced by perennation from the long-lived subterranean holdfast during

more favorable conditions. Selection for this strategy is amply represented in terrestrial environments as shown by the multitude of vascular plants that crown sprout after physical forces such as severe storms, fires, freezes or overgrazing have destroyed the above-ground canopies. However, the only relevant marine example (Heck and Valentine, 1995) is the seagrass *Thalassia testudinum* which is able to compensate for short-term grazing losses on emergent shoots by mobilizing stored carbohydrates from the rhizomes (see Tomasko and Dawes, 1989).

Ecological Role

The advantage of the deeply rooted morphs of *Avrainvillea* in open-water sedimentary seagrass environments, such as Curlew Cay where the water column nutrients are consistently low, lies in the fact that these plants can avoid physical catastrophic losses while tapping into the much higher concentrations of interstitial pore-water nutrients (e.g., >200 $\mu\text{mol N}$, Williams and Fisher, 1985). These findings add a further dimension to observations of nutrient-limited productivity of benthic algae on tropical reefs (Kinsey and Domm, 1974; Kinsey and Davies, 1979; Smith et al., 1979; Hatcher and Larkum, 1983; Lapointe et al., 1987). Conversely, we have shown that colonial adaptations of *Avrainvillea* that take advantage of high nutrient, but anoxic, environments, such as commonly found in mangrove interior creeks, lakes and ponds, result in some of the most prolific communities known. Documentation of such ecosystem level differences in nutritional state and productivity, relative to the functional morphology of the dominant primary producers, is critically needed in the construction of successful models of benthic productivity for tropical marine systems.

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