ALGAE.
AND DENNIS RUSSELL
1970s-1990s
PART 2 of 2

G.H. BALAZS
COLLECTION

Maui algae bloom focus of environmental plan

11/16/92 THE MAUI NEWS

By JILL ENGLEDOW
Staff Writer

WAILUKU — West Maui could become a model for managing a developed watershed area in order to limit the degradation of the ocean environment by runoff from human activities on shore.

Algal blooms that have fouled West Maui waters in recent years provided a focus for a comprehensive plan being developed by the state Department of Health in cooperation with the U.S. Environmental Protection Agency.

The plan is designed to define the problem of seaweed infestation and land-based pollution in the ocean and to minimize the impact of development on nearshore water quality, said Bruce Anderson, state deputy director for environmental health.

No one knows why several species of seaweed have bloomed so heavily in West Maui recently. However, land-based pollution is a major suspect there and where algal blooms have occurred in other areas of the state.

Algal blooms are the problem that focused the planning but "the larger problem is the incremental degradation that may occur without proper planning in that area," Anderson said. The lessons learned in West Maui can be applied elsewhere.

The plan contains a "smorgasborg of ideas," some of which already are being implemented, Anderson said. Others will be made possible with congressional appropriations sought by Sen. Daniel Inouye, D-Hawaii.

The EPA has said it will pass along most of a $400,000 appropriation to the Health Department. Another $450,000 was appropriated for the National Oceanic and Atmospheric Administration. How that money will be used hasn't been determined yet, but the plan anticipates those funds would be used for some of its proposals.

Among the proposals is for EPA and NOAA funds to be used to pay a full-time project coordinator for a period of two years. The coordinator would chair the West Maui Watershed Management Committee, which would be an expanded version of the existing West Maui Algal Task Force.

This person would work with the committee and with county, state and federal officials to plan and manage research projects, keep track of all projects and information gathered, and keep everyone informed.

The coordinator and the committee also would develop a management plan for selected West Maui watersheds to translate research findings into management projects.

A couple of watershed management plans are under way on Oahu, but the proposed Maui plan would be more comprehensive, Anderson said. It would look at a number of possible pollution sources, not just a specific source.

The task force has been working on the algal bloom problem for the past year, and it will be asked to comment on the proposal. The proposed permanent committee would be formed somewhat as the task force is, with membership from government, scientific and other affected sectors working together.

Other projects included in the proposed plan:

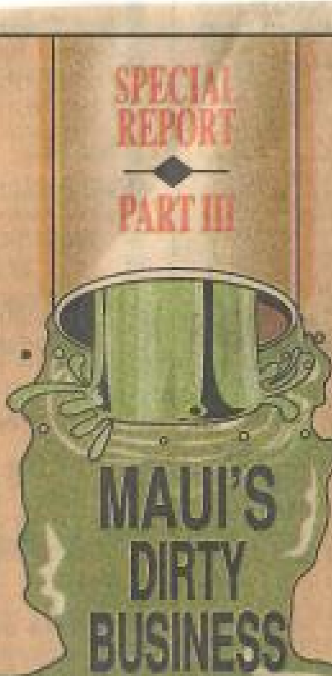
■ A preliminary nutrient fate study, beginning this month with EPA funds, focusing specifically on the effluent from sewage injection wells at the Lahaina Wastewater Treatment Plant.

■ Mapping locations of seaweed species and studying their relationship to the ocean environment, beginning in January with state funds.

■ Geographic Information System, an ongoing in-house DOH project to map selected subwatersheds in West Maui and monitor their groundwater, rainfall and other features.

■ Reviews of fertilizer application on agricultural lands, golf courses and other areas, measurement of nutrient runoff entering nearshore waters and aerial infrared photography surveys of coasts along Lahaina, Kihei and Kahului, where major seaweed blooms have occurred.

6/24/93 Honolulu Star-Bulletin A1



Algae tainting crystal waters

☐ Residents want an end to the green growth

Third in a series

By Peter Wagner
Star-Bulletin

IT'S ugly, it's slimy and it stinks. But where does Maui's awful algae come from?

Shore-front property owners from Kahului to Kaanapali don't really care. All they want is an end to the rotting "blooms" of algae that for three years have chased tourists and residents from otherwise idyllic shores.

The mystery, now under intense study, points strongly in the direction of four injection wells at the county's Lahaina sewage treatment plant as a major source of nitrates apparently feeding the overabundant blooms.

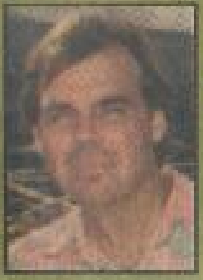
Nearby agricultural fields, another suspect in the case, may be a tougher problem to control, putting the county's wells on the hot spot.

Snorkelers accustomed to Kaanapali's crystal waters are finding a murky wall of seaweed smothering coral reefs. A Lahaina tour company has gotten rid of its glass-bottom submersible. And residents are paying thousands of dollars to have tons of smelly algae bagged and hauled away.

Whatever is happening, environmentalists say, nature is seriously out of whack.

"It used to be beautiful, just like you see in the ads," said Eve Clute, who moved to Napili 14 years ago for the pristine beaches. "It got so I couldn't go to the beach because it was filled with flies and hydrogen sulfide gas."

See **ALGAE**, Page A-6



> *"It's still not clear why we have problems with algae in the area."*
>
> **Bruce Anderson**
> Deputy director, state Health Department

Star-Bulletin

Kaanapali resident Eve Clute scoops up some of the algae Hypnea in Napili's Alaeloa Cove. See story, Page A-6.

ALGAE: Maui waters tainted by blooms

Continued from Page A-1

The beach buildup is more than a nuisance. Rotting algae releases hydrogen sulfide gas, a toxic substance that some say is making beach goers nauseous. And as the algae decomposes, it puts nutrients back into the water to fertilize the next crop.

The problem in Kihei is so bad that hotels for years have paid a contractor to bulldoze beaches, a practice Clute said is carrying away sand.

The unsolved mystery

Something is happening, biologists say, but what? Among the unsolved mysteries is why the algae, predominant in spring and summer, seems to come and go. If nutrient sources are year-round factors, why isn't the algae?

Last summer a heavy crop turned away tourists and spoiled real estate deals. But recently, it has been difficult to find the offending algae on most Kaanapali beaches.

One exception is Alaeloa Cove, a tiny enclave on the Napili coast that stays clogged with algae most of the year.

"It's still not clear why we have problems with algae in the area from time to time," said Bruce Anderson, deputy state Health Department director. "Right now, there isn't an algae problem in the area as far as I know, yet the use of fertilizer hasn't changed. There certainly are other factors to look at besides nutrient loading."

Those could include seasonal changes in sunlight, currents and wave action, he said. But with the heaviest concentrations of algae often found near sewage injection wells, the evidence is incriminating.

That near-shore waters from Lahaina to Kapalua are high in nutrients from land-based activities is undisputed. A soon-to-be-released study funded by the Environmental Protection Agency points to shore-line sewage injection wells in Lahaina as a prime source of nutrients in the area's water. Other sources identified include fertilizer runoff from nearby sugar and pineapple fields.

No clear connection

William Magruder, a marine botanist at the Bishop Museum, said algae is everywhere and will bloom where nutrient levels in the water are high. Like a backyard garden, he said, the sea's plants respond to sunlight and other growing conditions.

But making a clear scientific link between the nutrients and the algae is a big step that biologists and governmental regulators aren't ready to take.

Scientists studying the problem haven't ruled out that the blooms may be natural.

Like the coral-eating crown of thorns starfish that swarmed into Hawaiian waters 20 years ago, Maui's algae could simply pick up and leave without explanation.

Still, it's hard to ignore the county's sewage treatment plants and their injection wells. All three plants, in Kahului, Kihei and Lahaina, are putting millions of gallons of nutrient-

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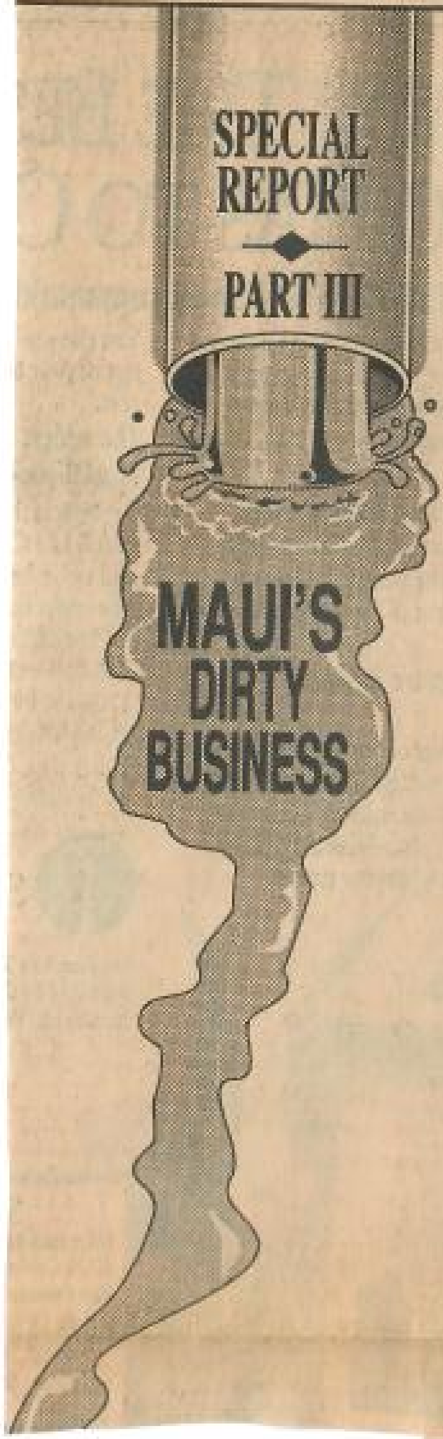
TOMORROW: Recycling



Seaweed tangles clog isle cove

☐ Property owners say the problem is getting worse

By Peter Wagner
Star-Bulletin

IT'S a cozy little piece of paradise, much farther from the world than its electronic gates and neatly clipped homes suggest.

There are broad green lawns, a shady clubhouse and a private cove that seems under a sultry spell.

But Alaeloa Cove, a dimple in Maui's Napili coast, has not been a happy place lately. For some reason, the island's on-again, off-again algae bloom has taken up permanent residence in the cute little cove, turning nirvana into a festering garbage pit.

"It stinks like a sewer in the summer," said Wally Working, a resident tired of wrestling with relentless seaweed.

In April, when beaches from Lahaina to Kaanapali were enjoying a respite from the invasion, Alaeloa Cove was choked with Hypnea, a reddish-brown seaweed with a powerful smell. On the beach, tons of the stuff was baking in the sun. In the ocean, a mass of matted algae heaved heavily in the currents.

Last summer, three times as much algae had piled up each day, turning the cove into a reeking caldron of decay.

"I don't want to sound like an ogre, but you can see we have a problem," Working said. "Someone should come in and bag it up and take it away and then find out where it comes from and stop it," said Working, president of the community's association.

According to Working, things have gotten worse in the past three years. Four years ago, he said, the cove was crystal clear. Now residents can no longer go for a swim.

County officials have been hauling away what residents pay to have bagged three times a week, a $15,000- to $25,000-a-year project that doesn't sit well with the Alealoa Community Association. But no one's gathering up the algae in the water, which the county says is a state jurisdiction.

While scientists study the problem under federal and state grants, residents have their theories. An open drainage ditch that feeds the cove this winter was a raging river of red dirt. The only obvious source of such runoff, Working said, is nearby pineapple fields.

Photographs taken during recent winter rains show the cove turned red.

Another possible suspect, the county's sewage injection wells in nearby Lahaina, could also be overfertilizing the sea, Working believes.

Whatever the case, residents of Alealoa want an answer.

"We need to get to the source of the problem without studying this thing to death," Working said.


Star-Bulletin

More than 20 tons of algae pile up in Alaeloa Cove weekly, costing residents $15,000 to $25,000 a year to have it hauled away.

rich effluent into injection wells each day.

Unlike ocean outfalls that carry effluent far from land, the 380-foot wells go directly into the ground for dispersal. Recent studies suggest the effluent may be riding out on an underground tide.

An emotional issue

The EPA's $117,000 study, by California-based Tetra Tech Inc., said the Lahaina wells are putting high levels of phosphorus into nearby waters.

The same study found nitrogen fertilizer from agricultural fields in equally high amounts. The study will be followed by dye tests to find the leaks.

Not everybody is ready to get on the anti-algae bandwagon. Mayor Linda Crockett Lingle wants to be sure that sewage indeed is the culprit before pulling up injection wells.

"We don't want to make the problem worse, but there is no scientific evidence connecting the wells with the algae," Lingle said. "I think it's just more complicated than people realize. It's become a very emotional issue."

Lingle took a ride on the commercial submarine Atlantis last year and saw little evidence of a problem in 100-foot-deep waters.

But operators of the Nautilus, another submersible that until recently cruised Kaanapali's more shallow waters, said Lingle missed the boat.

"If she'd gone out with us the same week she did on the Atlantis, she'd have seen algae so thick she wouldn't have believed it," said Kahren Bellamy, spokeswoman for Hawaiian Reef Divers. The problem is in water less than 30 feet deep, she said.

So bad was a bloom just outside Lahaina Harbor late last year that Bellamy's company was losing customers. "It was killing our business," she said.

The Lahaina bloom "seems to have moved progressively south in the last few years," Bellamy said.

Video footage taken by recreational divers in 1991 shows staghorn corals dying in shallow waters off Kaanapali. The area's reefs were shown buried under a thick blanket of green algae the year before.

A major economic threat

Meanwhile, Anderson and Lingle have locked horns over the county's wells.

"They've been focusing their efforts on vindicating those injection wells," Anderson said.

But Lingle's cautious attitude is understandable. As caretaker of a relatively small county budget, she's got to juggle other costly problems, including road repairs and impending landfill closures.

Coming up with an alternative to injection wells looks to be another expensive proposition, and Maui still needs a place to dump the discharge from a system fed by 130,000 residents and tourists.

Whatever the cause, Magruder said, Maui's algae blooms pose a major economic threat.

"Hawaii spends a lot of money promoting its environment, especially its marine environment," he said. "I worry about the tourist's image of Hawaii."

Property owners lobbied the state Legislature this year to push for a solution and an immediate beach cleanup.

But while state and federal lawmakers have put more than $1 million into studying Maui's algae problem, none is earmarked for cleanup. A resolution was passed giving the county responsibility for algae on shore and the state for nearshore waters. But, strapped for money, the county has agreed only to haul away what residents pay to have bagged.

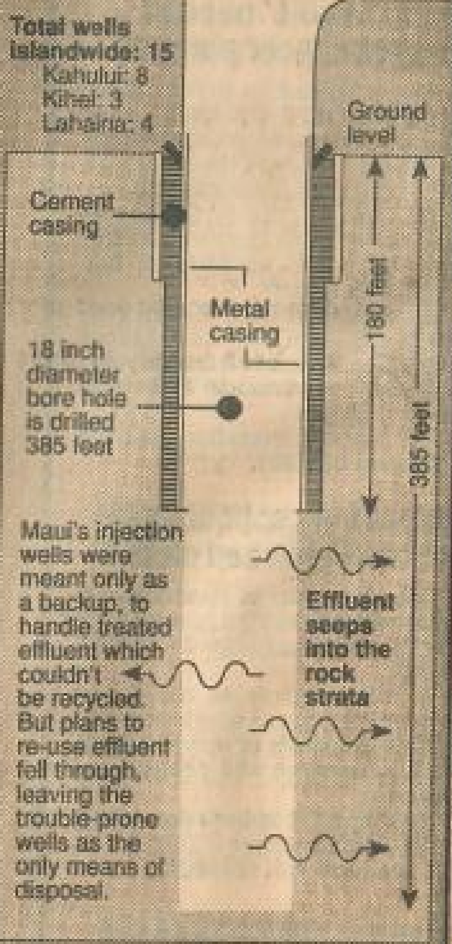
While the algae has brought a pile of worries, it has also triggered something good, Lingle reflected.

"It focuses people's attention on the fact that water is a valuable commodity," she said.

Meanwhile, west Maui residents are clamoring for a cleanup.

Injection well

Total wells islandwide: 15
Kahului: 8
Kihei: 3
Lahaina: 4



Maui's injection wells were meant only as a backup, to handle treated effluent which couldn't be recycled. But plans to re-use effluent fell through, leaving the trouble-prone wells as the only means of disposal.

By Joan Bossert, Star-Bulletin

ffluent could solve Maui's sewage and water woes.

February 13, 1980

Dr. Dennis J. Russell
Miller Science Learning Center
Seattle Pacific University
Seattle, Washington 98119

Dear Dennis:

Many thanks for your recent note and the results of the first set of turtle gut samples that I previously sent you. At your earliest convenience, I look forward to receiving the results of the second batch, as well as return of the actual samples for storage in my permanent collection. Incidentally, I noted in my latest computer printout that you have not yet requested payment from the U.H. Research Corporation. Hopefully you will do so in the very near future. I do indeed have another group of samples, but there will only be about 25 vials in the shipment. I will write out an additional Purchase Order in the near future.

I noted with interest that GB-425 may be *another* species of *Codium*. This is exciting. Have you made any progress on the *Codium* publication?

Dennis, if young green turtles live in the pelagic environment until they are approximately 35 cm in length, would you expect them to acquire algal growth on the shell and skin during this period? On a number of occasions I have observed and captured turtles 35-37 cm living close to shore that were exceptionally "clean" - that is, free of algal growth. I suspect that these turtles are new recruits from the pelagic environment - recent arrivals that have taken up residency in the shallow water foraging pastures. Does this sound reasonable to you? What factors would prevent young turtles from acquiring algal growth in the pelagic environment? Any assistance that you can provide in this important matter would be greatly appreciated.

Best personal regards to you and family.

Sincerely,

George H. Balazs
Assistant Marine Biologist

GHB:md

Enclosure

March 4, 1980

Dr. Dennis J. Russell
Miller Science Learning Center
Seattle Pacific University
Seattle, Washington 98119

Dear Dennis:

I appreciated your letter of 26 February, which was my birthday of 37 years.

Algae seems to be in the news a lot lately, as you will note from the attached article.

Your comments on possible algal colonization of young turtles in the open ocean provided me with some valuable insight. However, I am still wondering if the probability for such colonization would not be far less than for a turtle living in a nearshore algal rich environment. Concerning sloughing, yes small particles of shell and skin are continually coming off. If comparatively rapid growth is occurring under pelagic conditions, than I suppose shell and skin sloughing would also be taking place at a faster rate. As far as secretions are concerned, I am not aware of any such action in sea turtles. I'll look into this further. Yes, I would indeed like to have a copy of the bibliography on the subject of colonization.

I have also enclosed a Xeroxed copy of the RCUH purchase order which I just wrote today. This should cover the samples I will be sending in a few days (GB series 600), as well as return postage and some pressed specimens I would like to have you look at. Dennis, please request payment for this amount immediately, as my funds are rapidly drawing to a close. You didn't mention it in your letter, but I certainly hope you requested payment on the earlier outstanding purchase order.

Best regards - looking forward to hearing from you soon.

Sincerely,

GEORGE H. BALAZS
Assistant Marine Biologist

GHB:ec
Enclosures

14 April 1980

Dear George,

Here are the references I promised you:

Bibliography concerning the algae that have been found growing attached to turtle shells.

Hollenberg, George J. 1968. An account of the species of the red alga *Polysiphonia* of the central and western tropical Pacific Ocean. II. *Polysiphonia.* Pac. Sci. 12(2): 198-207.

Jackson, Crawford G., Jr. and Arnold Ross. 1971. Molluscan fouling of the ornate diamond-back terrapin, *Malaclemys terrapin macrospilota* Hay. Herpetologia 27(3): 341-344.

Jackson, Crawford G., Jr., Arnold Ross and George L. Kennedy. 1973. Epifaunal invertebrates of the ornate diamondback terrapin, *Malaclemys terrapin macrospilota*. Am. Midl. Nat. 89(2): 495-497.

Kitami, Takehiko and T. Koseki. 1971. Supplementary notes on the foreign barnacles found along the coasts of Niigata and Sado Island. Collect. Breed. (Tokyo) 33(7): 1970-1971.

Ross, Arnold and Crawford G. Jackson, Jr. 1972. Barnacle fouling of the ornate diamondback terrapin, *Malaclemys terrapin macrospilota*. Crustaceana 22(2): 203-205.

Tsuda, Roy T. 1965. Marine algae from Laysan Island with additional notes on the vascular flora. Atoll Res. Bull. 110: 1-31.

I am supposed to have another reference for Kitami (1972) and will have to look it up in one of my later editions of my literature review. I will send that reference to you if it is not just a typo in the edition I am looking at now.

I have mailed all of the samples back to you except for the latest ones on cards and in vials (GB 600 series) and one sample No. 533 which I have kept to tag with their names so you will not have to send them back after they are pressed. Don't worry about the samples, they should be arriving your way soon. I sent two packages about a week apart from each other. Yes, please send more samples and do you want me to send the bill in as soon as I get it? I assume you do and then I will wait for the samples to arrive afterwards.

It was interesting to see Hypnea musciformis in the Waimanalo Bay sample (GB 533) since this alga was confined to Kaneohe Bay alone when I left Hawaii. It is known to spread like made in Florida, the place it originally came from. That alga and A. spicifera were the only two exotics on the list. Hypnea musciformis is the only alga in Hawaii with the prominent hooks on its branches, so it is very easy to identify. Let me know if you find it anywhere else.

Thank you for the V. B. Vozzhinskaya article on free-floating plants in the Pacific. When I begin writing on the Codiums you found on floats I will send you a rough draft and we can complete it together. The Hypnea information will probably go into a paper that I hope to present in Scotland in August 1980. I will mention the fact that is seems to be spreading rapidly, but it will not be enough for a separate publication. It would, however, be a good place to mention your research and your grant numbers. Please send that information or wait until I send you the paper, before I present it, so you can add what you wish.

Sincerely,

Dennis J. Russell

SEATTLE PACIFIC UNIVERSITY
SEATTLE, WASHINGTON 98119

February 26, 1980

Mr. George H. Balazs
Assistant Marine Biologist
Hawaii Institute of Marine Biology
P.O. Box 1346 (Coconut Island)
Kaneohe, Hawaii 96744

Dear George:

Thank you for the letter and the news clipping. I also enjoyed reading the article you sent last time concerning the sea turtle research symposium. I will finish the latest samples (GB 501 - 532) by the end of March at the latest and then will send the samples to you. I will send the first set of samples this week, however, rather than waiting to send them all together.

You asked me if I would expect algae to grow on very young turtle shells of animals that are pelagic. A great deal of my literature review was concerning "fouling" of many kinds of organisms and surfaces in the sea and there are several factors that would tend to keep a turtle shell clean. Algae would have a difficult time remaining attached to a shell surface which is being sloughed-off rapidly, or one that was secreting a mucus or other slick substance, or a toxin, or inhibitor. Even a slight secretion could prevent "fouling" or what I call colonization of the shell. Does the shell have an epithelium? Would you like me to compile a bibliography on the subject? I would not use clean shells as an index of new recruits from the pelagic environment without further considerations. Send those 25 samples and I will finish them by March 31, 1980 - Promise.

undergoing rapid growth?
consider young in captivity?

Sincerely yours, Dennis J Russell

UA can be considered. DLNR parks forestry divisions will submit a new UA proposing to establish heliports so copter use can be regulated. In the ntime, a limited temporary permit been issued to the two operators, continue their Na Pali landings and s.

—Environmental groups, hunters and r residents filed suit in May against helicopter operators, the DLNR and hairman, charging that the EIS inade-ely discusses impacts to the rich his-al artifacts along the coast and to the onment—particularly to such wildlife e endangered shearwater which nests e area.

The suit calls for revocation of all which calls for another permit to regulate the helicopters' impacts on the shoreline. Following a June hearing, the County Planning Commission approved the permit.

PUBLIC PROS AND CONS

Testimony at the SMA hearing leaned in favor of the helicopters operations. Several people, however, felt the growing numbers of people brought to Na Pali valleys by the helicopters would diminish the experience of wilderness.

"There are areas in the Na Pali coast that should not be accessible by helicopters," one resident said. "We're talking about an area that's not a large area but certainly one of the most unique places in the U.S. We've seen the pressures on Kalalau."

Kalalau Beach: end of the Na Pali trail and destination for backpackers and other visitors.

Proponents of continued tours to the area included helicopter operator Richard "Red" Johnson.

"We are not asking to increase business, that is, the number of people using the park areas, but to maintain the services we have historically provided the visitor, the community and the government," Johnson said.

MANAGEMENT PLANS

The County's approval of the SMA permit hinges on the operators' compliance with a temporary plan aimed at balancing the Na Pali use with resource preservation. DLNR officials adopted this "interim management plan" which took effect in January and will remain in use until the State parks division completes the permanent, more detailed plan in about a year.

In 1975, when the DLNR began issuing permits to campers in Kalalau Valley, about 2,000 people used that area. In 1978, however, about 10 times as many campers used Kalalau.

In order to help the Na Pali area sustain such population pressures, the interim plan calls for gathering more information on the environmental, historical, and recreational resources—studies now in progress.

The plan's objective on public access, however, may influence permit decisions in the helicopters case. This objective provides for access by helicopter and boats to Kalalau, Nualolo Kai, Miloli'i and Polihale valleys, as well as from Ha'ena to Kalalau Valley via existing trails.

Other objectives include: limiting the density of use at any one time; preservation of archaeological sites by discouraging public use of historically sensitive areas, and by placing qualified areas on the State and national historic registers; preservation of marine life, ecosystems and scenic values by regulating hunting, fishing and building in these areas; and restricting heavy use to areas with great recreational potential and minimal risk to resources.

No matter how government manages the Na Pali area, public reaction remains split in two fairly distinct categories: those who prefer leaving the coast alone and those concerned that more people should have access to the region.

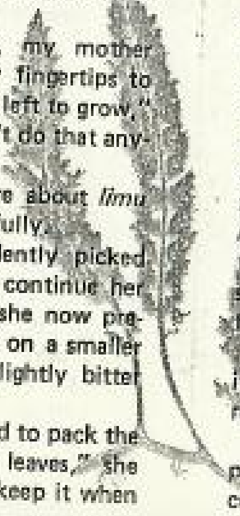
PEOPLE

Limu kohu — a family tradition

Anyone living on the northeast coast of Kaua'i will encounter the salty, reddish-purple seaweed called *limu kohu*, but probably not to the degree that Pearl Rapozo has.

Residents of the area regard Rapozo and her sisters as keepers of a *limu* tradition practiced most actively by their late mother, Daisy Lovell.

Rapozo stooped on the wave-washed reef flats in Anahola Bay to pick a *limu kohu*.



"When I was a girl, my mother taught me to use just my fingertips to pick *limu*, so the 'roots' are left to grow," she said. "Most people don't do that any more.

"I wish I learned more about *limu* from her," she added regretfully.

However, Rapozo evidently picked up enough information to continue her mother's business, though she now prepares and sells the product on a smaller level, giving some of the slightly bitter condiment to friends.

"In the old days, we used to pack the *limu* in cans lined with *ti* leaves," she said. "That was enough to keep it when we shipped it to another island."

Today, Rapozo cleans the coral and sand from the *limu*, soaks it to diminish the iodine taste, and squeezes it in the traditional way, but has gone the modern route of refrigeration and packing in plastic bags. She pulled out a bag filled with a snug roll of prepared *limu kohu*, picked during a particularly fruitful period in July.

In ancient times, this seaweed's distinctive flavor set it apart, and *limu kohu* was reserved for royalty. Now, if one braves the risk of coral cuts and the sometimes vigorous wave action the plant requires, the field is open. Many people actually prefer donning their own rubber *tabis* (shoes) to gather their private *limu kohu*, rather than running to the store as they do for many other seafoods, Rapozo said. This stems in part from personal preferences in preparation of the seaweed.

But *limu kohu*, always an expensive item, has not escaped inflation—another reason to "get your own."

"I've seen it selling for four dollars a pound," she said. "I remember when it cost twenty-five cents."

Hawaii Coastal Zone News Aug 1979

Received 11 February 1980

George,

I will have a light teaching load after March 1980. Send more samples and I'll work them up before June.

I'm now working on your second shipment.

Dennis

SCHOOL OF NATURAL & MATHEMATICAL SCIENCES
SEATTLE PACIFIC UNIVERSITY
(206) 281-2140 - SEATTLE WASHINGTON 98119
Dennis J. Russell

28 Nov 1978

Dear George,

Thank you for your letter of the 21 November 1978. How many samples do you have this time? I will be glad to identify them for you, just send them to Miller Science Learning Center, Seattle Pacific University and I'll get to them post haste, since I may have a bit of a vacation in Dec-Jan. I have been thinking about the Codium paper or note that should be written and will be getting to that as soon as I have sent off my dissertation (hopefully in final final form) to my dissertation committee.

I do not know what the future holds for me, but SPU is a good school and 80% of the faculty are tenured and most of those have been here for 20 years. The pay is good, working conditions are good, the people are open to each other and very helpful. They have a marine laboratory on Whidby Island, and another one beginning on Blakely Island. You may have heard of Dr. Ron Phillips, a sea grass man that is working with Dr. Ken Bridges. Dr. Phillips has been at SPU for about 15-20 years. There is only one trouble -- it isn't Hawaii! Leaving the islands was just about the hardest thing I have ever had to do. Take good care of yourself George and thanks for writing.

Sincerely,

Dennis J Russell

Dennis J. Russell

Mr. and Mrs. Dennis Russell
4408 - 237th Pl. S. W.
Mountlake Terrace, Wa. 98043

4 November 1979

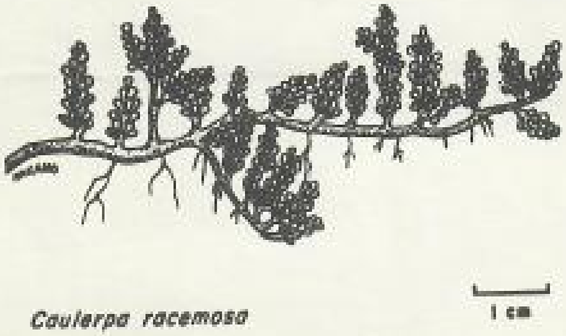
Dr. George,

This is just a short note to let you know that I have received your latest box of samples.

Dennis

THE LIMU EATER. A COOKBOOK OF HAWAIIAN SEAWEED. Heather J. Fortner, 115 pp, November 1978. $4.75. UNIHI-SEAGRANT-MR-79-01. Sea Grant Publications, 2540 Maile Way, Spalding Hall 253, Honolulu, HI 96822.

A cookbook was created as a result of a study of Hawaiian seaweed, or limu. *The Limu Eater* introduces a food that is unfamilar to most and identifies where it grows, what it looks like, how it is used, and where it can be found. In addition, it describes some of the ethnobotanical uses of local seaweed such as for medicine or in ceremony. Thus, what started out to be just a collection of recipes has become an introduction to the art and craftmanship of limu eating. The recipes are from several sources. About a third of the 86 presented have already been published either locally or nationally.



Caulerpa racemosa

SCHOOL OF NATURAL & MATHEMATICAL SCIENCES

SEATTLE PACIFIC UNIVERSITY

(206) 281-2140 · SEATTLE WASHINGTON 98119

J. H. Balazs
HIMB
P.O. Box 1346
Coconut Island
Kaneohe, Hawaii
96744

29 October 1979

Dear George,

I've received your note indicating that more samples will be coming my way. Thank you for your confidence: the ID of your last samples will be done soon. Tomorrow I'm sending my final copies of my dissertation to the Botany Department. I hope this will be final.

aloha,
Dennis

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Hawaii Institute of Marine Biology
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17 July 1979

Dear George,

I have just returned from teaching a Marine Biology course in Sitka, Alaska, and will begin on the ID of your samples forth with. I have received 20 specimen jars and 31 vials plus the one jar from the float. Thank you for sending them as wet specimens, I feel more sure when they have not been reconstituted.

Sitka was quite a bit different from Hawaii both with the low temperatures and cloudy skies, however, the islands, natives and slow pace of life was refreshing. I was teaching at a small, but old, college (Sheldon Jackson College) and will be making plans to teach there again sometime in the future. I am nearly finished with the final polish on my dissertation and have the Codium paper of ours next on the list. Say hi to Al for me!

Dennis

THE POTENTIAL FOR ADDITIONAL MARINE CONSERVATION DISTRICTS ON OAHU AND HAWAII

by

William J. Kimmerer
Woodrow W. Durbin, Jr.

Sea Grant Technical Report
UNIHI-SEAGRANT-TR-76-03

December 1975

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APPENDIX I. LIST OF ALGAL SPECIES BY SITE AND HABITAT. (For site and habitat abbreviations, refer to pages 23 and 24.)

Relative abundance indices are as follows:

4: Abundant
3: Common
2: Uncommon
1: Seen in the habitat but not counted on any transect

	KE-IN	KE-MR	KE-OR	KO-IN	KO-MR	KO-OR	PK-IN	PK-MR	PK-OR	HO-IN	HO-MR	HO-OR	HA-LO	HA-CO	HA-SA	HA-SH	KA-LO	KA-CO	KA-MK	KA-PV	KA-DP	MP-PL	MP-LG	MP-DP	PP-CA	PP-TP	PP-BE	PP-BO	PP-DP	KB-PR	KB-BP	Kapapa
Phylum Chlorophyta																																
Boodlea composita													1					2						3			1					
Hormetella sp.																2																
Caulerpa sp.	1	1						1	1		2						1															
Cladophora sp.													1			2																
Cladophoropsis sp.																				1												
Codium arabium													1															1	1			
C. edule																								3					1		3	
Dictyosphaeria cavernosa		1	1	1	1	1		2	2		1	1	1	3		2		1	1				1				1		1	4	3	
D. versluysii														1				1	1	1			1			2	2	3		3	4	
Enteromorpha sp.													1														1					
Halimeda discoidea													3	3				2	1	3	1	1	2	3				3	3	2	3	
H. opuntia		1	2		1	1	1	2	2		2	2																				
Microdictyon sp.		2						1	1		1																					
Neomeris sp.									1				3			1			1		1	2	1	3								
Ulva lactuca																										3	3					
U. reticulata																												1				
U. sp.							1																									
Valonia ventricosa		1	1		1	1		1	1		1	1																				
Phylum Phaeophyta																																
Chnoospora sp.																											3					
Colpomenia sinuosa																											3					
Dictyopteris australis		2			1						2		1		2							3	3									
Dictyota sp.					1								3	1	2	3		2		2	2	2	4				3	2	1		3	
Ectocarpus sp.													2			3						2										
Padina crassa																											3					
P. japonica																1				1	1	1					3	1	2			
Sargassum sp.								1			2																3					
Turbinaria ornata				2	1	1	1				1	1				3						1	3					3			3	
Zonaria hawaiiensis													1														2					
Phylum Rhodophyta																																
Acanthophora spicifera																										3	3					
A. sp.							2																									
Ahnfeltia sp.																											2					
Amansia glomerata												1	4	4		3	3		3	3			3					4	3			
Asparagopsis sp.													3	2										3								
Ceramium sp.					1		1				1			3																		
Chondrococcus sp.													3	3				3	2		1		1					3	1	2		
Corallina sp.																2												2			3	
Dasya sp.																															2	
Desmia hornemanni						1		1	1		2																					
Galaxaura sp.		1	2		1		1	1	1		1	1	2			3		3	2		2			3		3		3	3			
Gelidium sp.																											3					
Halymenia sp.																			1			1						2	1			
Hemitrema sp.		1									1	1				2				3			1	3				3	2			

APPENDIX I. LIST OF ALGAL SPECIES BY SITE AND HABITAT (continued).

	KE-IN	KE-MR	KE-OR	KO-IN	KO-MR	KO-OR	PK-IN	PK-MR	PK-OR	HO-IN	HO-MR	HO-OR	HA-LO	HA-CO	HA-SA	HA-SH	KA-LO	KA-CO	KA-MX	KA-PV	KA-DP	MP-FL	MP-LG	MP-DP	PP-CA	PP-TP	PP-BE	PP-BO	PP-DP	KB-FR	KB-RF	Kapapa
Phylum Rhodophyta (continued)																																
Hypnea sp.													1										1									
Jania sp.		2	3		2			1		2	3	2		2		2	2	2														
Laurencia sp.												1	1	2						2							1	3	3			
Liagora sp.													1			2				1		1		3								
Porolithon sp.													4			2	4	2	4	2		3	4	3			3	4		3	3	
Spyridia sp.																												1				
Trichogloea sp.																			1													
unspecified encrusting		2	3	2	3	3	3	3	4	4	3	3	2									3	3		3		3	3		3	3	
Phylum Cyanophyta																																
Lyngbya sp.				1	2	1	2	2	2		2																					
unidentified filamentous																			1							4						

ECOLOGICAL RESPONSE TO SEWAGE STRESS AT MŌKAPU, O'AHU, HAWAI'I

WATER RESOURCES RESEARCH CENTER • UNIVERSITY OF HAWAII
2540 Dole Street, Holmes Hall 283/Honolulu, HI - 96822

PROJECT BULLETIN

Project Bulletin No. 18/June 1979

by Mr. Steven J. Dollar, and Drs. Anthony R. Russo and E. Alison Kay

(Editor's Note: Mr. Steven J. Dollar is a WRRC consultant, University of Hawaii; Dr. Anthony R. Russo is an instructor of Mathematics and Science, Leeward Community College, Pearl City; and Dr. E. Alison Kay is Acting Dean & Acting Director of Research, and professor of General Science, University of Hawaii. An interim progress report will be published as Tech. Rep. No. 122.)

NATURE OF REPORT

A study of the Kailua Bay area was conducted in 1973 (Kay et al. 1973) to examine the benthic biota in a baseline study from Mōkapu Pt. to Mokulua Island off Lanikai. During December 1975, prior to the completion of an outfall with a discharge of secondary effluent, a study was made of the benthic marine community at five stations from Mōkapu Pt. to Alāla Pt. north of Lanikai (Russo et al. 1977).

The outfall, constructed for the City and County of Honolulu, began operation in December 1977. In February 1978 the Kailua Bay outfall was connected to the Mōkapu line, and in May of the same year the Kane'ohe Bay outfall was also diverted to Mōkapu. The total volume rate of sewage discharge is approximately 0.4 m^3/s (9 mgd). Outfall length is 1 524 m including the diffuser which is 293 m long and which lies in 25.9 to 30.5 m of water. The outfall pipe, laid on the bottom, was covered with large boulders for its entire length.

In compliance with a request from the City and County of Honolulu, this study, conducted in June 1978, is a follow-up to the 1975 survey.

OBJECTIVES

To determine changes, if any, made in the benthic community, the 1975 stations were reexamined in the summer of 1978 (Fig. 1). At station depths of 6, 12, and 18 m (20, 40, and 60 ft), the following were monitored, and results of the observations were compared with the 1975 data:

1. Fish population abundance and diversity
2. Biomass of attached algae
3. Diversity and cover of hermatypic corals
4. Distribution, abundance, and diversity of micromollusks.

RESULTS

Water Quality

Possible causative factors of stressed benthic communities receiving particular attention in the past include increases in nutrients, turbidity, sediments, and various toxins. It is possible that all

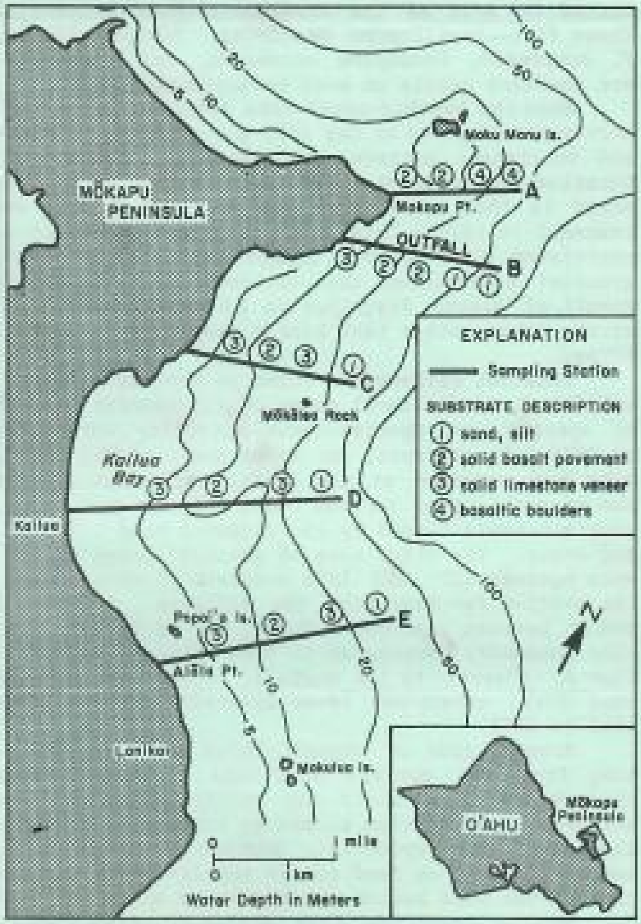

FIGURE 1. SAMPLING STATIONS, 1975 AND 1978, MŌKAPU OUTFALL, KAILUA BAY, O'AHU

Fish

Fish communities at each of the stations and depths remained essentially the same from 1975 to 1978 except for station B, the outfall site. Similarity indices show that the communities did not change their composition by species. Such change would only be expected under extreme stress, e.g., depletion of food supply, destruction of substrate habitat, change of physico-chemical environment. Changes of this type were not observed in this study. Results also show little change in total fish, diversity index, and species richness at each station of Honolulu, began operation in December 1977. In February 1978 the Kailua Bay outfall was connected to the Mōkapu line, and in May of the same year the Kane'ohe Bay outfall was also diverted to Mōkapu. The total volume rate of sewage discharge is approximately 0.4 m^3/s (9 mgd). Outfall length is 1 524 m including the diffuser which is 293 m long and which lies in 25.9 to 30.5 m of water. The outfall pipe, laid on the bottom, was covered with large boulders for its entire length.

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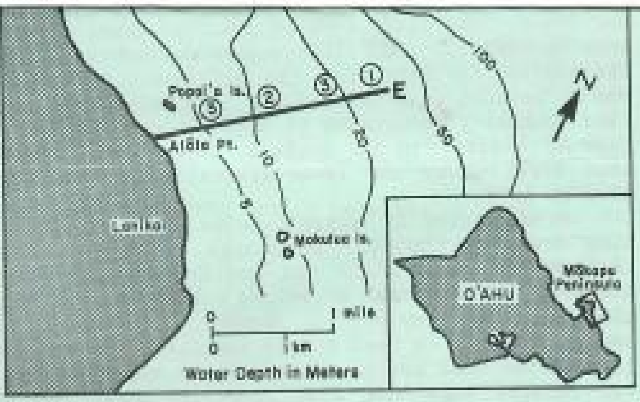
RESULTS

Water Quality

Possible causative factors of stressed benthic communities receiving particular attention in the past include increases in nutrients, turbidity, sediments, and various toxins. It is possible that all exert some influence on the distribution and abundance of the benthos with some effects probably being synergistic. In addition, human activity, such as sewage discharge, can alter community structure by creating artificial changes in the above variables. When organic matter (sewage) is discharged into an environment, the major environmentally significant aspect is nutrient loading and consequent cultural eutrophication of the ecosystem. Community structure is often defined, at least in part, by its diversity. In polluted environments there normally is less diversity because pollution intolerant species give way to more tolerant forms. With less competition for existing resources, the tolerant forms tend to become more abundant.

During scuba dives at 26 to 27 m over the diffuser, the effluent discharge was noticeable directly over the diffuser but disappeared 15 to 20 m from the outfall. The outfall plume discolored the water slightly as it rose and formed a 2- to 3-m turbidity layer over the outfall. The mixing is vigorous in these waters, and it is speculated that little nutrient enrichment takes place except at the diffuser.



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At station B results showed a large increase in all major fish families, especially over the large rocks covering the outfall. Transects laid at a distance 10 m from the outfall line showed less fish than directly over the outfall but still showed more total fish and greater species richness than in the 1975 survey. At station B depths where no fish were seen in December 1975, fish now abound. At station B20 there was an 82% increase in total fish counted, a 77% increase in numbers of species, and a 123% increase in diversity.

The new substrate provided by the large rocks covering the outfall attracted large numbers of fish, providing not only substrate for the growth of attached algae but also habitat space. The rudderfish *(Kyphosus cinerascens)* was abundant over the outfall substrate. This fish, a strict herbivore, was seen grazing on attached algae. Acanthurids, also generally herbivorous, were abundant along the outfall. Large numbers of weke *(Mulloidichthys auriflamma)* were observed feeding on the sediment alongside the outfall. Weke normally feed on benthic infauna and may have been feeding on organic detritus in the sediment. Whatever their food source, the outfall site attracted them in large numbers. Also observed were large aggregations of *Lutjanus kasmira* (taape), *Acanthurus olivaceus*, *Mulloidichthys flavolineatus* (weke), and *Parupeneus porphyreus* (kumu).

These observations are consistent with many studies done on the increased numbers of marine organisms, especially fish, associated with the construction of artificial reefs.

Corals

The environmental variables seeming to affect coral community structure most directly are wave energy which causes skeletal breakage and tissue abrasion, available light energy associated with photosynthetic and calcification processes, available solid substrate for larval settling, sedimentation that may smother corals, and interspecific competition between corals and other benthic forms.

Of the thirteen species of coral encountered, five can be considered dominant species as they occurred on most of the coral-containing transects. These five—*Pocillopora meandrina, Porites lobata, P. compressa, Montipora verrucosa*, and *M. patula*—are dominant corals on most Hawaiian reefs.

When the benthic-cover data obtained in the 1978 survey are compared to the 1975 survey data, no clear and consistent patterns associated with the outfall location are evident. The only pattern that does occur is that coral cover was lower on all but one transect in 1978. This trend does not appear to be correlated with outfall stress, since some of the greatest differences that occurred appear to be the result of patchy distribution of benthos and substrate types rather than biotic responses to outfall stress.

Greatest differences occur at the outfall site, station B, where coral cover of all species, number of species, and species-cover diversity were lower in 1978. In contrast, an enhancement of the community had occurred at the outfall structure itself due to settling of various benthic forms on the complex substrate formed by the diffuser pipe and boulder cover. The other area of greatest cover difference between 1975 and 1978 occurred at station E60, the station furthest from the diffuser. Stations C and D, between stations B and E, had relatively similar community structures at both surveys while station A, closest to the outfall, showed consistently less coral cover and lower diversity in 1978 relative to 1975.

Accumulation of organic-laden sediment emanating from the outfall discharge or from passage through the food chain can modify or cover substrates, causing them to become unsuitable for settlement of many epibenthic species. An effect of sewage effluent on reef corals appears to be domination of the hard bottom substrates by epifaunal filter feeders dependent upon suspended organic materials in the water column. These filter feeders can successfully outcompete corals for space and light. Benthic algal blooms attributable to eutrophication from sewage outfalls also have resulted in reduced settlement and smothering of corals in nutrient-rich outfall areas.

No benthic filter feeders or benthic algae common around other shallow outfalls were observed in the immediate vicinity of the Mōkapu outfall. The only abundant filter feeder that has colonized the area is *Triphyllozoon hirsutum*, and this species is presently limited to the outfall structure itself, appearing in relatively small clumps with large areas of bare substrate still available. Possibly the combination of a relatively low discharge rate (0.4 m^3/s or 9 mgd) and relatively strong and variable current velocities of up to 1 knot around Mōkapu Pt. disperse the effluent rapidly enough that the increase in suspended organic material is not great enough to support dominating communities of filter feeders or leafy benthic algae.

Algae

Results of the algae collection showed that there was no large change in biomass or species composition from 1975 to 1978. *Dictyopteris, Amansia, Asparagopsis*, and *Desmia* were the predominant genera.

Micromollusks

Micromollusk samples are separated into two groups of stations, at 6- to 12-m depths and 18- to 30-m depths. This separation was suggested by differences in species composition which were confirmed by analysis using the Sorenson coefficient of similarity which demonstrated two clusters of assemblages, a shallow water assemblage at 6 to 12 m and a deeper water grouping at 18 to 30 m.

As in the 1975 samples, gastropods comprise between 86 and 100% of the micromolluscan assemblages. Most of the gastropods are epifaunal; hence, the assemblage rather than one associated with infauna.

At the 6- to 12-m depths interval abundance, species composition, and distribution are in the main similar to those described in 1975; but some differences, especially with respect to the proportions of species present are apparent. Overall abundance at this depth interval was little changed with respect to the 1975 abundance figure (7.1 shells/cm^3 vs. 7.6 shells/cm^3). Four groups of micromollusks comprise about 50% of the assemblages on each transect: the archaeogastropod *Tricolia variabilis*, the rissoids *Rissoina ambigua* and *R. miltozona*, the cerithids *Bittium parcum* and *B. zebrum*, and species of the family Triphoridae. At depths of 18 to 30 m, average abundance is slightly higher, 9.1 shells/cm^3, than at the 6- to 12-m depths. Dominant species at the deep stations are the rissoid *Vitricithna marmorata*, the dialid *Cerithidium perparvulum*, species of the family Triphoridae, and Tricolia.

Differences between the 1975 and 1978 micromolluscan assemblages include:

1. A generally lower abundance (number of shells/cm^3 of sediment)
2. Increased proportions of *Bittium parcum, Tricolia variabilis*, and *Vitricithna marmorata* at the 6- to 12-m depth levels and concomitant decrease in the proportions of Triphoridae
3. Increased proportions of *Vitricithna* at the 18- to 30-m depth levels.

The differences may in part be ascribed to the effects of the diffuser system on the biota and may in part be associated with the patchy nature of the substrate.

REFERENCES

Kay, E.A.; Reed, S.A.; and Russo, A.R. 1973. A baseline survey of benthic biota in Kailua Bay. In *The quality of coastal waters: Second annual progress report*, Tech. Rep. No. 77, Water Resources Research Center, University of Hawaii.

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University of Hawaii

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NOTES ON FREE-FLOATING PLANTS IN THE PACIFIC

V. B. Vozzhinskaya, Institute of Oceanology USSR Academy of Sciences, Moscow, USSR

Materials on the free-floating plants were collected from the Pacific ocean by various expeditions (r/b "Vitjaz" and others) in 1955-1964.

Over 100 species of marine plants (green, brown, and red algae and to the higher flowering were found in samplies: marine grasses). More than a half of the most species number belonged to Fucales and Laminariales. Its morphological structure of the thallus may permit its floating.

As a rule distribution of the floating plants in the Pacific reveal a strictly latitudinal character and may be subdivided into groupings. In accordance to the phytogeographical scheme developed by A.Zinova, 1962 these groupings may be named: the upper-boreal - boreal, the lower-boreal-subtropical, tropical, notal, and lower-antarctic grouping. The first grouping is characterized by representatives of Genera <u>Alaria</u>, <u>Lessonia</u>, <u>Nereocystis</u>, <u>Macrocystiis</u>, <u>Cystophyllum</u>, <u>Fucus</u>; the main genera in the second grouping is <u>Sargassum</u>: to a lesser extent other genera such as: <u>Cystophyllum</u>, <u>Hizikia</u>, <u>Pelvetia</u>, <u>Undaria</u> the mass of the third grouping is represented in; <u>Sargassum</u>, <u>Turbinaria</u> and <u>Hormophyza</u>; a considerable number of <u>Carpophyllum</u>, <u>Macrocystis</u>, <u>Durvillea</u> (are marked in the fourth grouping; <u>Durvillea</u> and <u>Phyllogigas</u> with <u>Macrocystis</u> (rarely) occur in the fifth grouping. All these algae form the basic floating masses.

The distribution of the floating plants in the Pacific is strongly influenced by currents. The greatest accumulation of floating is noted at the junction areas of currents. The highest number of floating plant packs has been observed along the shores of Kamchatka, the Komandor islands, the Aleutian isl., Kurile isl., islands of the Japan (the paper of Japanese algologists contain similar data of mass occurrence), Philippine isl., New Guinea, and the neighbouring areas and New Zealand.

Floating alage occur in the central Pacific very rarely. The occurrence floating plants near the coasts where they grow points to their coastal origin.

The Pacific ocean has a more abundant number of floating plan species than the Atlantic and Indian oceans. In some areas the floating algae mass of the Pacific may be compared to the waters of the Saragass sea of the Atlantic.

Abstracts of Papers

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Tokyo, 1966

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* *

Catherine Craine Unabia, Department of Botany, University of Hawaii

SMARAGDIA (GASTROPODA: NERITIDAE), A SEAGRASS ANIMAL

(Major Professor: Dr. Kent W. Bridges)

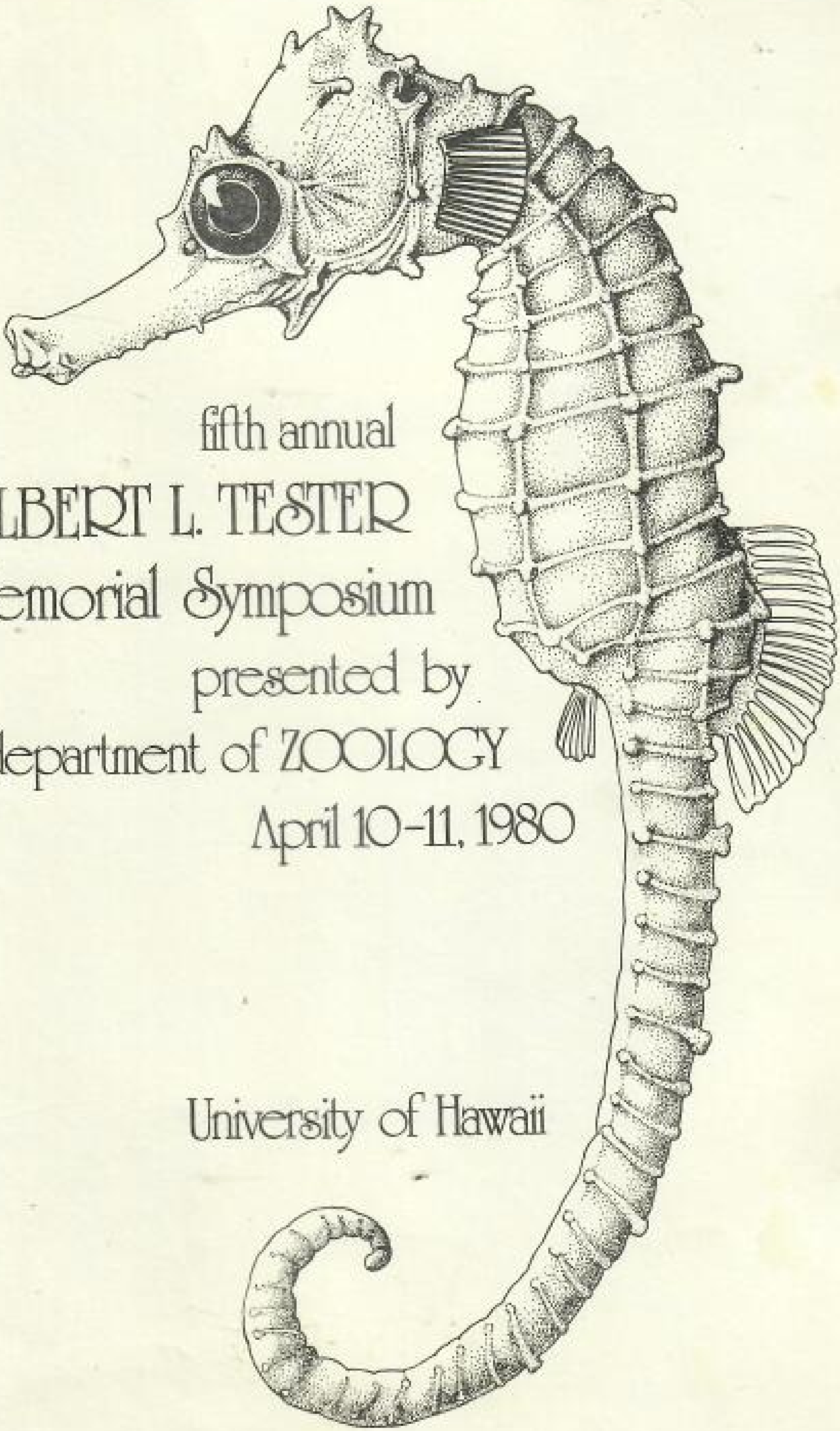
In Hawaii, the snail Smaragdia bryanae grazes the seagrass Halophila hawaiiana, and is limited in its distribution to that plant. They are found together around the islands of Kauai, Oahu, Molokai and Maui. This grazing interaction is unusual because most invertebrates associated with seagrasses or other aquatic macrophytes feed on epibiota of the leaf surface rather than higher plant tissue. The utilization of such tissue by gastropods requires specialization of the digestive system and radula. The radula of Smaragdia has been described as unique among the Neritidae, having modifications approximating the type found in higher snails, where a diet of macrophytes is usual for herbivorous species.

Feeding behavior of the snail has been observed in field and aquarium. Patterns of leaf damage were recorded photographically at various magnifications. A consistent and characteristic damage pattern can be recognized.

The Smaragdia - Seagrass association in Hawaii seems to be part of a widespread phenomenon. Reports of Smaragdia on seagrasses have been found from southern Japan, the Seychelle Islands, the Mediterranean and the Caribbean. Compilation of recent and fossil (Tertiary) Smaragdia records indicates that the range of the genus is Pan-tropical plus Mediterranean, distribution limits probably being set by temperature. Seagrasses are found at all sites where the snail is reported, but an exclusive relationship with Halophila is ruled out by the presence of the snail in the Mediterranean, where that plant is but a recent migrant since the opening of the Suez Canal. From records found so far, the trend seems to be that the snail is found most frequently where large numbers of seagrass species co-occur.

The ecological nature of this plant-animal association in other areas is not yet known, but extremely similar patterns of leaf damage have been seen on herbarium specimens of Halophila from the Philippines, the Caroline Islands, Australia and Madagascar. Marks that could be Smaragdia grazing damage were also seen on specimens of Thalassia and Halodule from the Philippines. It is suggested that Smaragdia may be, like the dugong, a true 'seagrass animal', ecologically dependent upon the marine angiosperms.

* *



fifth annual

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University of Hawaii

Tester Symposium
10-11 April 1980

William J. Walsh, Department of Zoology, University of Hawaii

RESPONSES OF A HAWAIIAN REEF FISH COMMUNITY TO A CATASTROPHIC STORM

(Major Professor: Dr. Leighton Taylor

On January 8 and 9, 1980, a severe cyclonic storm struck the west (Kona) coast of the Island of Hawaii. Wind-generated waves in excess of 8m. battered the normally protected leeward fringing reefs and caused widespread reef destruction and shoreline alteration. Extensive near shore shallows (1-3m.) were denuded of almost all bottom cover and marine life. Corals, sand and debris from this area were thrown up on shore or swept seaward over the edge of a 5m. drop-off approximately 75m. off shore. Such material was deposited at the base of this drop-off forming a layer of rubble up to 1.5m. thick which buried previously deposited rubble and live Porites lobata corals. Other storm-associated damage to the massive corals below this drop-off was minor. Beds of the fragile finger coral, Porites compressa, which lay seaward of this zone sustained substantial damage, however. In 9m. of water, up to 60% of the coral cover was smashed and in 13m., over 25% was damaged. Broken and dislodged heads could be observed down to a depth of 27m.

Fish mortality directly attributable to the storm was remarkably slight. The small numbers of fishes which were killed tended to be either surge zone species of individuals normally residing in tide pools. Large numbers of individuals left the shallows during the storm for deeper water around and below the 5m. drop-off. Transects in these areas showed the influence of these refugees in terms of increased numbers of juveniles, adults and species present. Population increases are still evident after almost three months.

A major faunal shift resulting from the storm was the ten-fold increase in the numbers of the territorial damselfish, Stegastes fasciolatus in the areas below the drop-off. Within two weeks after the storm, barren rock and calcareous rubble developed a heavy growth of bright green filamentous algae, particularly of the genus Enteromorpha. This algae was heavily grazed by herbivores and by one month after the storm had disappeared except in the extreme shallows and within some of the damsels' territories. This abundance of early colonizing algae enabled this highly site-oriented species to successfully shift to a new habitat area. Recolonization of the shallows is presently underway, primarily by small species and juveniles.

In the deeper water finger coral areas, diurnal fish populations remained unchanged or increased slightly, even in light of substantial coral destruction. Algal growth on dead coral rubble opened up new feeding areas to wandering herbivores. During daylight and twilight periods there was an increase in disordered movement about the reef due primarily to disruption of path habits, and changes in topography. An unusually high number of predatory events was observed shortly after the storm. Night transects did not show significant reductions in nocturnally active or diurnally inactive individuals. Porites lobata areas of high relief and large numbers of diurnal shelterers remained mostly intact. Heavily damaged finger coral areas had few inactive adult shelterers but damage was patchily distributed and total area of suitable shelter was still high.

* *

26 Jan '79

Dear George,

Thank you for the calendar. Probably one of the best memories I have of atolls concerns the seabirds. You really captured the best of their personalities. I can see you enjoy them too. Beautiful work!

I received your two boxes, and have not forgotten you. There are 35 vials total. My goal is to have them identified and back to you in March. Is that soon enough?

My family and I miss Hawaii, especially our friends and the people.

Aloha,

Dennis Russell

Is Alan still working for you?

Dennis Russell
MSLC
Seattle Pacific Univ.
Seattle, Wa 98119

SEATTLE WA 981
PM

anyone for limu lepe 'ula 'ula pupus?

by DONDA JOW
news editor

If you like seaweed, you might want to try some limu lepe 'ula 'ula pupus or an ar-ar-o-sip Filipino salad from Heather Fortner's up-coming cookbook.

Fortner, a University of Hawaii liberal studies senior, uses 18 different kinds of seaweed in her cookbook, tentatively titled "A Cookbook of Hawaiian Limu."

HEATHER FORTENER

TWO YEARS AGO, a project on edible seaweed for a "Living the Wealth of the Oceans" class stirred her interest. She began eating seaweed quite often and "thought it would be interesting to come out with a cookbook for Hawaiian seaweed."

Fortner submitted her proposal to the Marine Option Program last year and received funds from Seagrant to conduct her research on the Big Island.

Information for the cookbook comes from three sources:

—Recipes published previously,

—interviews with old Hawaiians on names and uses of seaweed, and

—imagination. Fortner made up recipes by substituting one type of seaweed for another, modifying recipes and using seaweed when an ingredient was unavailable.

THESE INCLUDE limu palahalaha guwumpkies (stuffed limu,) candied ogo, crisp limu tuna patties and deep fat fried limu kala chips. Some recipes were found by accident.

Fortner has tested most of her recipies. She uses different types of seaweed for soups, salads, main dishes, condiment and pickling.

The seaweed used most often in her cookbook is limu manauea—or ogo.

"**SEAWEEDS** are as diverse as land vegetables. There is a movement to call it sea vegetable instead of seaweed," Fortner said.

Because many people "consider seaweed a nuisance," the cookbook also tries to "inspire people to appreciate it as the old Hawaiians did," Fortner said.

> *'There is a movement to call it sea vegetable instead of seaweed.'*

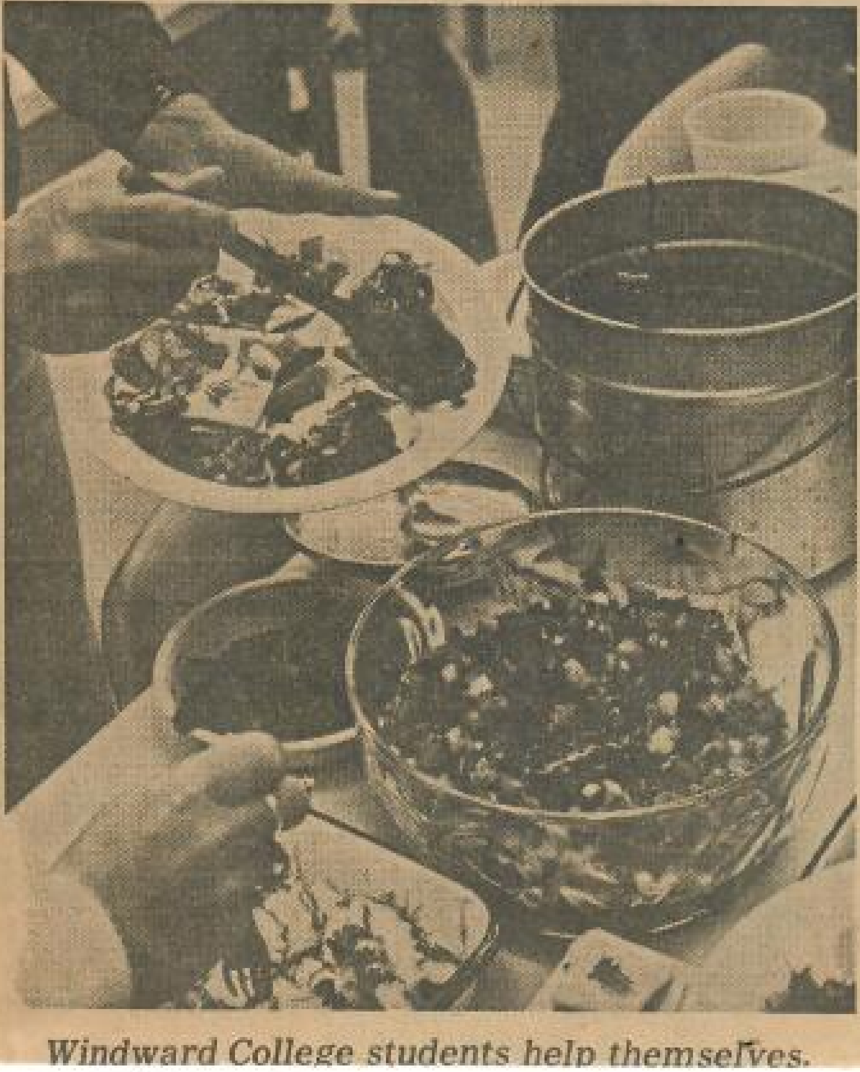
Fortner collects her own seaweed in places such as Ewa Beach, Laie, Castle Beach and Diamond Head. She recommends low tide as the best time.

After picking, the seaweed must be cleaned: "a slow, tedious chore, but well worth it," Fortner said. "You can spend the whole day at the beach and come home with your dinner," she said.

"There's no limit to what you can do with seaweed. It's all up to the imagination. It's exciting to experiment and come up with something delicious."

G

Food F



Windward College students help themselves.

Limu—getting to kn

By SANDRA MATSUKAWA HU
Advertiser Food Editor

Whenever there's talk about the scarcity of food to feed an ever-expanding world population, seaweed is mentioned as an alternate food source. But like most plans to utilize anything nutritious but unfamiliar, everyone seems to want the starving masses elsewhere to eat it, saving the more traditional steak and potatoes for himself.

Not so in Hawaii, where seaweed or limu is a well-loved delicacy. Hawaiians have always eaten limu to suppliment the diet and to break the monotony of meals that have consisted mainly of fish and poi. Today, most ethnic groups in Hawaii have their favorite recipes for this crunchy, flavorful food.

At one time, between 70 to 80 kinds of limu were eaten in Hawaii. Now the number has dwindled to about 20, according to Jeffrey Hunt, instructor of botany at Windward Community College. Hunt points out that there are no poisonous seaweeds — only some unpalatable ones.

"In general, if the limu is not hard and stony and if it's not too tough or bitter to your taste, I wouldn't worry about eating it," he said. But Hunt cautioned against eating blue-green colored algae, since some varieties can cause a rash or itch.

Why eat seaweed? Seaweed provides a substantial amount of trace minerals and vitamins in the diet, Hunt explained. It is high in vitamins A, B-complex, D and E as well as K, and a valuable source of iron and zinc. But Hunt doesn't expect limu to ever be a staple like potatoes or poi. "You'd have to eat a lot of seaweed to get all the protein and carbohydrates as you would get from steak and potatoes."

In Hunt's botony 181, Plant Life of the Sea, a seaweed-tasting lab is scheduled. Each student gathers some seaweed and brings it to class for final preparation. The school provides basic seasonings like soy sauce, sugar, chili peppers. Then the students experiment with recipes, winding up with a tasting session when the cooking is done.

"Lately, a lot of local people have been attending our lab," Hunt said. "Since usually half the students are newcomers to the Islands, it's fun when everyone interacts. The students learn from the local people how to clean a squid, how to eat raw fish, how to make poki."

During a recent lab session, a group of students combined red and green limu into a salad, tossing the leaves together with salad dressing. The dish looked and tasted somewhat like the conventional tossed greens. Other recipes tested at the lab session are given below.

If you're gathering limu for the first time, Hunt offers these suggestions: "Don't overharvest. I've seen people take more than they need and it just goes into the garbage can."

Because it is important not to deplete natural supplies, Hunt does not recommend any one spot for limu gathering. "There's enough limu on the reefs around here if you just look. The best time to go is at low tide."

When you harvest the limu, clean it well. All kinds of animals live in the fronds and there can be a lot of dirt. "Every time I don't clean limu carefully I eat a snail or bite down on a piece of rock," Hunt said.

The two sketches of commonly eaten limu in this article are from the book, *Limu, an Ethnobotanical Study of Some Edible Hawaiian Seaweeds*, By Isabella Aiona Abbott and Eleanor Horswill Williamson.

Hunt also is writing a book on limu with William C. Magruder, a graduate student in psychology. The book, which will be published by the University Press in January, will contains color photographs of the 100 most common seaweeds in Hawaii.

Here are some interesting recipes to try that are especially appropriate for first-time tasters. Hunt collected them from various sources, including old-time tutus and skilled local cooks.

Poki (Hawaiian)

Limu manauea, limu huluhuluwaena, limu lipe'epe'e, limu mane'one'o or ogo
Raw fish, like ahi or aku
Hawaiian salt

Manauea or ogo (Gracilaria coronopifolia)

eport

Honolulu
Advertiser
Wednesday, May 4, 1977

Advertiser photos by Ron Jett

Limu can be prepared in many different ways.

ow it and like it

Cube raw fish into 1-inch squares. Add Hawaiian salt to taste. Chop limu into 1-inch pieces, adding it to salted raw fish. Mix with hands. Serve cold. The same ingredients may be used with raw opihi instead of raw fish.

Limu wawae'iole, palahalaha and huluhuluwaena may be added in small quantities with the manauea or ogo.

VARIATION: At the latest limu lab, a Hawaiian cook made the poki with limu kohu and added some mashed kukui nut to the mixture.

Limu tsukudani (Japanese)

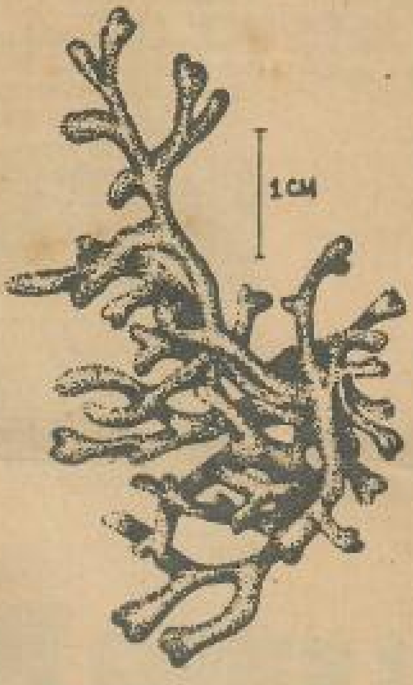
1 pound limu manauea or ogo
1¼ cup brown sugar
½ cup mirin (sweet rice wine)
1¼ cup soy sauce
¼ teaspoon msg

Clean limu. Bring sugar, soy sauce and mirin to a full boil. Pour boiling water over limu and let stand for 3 to 4 seconds. Drain and rinse with cold water. Drain. Place limu in container and cover with sauce. Sesame seeds and chili peppers maybe added to taste.—Ray Rounds.

Pickled shredded guso (Filipino style)

1 cup shredded guso*
4 slices ginger
¼ onion, cut into small pieces
¼ cup wine vinegar
4 tablespoons sugar
½ teaspoon salt

Select pieces of fresh guso about ½-inch in diameter. Pass these through coarse part of a shredder. Boil 2 cups water, add guso and keep boiling 3 minutes. Strain guso. Put vinegar in a small pan add salt, sugar, onion and ginger and simmer. Pour the mixture into the guso and mix well.—Dr. Gertrude Santos.



Wawaeiole (Codium edule)

*Guso, imported from the Philippines, now grows in Kaneohe Bay.

Korean-style ogo

1 pound ogo or limu manauea
1 cup rice vinegar
½ cup soy sauce
1 tablespoon sesame oil
1 tablespoon roasted sesame seeds
2 tablespoons brown or white sugar
2 tablespoons mirin (sweet rice wine)
Ko Choo Jung (Korean hot sauce) to taste
Chili pepper to taste
Chopped garlic to taste

Clean limu well; pour boiling water over limu until color just turns greenish. Mix remaining ingredients together. Add limu to mixture. For best results, refrigerate overnight in containers.

Polish-style stuffed palahalaha

15-20 fronds or blades of limu palahalaha*
1½ pounds ground pork or pork-beef mixture
4 teaspoons chopped onions
2 beaten eggs
1 teaspoon salt
½ teaspoon thyme
½ cup chopped limu manauea or ogo
1 teaspoon black pepper
Garlic to taste

Clean limu palahalaha in fresh water. Simmer in a pot of fresh water over low heat while preparing the stuffing. To prepare the stuffing, mix together the remaining ingredients.

Remove limu palahalaha from water and lay fronds flat, placing two together, depending on the desired thickness. Place golf-ball size portions of the meat mixture on each set of fronds. Fold the sides over and roll. Secure with a toothpick. Place the stuffed limu in a small casserole dish, dotting each with butter. Pour ¾ cup tomato sauce into casserole and bake for one hour at 375 degrees F. Strength of limu flavor will depend upon the amount of limu palahalaha, manauea or ogo used.—Sea Grant and Marine Options Program.

*The best fronds of limu palahalaha to use are about 4 to 6 inches X 2½ to 3½ inches.

KEY TO THE FREQUENTLY FOUND GENERA

of

HAWAIIAN SEAWEEDS, Edition 13

The following is what biologists call a "key". It is so constructed that one is presented with a series of alternate choices or pathways. If one makes only "right" choices, with regard to one of the conspicuous seaweeds (LIMU) commonly found around the Hawaiian Islands, the pathways will always lead to the same end, a generic name for that seaweed.

Approximate sizes of American coins in the metric system.

	Diameter in mm	Thickness in mm
dime	18	1
penny	19	1.5
nickel	21	2

Ten millimeters (mm) equals 1 centimeter (cm) and 100 cm equals 1 meter (m), and one meter is about 39 inches.

The whole individual alga is referred to as a thallus (plural: (thalli). There are pairs of diagrams illustratin unilateral, dichotomous and pinnate branching on the last page, below. The word blade is used for any flat form attached by one edge or end like the blade of a leaf of grass, castor bean, kiawe, etc. A crust is a thin piece of hard matter attached to something larger by one flat side; like the brown material on the outside of a loaf of bread. Stony mea inorganic and usually of carbonate or limy material like concrete. Filaments are cylindrical objects not unlike hairs. Prostrate mean lying down. Reproductory lumps, i.e., conceptacles or cystocarps, are swellings or spots on the surface in which the reproductory structures are born.

1. Thallus a crust or consisting of inflexible branches or knobs.........2

1. Thallus an erect pliable frond, lump, or mass of filaments (hairs)...14 (p. 3)

2(1) - Thallus stony, with lime ($CaCO_3$) deposits throughout.....3

2(1) - Thallus not stony though it may be hard; red,
to black, green or brown in color............10 (p. 3)

3 (2) - Thallus of erect rigid knobs or branches......................4

3(2) - Thallus a crust or series of small horizontal shelves...........7

4(3) - Thallus surface smooth, without spiral pattern......5

(ALSO, NO MULTIPORATE ASEXUAL CONCEPTACLES) APRIL, 1993.

4(3) - Thallus surface sculptured in spiral whorls, with knob-like protoberances................*Tenarea*

5(4) - Thallus surface having a mosaic pattern, purple, lavendar or maroon..*Hydrolithon*

5(4) - No mosaic pattern; protuberances branched, slender and rigid.........6

6(5) - Texture of thallus powdery due to fields of clumped large cells.......*Porolithon gardineri*

6(5) - Texture of thallus glazed and smooth, often forming nodules..........*Lithophyllum* (*kotschyanum*)

7(3) - Thallus a flat crust usually on dead reef or lava rock, intertidal in areas of wave shock; texture powdery due to fields of clumped large cells................................*Porolithon onkodes*

7(3) - Subtidal crusts on rock or other seaweeds, no fields of large cells....8

8(7) - Thalli very thin (far less than 1 mm thick, growing on frondose algae......................9a

8(7) - Thalli thick, growing on rock...................9b

9a(8) - Some reproductory lumps (conceptacles) having many pores in their upper surface; thalli about 0.1 mm thick; on other algae, glass and different substata...................................*Melobesia*

9a(8) - All reproductory lumps having but one pore in their upper surface; on noncalcareous algae................................*Fosliella*

9b(8) - Obvious reproductive lumps with many pores, texture rough, only found in deep water.....*Lithothamnium*

9b(8) - No obvious reproductive lumps, texture glazed and glossy; deep maroon when in shade and green when in direct light, found in both deep and shallow water................................*Sporolithon*

HONOLULU STAR-BULLETIN 1-17-85 A:17

Tests to Grow East Coast Seaweed OKd

By Pat Guy
Star-Bulletin Writer

Hawaiian Marine Enterprises got permission yesterday to bring in 100 pounds of an East Coast seaweed that it hopes eventually to grow here commercially.

The state Board of Agriculture approved Richard Spencer's request to bring in the limu called Gracilaria tikvahiae.

Spencer, one of the partners in Hawaiian Marine Enterprises, plans to conduct research to determine whether the limu can be produced for local consumption.

Limu, or edible seaweed, is a popular food in Hawaii but there is no commercial production. Limu pickers here scout the beaches for it.

ACCORDING TO Spencer, about half a million pounds of limu are consumed here annually. He wants to capture a quarter to half of that market.

He began cultivating two local types of limu about 18 months ago, but the results were not good. In winter, the plants would "go sexual," he said, breaking up into small pieces and growing slower.

"I saw the writing on the wall," Spencer said. He then contacted J.H. Ryther who has been cultivating limu for six years in Florida as part of an energy research project.

"His plants grow all year round and are just gorgeous," according to Spencer.

Spencer estimated that research here with the introduced species of limu would take "no longer than three months."

IF ALL GOES well, he then will return to the agriculture board for permission to sell the limu commercially.

The board set several conditions for the importation of the new limu to make sure it does not escape to the ocean.

Stanley Y. Higa, plant quarantine branch manager, said the limu-growing facility at Kahuku is well guarded and located far enough inland to prevent it from getting into the ocean, where it might become established and change the ecology of the area.

ALGAE FILE -

JULY 1984

Note -
Abbott's 4th ed. limu books mentions "Limu Honu" - Sargassum - "probably because turtles eat it for food"

Resent
4/8/85

January 13, 1983

Dr. R. A. Fralick
Plymouth State College
University System of New Hampshire
Plymouth, New Hampshire 03264

Dear Dr. Fralick:

I recently had the opportunity to read your interesting paper on <u>Pterocladia</u> <u>pinnata</u> that appeared in the Proceedings of the 10th International Seaweed Symposium. Our green sea turtles (<u>Chelonia mydas</u>) here in Hawaii feed heavily on <u>Pterocladia</u> at a number of shallow coastal areas. As a researcher of these sea turtles, I would like to learn more about the ecology of this important food source. I wonder if you would be able to give me the citations to any other current publications dealing with <u>Pterocladia</u> that have come to your attention. I would greatly appreciate your assistance with this request.

The enclosed reprints cover some of the results of my work with Hawaiian sea turtles. Best wishes for the New Year.

Sincerely,

GEORGE H. BALAZS
Assistant Marine Biologist

GHB:ec

Enclosures

FORM 354 (08/81)

UH FEED AND FORAGE ANALYSES PROGRAM
AGRICULTURAL DIAGNOSTIC SERVICE CENTER
UNIVERSITY OF HAWAII AT MANOA
1800 EAST-WEST ROAD
HONOLULU, HAWAII 96822

University of Hawaii Cooperative Extension Service
U. S. D. A. Cooperating

NUTRIENT COMPOSITION*

DATE June 13, 1985

REQUESTED BY GEORGE BALAZS

ADDRESS National Marine Fisheries Service
P. O. Box 3830, Honolulu 96812

COLLECTION SITE Kahului, Maui

COLLECTION DATE 5/9/85

UH No.	SAMPLE DESCRIPTION	DM	Ash	CP	EE	NDF	ADF	PML	C		
		(%)									
F85-195	Codium Edule - Channel	5.6	60.1	8.2	.6	21.8	9.9	3.5	6.2		
196	Codium EdulÁ - Outfall	7.4	57.0	10.7	.5	26.6	10.7	3.4	6.8		
197	Acanthophora - Outfall	6.8	45.3	23.2	.1	23.1	16.2	6.8	8.9		
198	Codium Reediae - Outfall	5.1	53.5	12.2	.8	23.8	8.1	2.5	5.6		
	MINERALS TO FOLLOW.										

* Dry matter basis, except for DM, which is expressed on an "As Sampled" basis.
DM = Dry matter, as sampled; CP = Crude protein; EE = Ether extract (crude fat); NDF = Neutral detergent fiber; ADF = Acid detergent fiber; PML = Permanganate lignin; C = Cellulose.

George Balazs
National Marine Fisheries Service
P.O. Box 3830
Honolulu, Hawaii 96812 August 15, 1985

Dear George,

Enclosed are the results of the last series of samples you sent to me. The samples were sent to you yesterday and may arrive before this letter, since I had to organize and type the results. Also, you will notice there is a master list for all the algae that have been identified for you since we began in 1976. This list can be used to find spellings and authors of species. It probably contains a few mistakes (I hope not), and will be added to as we go, so for that reason I have dated to avoid confusion when species are added to it and corrections made.

You have sent me several articles, like the one on Tagging Turtles, the House of Representatives Committee on Ocean and Marine Resources agenda, Seagrasses, etc. and I thank you for them. I have filed them into notebooks that I am using as references. You have also asked me several questions that I have not answered yet.

April 13, you asked, "I found a piece of Pterocladia with Ulva fasciata attached firmly to the top growth on it. Is this common?" Yes, Ulva species are very opportunistic and will attach to and grow on just about anything that is rough and solid. At certain times, just about any species can be found as an epiphyte on another alga, especially the solid rough hosts.

August 8, you asked me about Acanthophora and my progress on the manuscript that describes its introduction, spread, physical requirements and competition with the native species of algae. Unfortunately, I began that manuscript and put it aside when an opportunity to write two chapters for a UNESCO book came around. My teaching load was 40 hrs/year last year and nearly killed me. This year it will be the normally high but lower 33 hrs/yr. I will be back at that manuscript and will also work on a review paper that is scheduled to be published in late 1986.

My tenure application goes to our Faculty Status Committee by October 18, 1985, so that hassle will be over soon. Promotions and tenure at SPU involve a lot of busy work, to show we are interested in, community, fine arts, church and school activities. All of this takes away from real productivity (publications are also required).

In the mean time, please feel free to look through my dissertation and glean what you can from it. I put in a lot of maps of Acanthophora distribution and tried to make it readable by just about anyone.

Thank you for the work. I am glad to be a service to you. Good luck on your report.

Aloha,

Dennis

School of Natural & Mathematical Sciences

Seattle Pacific University
Seattle, Washington 98119
Phone: (206) 281-2140

George Balazs
National Marine Fisheries Service
P.O. Box 3830
Honolulu, Hawaii 96812

August 28, 1985

Dear George,

I am sorry about the inconvenience made by the lost letter and identifications. I will be sure to have them done first thing after they arrive. Normally I only keep the last listing on my computer diskette and keep a hard copy in a notebook. The July 5th file was deleted when I entered the new information and I didn't make a xerox copy of it like I usually do.

Your volunteering to write a letter for my tenure file was very timely since I am coming up for tenure next month and the file is due, complete, by September 18. An unsolicited letter from you to the Dean of the School of Natural and Mathematical Sciences would be greatly appreciated. The tenure committee is especially impressed by the scientific significance of a project and international importance. Your evaluation of my work (a person outside SPU) will greatly enhance the quality of my file. Can I do the same for you? The letter should be addressed to:

Dr. Karl Krienke, Dean
School of Natural and Mathematical Sciences
Seattle Pacific University
Seattle, Washington 98119

After this tenure business is finished, maybe I'll have time to gather a few loose ends. Especially with writing.

Aloha,

Dennis

Congratulation to you and your wife on the birth of your son. I assume all are well & happy.

12/28/78 GCB

September 9, 1985

Dr. Karl Krienke, Dean
School of Natural and
Mathematical Sciences
Seattle Pacific University
Seattle, Washington 98119

Dear Dr. Krienke:

I recently heard that Dr. Dennis Russell of your University is currently being evaluated for tenure. This is an unsolicited letter to inform you of the significant contributions Dr. Russell has made over the past 9 years to our knowledge of the Hawaiian green turtle, *Chelonia mydas*. Since 1976 Dr. Russell has periodically analyzed and identified the algal component of food samples obtained in my research on free-ranging green turtles. As the result, a substantial body of data now exists on the dietary habits of this species at numerous resident foraging pastures throughout the Hawaiian chain. As the principal researcher of sea turtle in Hawaii, initially with the University of Hawaii and now with the National Marine Fisheries Service, I have repeatedly depended upon Dr. Russell's knowledge and expert advice in matters relating to Hawaii marine algae. Without Dr. Russell's willing and active participation these past 9 years, our understanding of the Hawaiian green turtle would be much less than it is today.

I should also mention that, along with other sea turtles, the Hawaiian green turtle is protected under the U.S. Endangered Species Act. An official Recovery Team has recently been appointed, and efforts have started to prepare a formal recovery plan. I look forward to Dr. Russell's involvement in this important work.

Sincerely,

George H. Balazs
Zoologist and Leader,
Hawaiian Sea Turtle Recovery Team

PEACOCK DAMSEL-FISH (**Pomacentrus pavo**). Plentiful in some of the larger pools. Brilliant metallic blue with yellow tints on the fins, in certain angles of light they are just as brilliantly green.

FIVE-BANDED SURGEON-FISH (**Acanthurus triostegus**). Quite plentiful. The transverse bands were often pale and indistinct.

BLUE-SPOTTED BOX-FISH (**Ostracion tuberculatus**). Plentiful in reef pools including some only a few inches deep. Most were rich brown with metallic blue spots, but some were rich yellow.

TOADS (**Tetraodon stellatus**). Some very large specimens seen in coral pools and along the beach shoreline on the incoming tide, average length about 20 inches.

SLENDER SUCKING-FISH (**Echensis neucrates**). One about 30 inches long caught from a launch anchored near ours when we were unloading on arrival at the island.

CLOUDED REEF-EEL (**Echidna nebulosa**). A small eel. Body colour creamy to whitish with dark blotches breaking up into star-like spots. Reef-eels have priority for the name **Echidna** over our monotreme Spiny Anteater.

TESSELATED REEF-EEL (**Lycodontus favagineus**). Colour black broken up by fine white lines giving the appearance of large black spots. A small specimen about 15 inches long was found among the coral.

SHARK. One about 7 ft long swam within a few feet of the shore late one afternoon. Distinct white tips were seen on the dorsal fins and the upper lobe of the caudal fin, suggesting it may have been a White-tip Shark (**Trienodon apicalis**).

BLUE-SPOTTED LAGOON RAY (**Taeniura lymma**). Many seen on the reef and along the coral sand shoreline, moving and feeding with the rising tide. Often fishermen, after catching a ray with a line, chop off the tail which is armed with a strong barbed spine and then release the ray into the water. It was of interest to see one that appeared to have received this treatment happily cruising along the shoreline in its search for food.

COMMON SHOVELNOSE RAY (**Rhinobatus armatus**). Extremely common everywhere and could be seen along the shoreline moving with the incoming tide. I stood in one spot and counted 26 in sight; they approached within an inch or two of the water's edge. Most were about 24 inches long and many were captured and eaten. The flesh is white and very palatable.

ALGAE ON A HAWK'S-BILL TURTLE

By A. B. CRIBB

There are numerous references in American literature (see Edgreen, R. A. et al., 1953) to algae growing on freshwater turtles, the name turtle in that country denoting any member of the Order Testudines which swims. In Queensland, green tufts of filamentous algae are common on the carapaces of freshwater members of the group, here known as tortoises, and the green alga **Basicladia ramulosa** Ducker (1958) has been described from **Chelodina longicollis** Shaw in Victoria and Queensland. However, there seems to be no previous record of the occurrence in Australia of algae on marine members—the turtles of local terminology.

A Hawk's-bill Turtle, **Eretmochelys imbricata** (L.), examined at North West I. during the Queensland Naturalists' Club expedition in Aug. 1968 carried crusts of calcareous algae on the plastron and an intermittent, fine fur of filamentous species over both carapace and plastron. Later examination of material scraped from the turtle revealed a surprising total of 38 algal species as follows:

CYANOPHYTA

Anacystis dimidiata (Kuetz.) Dr. & Daily
Calothrix crustacea Thur. in Born. & Thur.
Entophysalis conferta (Kuetz.) Dr. & Daily
Entophysalis deusta (Menegh.) Dr. & Daily
Microcoleus lyngbyaceus (Kuetz.) Crouan
Oscillatoria lutea C. Ag.
Schizothrix calcicola (C. Ag.) Gom.
Schizothrix tenerrima (Gom.) Dr.
Spirulina subsala Oersted

CHLOROPHYTA

Bryopsis pennata Lamouroux
Cladophora crystallina (Roth) Kuetz. prox.
Enteromorpha clathrata (Roth) Grev.
Ochlochaete ferox Huber
Phaeophila dendroides (Crouan) Batters
Pilinia sp.
Pseudopringsheimia sp.
Rhizoclonium implexum (Dillw.) Kuetz.
An undetermined filamentous species

PHAEOPHYTA

Ectocarpus irregularis Kuetz.
Ectocarpus mitchellae Harv.
Ectocarpus rhodochortonoides Boerg.
Sphacelaria furcigera Kuetz.
Sphacelaria novae-hollandiae Sond.
Sphacelaria tribuloides Menegh.

RHODOPHYTA

Acrochaetium catenulatum Howe
Acrochaetium daviesii (Dillw.) Boerg.
Acrochaetium robustum Boerg.
Acrochaetium sp.
Acrochaetium sp.
Ceramium gracillimum var. **byssoideum** (Harv.) Mazoyer
Ceramium serpens Setch. & Gard.
Erythrotrichia carnea (Dillw.) J. Ag.
Fosliella sp.
Gelidiella sp.
Goniotrichum elegans (Chauv.) Le Jol.
Herposiphonia tenella (C. Ag.) Ambronn
Lophosiphonia scopulorum (Harv.) Wom.
Melobesia sp.

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H

This section prepared by the staff of The Honolulu Advertiser.

Peter Tamada prepares aku poki at Ft. Ruger Market.

Advertiser photo by Gregory Yamamoto

For Tamashiro clan, poki's a big fish story that's true

from the sea

mike markrich

Tamashiro Market employees help their customers with many kinds of information. They suggest preparation methods for fish. They offer new ideas on how to flavor old favorites; if the favorites are out of stock, they give advice on how to cook fish they have.

But one thing they never give out is the secret to their poki. The traditional Hawaiian dish was once raw fish cubes, seaweed and salt; now it's a carefully prepared gourmet item involving different kinds of cooked as well as uncooked seafoods, seaweeds, vegetables and chili-pepper.

"It's top secret" said Cyrus Tamashiro of the flavoring methods they use to make their products distinct. "We instruct all of our employees not to give out the formulas."

With good reason. There are now more than 20 kinds of poki sold by Tamashiro Market, displayed like candy behind a large glass case. There is aku poki, aku poki with onions, tako poki, mussel poki, crab poki, imitation crab poki as well as the new flavors that the poki makers at the store come up with periodically.

It has become one of the store's most popular items for customers from many different ethnic and economic backgrounds.

But it was not always this way. Tamashiro, 32, said that until the mid-1970s the market sold only one kind of poki — chopped aku with limu (seaweed). Back then, as he and his mother explained, poki was thought of as an ethnic food with a limited market.

In fact the Tamashiro's chief poki maker for the last 10 years, Patsy Choy, 62, (who is Japanese American) said that when she was growing up, she never ate it. But then neither had Peter Tamada of the Ft. Ruger Market.

"I didn't even know what it was " said Tamada, 60, who has now been making it for 13 years. "When I first started my business was real slow and I was having to get rid of a lot of 2-day-old aku. So I told the people at the fish market that I couldn't afford to handle aku. They said, 'Why don't you make poki with it?' I said 'What is that?' "

He then began to mix poki and it has become one of his store's biggest sellers.

Cyrus Tamashiro said he's not sure when tastes changed. But he said that toward the mid-1970s, his father Walter Tamashiro noticed that the poki was selling out by mid-morning. He expanded the poki varieties and soon was selling more poki than meat.

"My dad took a gamble," Cyrus recalled. "He realized that we couldn't compete with the convenience stores. But we could sell a lot of poki."

Walter Tamashiro hired poki specialists to experiment and create new flavors.

The increase in demand caused concern among biologists at the university who worry that it has contributed to the large-scale taking of limu.

These concerns have found expression in hearings being held to restrict each person to 10 pounds per day of limu manuwea and ogo (another kind of seaweed) for commercial purposes.

"I think they're going to have to do something because the grounds are getting wiped out," said Tamada's son Gerald. "We use 150-200 pounds (of ogo) a week."

Gerald Tamada said the family store has been able to find a commercial grower of ogo, the most commonly used species of limu. But he has had to substitute sesame seed oil for the species known as limu-kohu because it has become so rare.

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Hana's Poisonous 'Limu' May Hold Key to Cancer

By Jeanne Ambrose
Star-Bulletin Writer

Star-Bulletin Health Page

Advice for You and the Latest Medical News

A powerful poison found in limu-make-o-Hana — the deadly seaweed of Hana, Maui — may lead to medical breakthroughs in cancer and heart disease research, according to a University of Hawaii chemistry professor.

Richard E. Moore has been the principal researcher in unraveling the potent secrets of limu-make-o-Hana to isolate the toxin and determine its molecular makeup — no easy task. In fact, Moore has been working on the problem for more than 20 years.

The results of his research look very promising, he said. The poison isolated from limu-make-o-Hana, called palytoxin, "ranks among the most poisonous substances in nature," Moore said. Those who have come in contact with it have become ill after touching or breathing the toxic substance.

"Most people think of poisons as things to stay clear of," he said. "But there are several poisons that are used in medicine."

For example, Moore said, tubocurarine, derived from curare which is used by natives of South America to poison the tips of their hunting arrows, is used as a muscle relaxant.

FURTHER RESEARCH is planned to determine if the palytoxin from limu-make-o-Hana can be used as an active ingredient in medicine. However, the toxin holds its most promise "as a tool for basic research" to find out exactly how some forms of cancer and heart disease progress, Moore said.

Research done in Japan has shown that palytoxin may be a cancerous tumor promoter — a substance that is not a cancer-causing agent but which acts on cells exposed to carcinogens, Moore said.

Although a carcinogen itself may be responsible for producing tumors, when a tumor promoter also comes in contact with the cells, the tumor may appear sooner than if the promoter were not introduced.

"There is intensive research in how tumor promotion actually works," Moore said. "The development of a tumor is exceedingly complex. This particular material is one of a few substances labeled as naturally occurring tumor promoters."

More research may lead scientists to discover exactly how the palytoxin works to promote tumors and what preventive action or drug is necessary to combat tumor promoters, Moore said.

"If we find out where and how it acts we should be able to develop materials that counteract that specific action," he said.

THE SAME HOLDS true for certain heart diseases because of the disturbances caused to the heart by the toxin, he said. If the mechanism of action were discovered, appropriate preventive measures or counteraction could be taken, he said.

Moore's work with limu-make-o-Hana began in the early 1960s after a group of collectors from the Hawaii Institute of Marine Biology gathered the limu from a tidal pool in Hana.

Moore and his co-workers were interested in finding out about an ailment that affected some people after they ate certain coral reef fishes. They suspected the ailment, called ciguatera, was caused by an algae eaten by the fish and passed on through the food chain to humans who ate those fish.

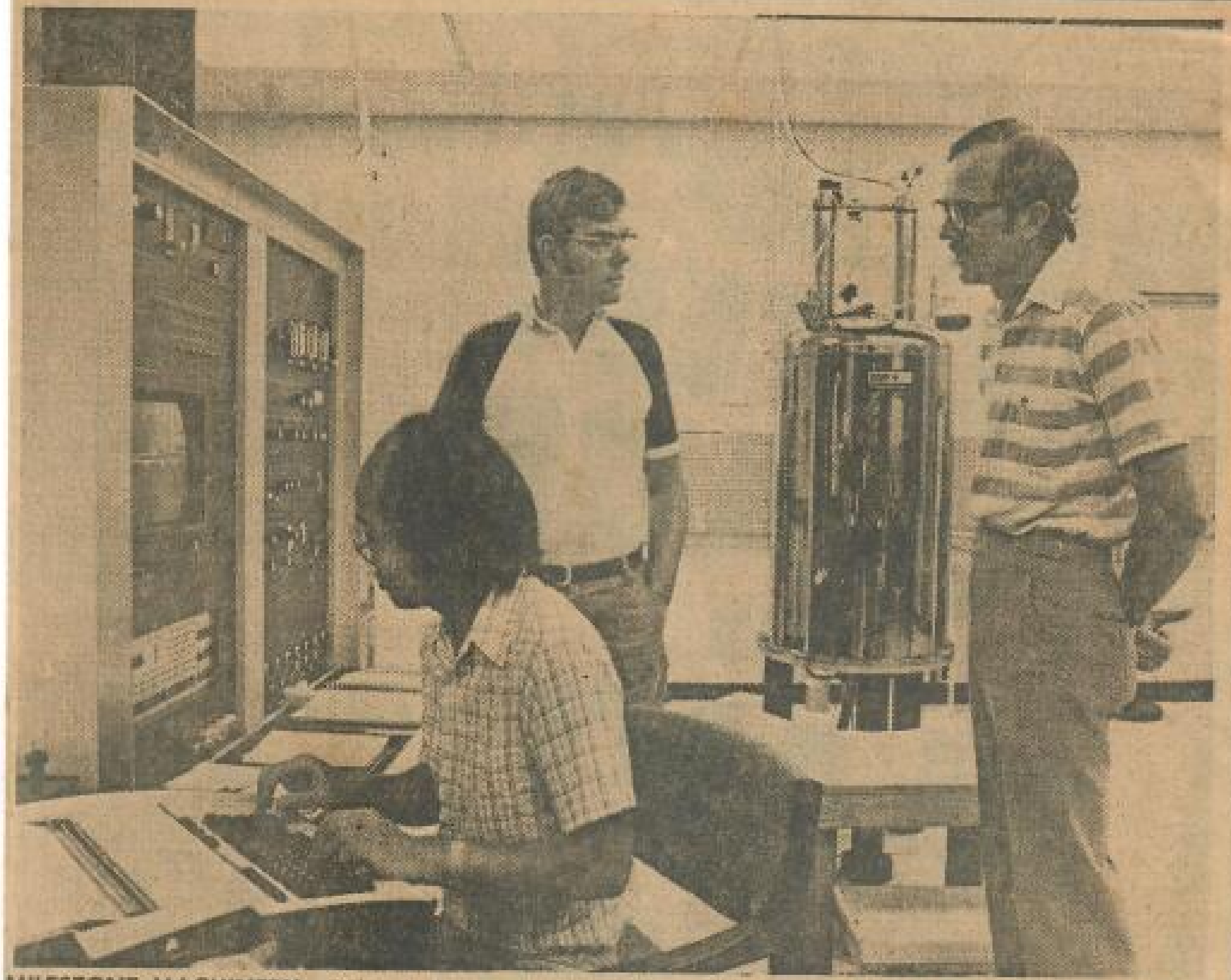
All signs pointed to limu-make-o-Hana as the culprit, so it was gathered for further study. Research revealed that the limu isn't really seaweed after all, Moore said. Instead, it is a coelenterate, an invertebrate animal lacking a true body cavity, such as jellyfish, sea anemones and coral. It is known scientifically as Palythoa toxica.

Moore's efforts to isolate the toxin from the so-called limu and determine its molecular structure spanned two decades and several states.

BECAUSE HAWAII did not have the equipment Moore needed to study and break down the components of the toxin, he traveled back and forth to the Mainland about once a month using equipment available there.

Recently the University of Hawaii obtained its own $300,000 nuclear magnetic resonance spectrometer so similar research can be done here. The spectrometer "is the most important tool for modern organic structure determination," Moore said. Cost of the equipment was shared by the National Science Foundation and the state of Hawaii.

Moore's research into limu-make-o-Hana included tracking down Hawaiian legends about the organism. According to legend, it be-



MILESTONE MACHINERY—Richard Moore, at right, talks to visiting researcher Robin Kinnel of New York while graduate student Roy Okuda types information to be sent to the computer. In the background is the University of Hawaii's new nuclear magnetic resonance spectrometer, a machine similar to the one Moore used to isolate the components of the limu-make-o-Hana toxin. —Star-Bulletin Photo by John Titchen.

came poisonous after a group of fishermen, suspicious of a man they thought was responsible for the disappearance of several missing fishermen, set the suspect on fire and threw his ashes into the sea.

Where the ashes were strewn, the poisonous limu-make-o-Hana grew. It appeared that pool in the Mu'olea region of Hana below the slopes of Haleakala was the only area where the poisonous substance could be found.

THE BAD LUCK attributed to the limu-make-o-Hana even seemed to affect the biologists who collected it. On the day the first major batch of seaweed was gathered by marine biologists, a mysterious fire destroyed the Hawaiian Marine Laboratory where the investigation originated, Moore said.

That was Dec. 30, 1961, and shortly thereafter, when another collection was made, the collector became dizzy and developed flu-like symptoms that lasted more than a week. He had been exposed to the toxin, which seeped into tiny scratches in his bare feet and hands while he was gathering the limu.

Several similar stories abound, but they didn't deter Moore in his research. The results of his work have earned him national recognition.

He was featured in a recent article in Chemical and Engineering News, the journal of the American Chemical Society. He also received the UH Chancellor's Award with Distinctive Merit for his work with palytoxin and other achievements.

A story about the "deadly seaweed of Hana" written by Moore, Philip Helfrich, director of the Hawaii Institute of Marine Biology, and Gregory M.L. Patterson, a biologist in the UH chemistry department, is scheduled to appear in Oceanus magazine this summer.

CURRICULUM VITAE

Dennis J. Russell, B.A., M.S., Ph.D.
Associate Professor of Biology
Seattle Pacific University

Personal:

Birth:	October 15, 1945, Portland, Oregon
Family:	Susan Carol Russell (married June 17, 1967) Allen Jess Russell (January 5, 1969)
Education:	Cascade College, B.A. (Biology-German, 1967) University of Washington, M.S. (Botany, 1969) University of Hawaii, Ph.D. (Botany, 1981)
Awards & Honors:	MAGNA CUM LAUDE, Cascade College, 1967 National Defense Education Act IV Fellowship, 1971 National Science Foundation Grant, 1976 FELLOW of Seattle Pacific University, 1984
Professional:	Associate Professor of Biology, Seattle Pacific University, 1985-1986 Assistant Professor of Biology, Seattle Pacific University, 1978-1985 Teaching Assistant, Department of Botany, University of Hawaii, 1974-1977 Teaching Assistant, Department of Botany, University of Washington, 1967-1969
Military:	Biological Assistant (MOS LGER 01H30) Munitions Command, Operations Research Group, Edgewood Arsenal, Aberdeen Proving Grounds, Maryland. United States Army, Specialist 5, 1969-1971 Honorable discharge
Laboratories visited:	Friday Harbor Laboratories, University of Washington, 1968-1969 Hawaii Institute of Marine Biology (HIMB), University of Hawaii, 1971-1978 Pacific Equatorial Research Laboratories (PERL), University of Hawaii, Kiribati, 1972-78
Societies:	American Phycological Society International Phycological Society Western Society of Naturalists

Consultant:	Environmental Consultants Inc., Kaneohe, Hawaii Sea Turtle Recovery Team, National Marine Fisheries (NOAA), Honolulu, Hawaii Natural Toxins & Cancer Research Laboratories, Honolulu, Hawaii
Civic:	Fine Arts Commissioner, City of Mountlake Terrace Editor, Cultural Arts, Stadium and Convention District of South Snohomish County, Newsletter.
Other:	Divemaster, Professional Association of Diving Instructors (PADI), 1984 Manager, The Kailuan Apartments, Kailua, Hawaii, 1976-1978 President, Graduate and Faculty Christian Fellowship, University of Hawaii, 1973-1976 Chairman, Faculty Research Committee, Seattle Pacific University, 1984-1985 Judge, for the best papers awards at the 64th Annual Meeting of the Western Society of Naturalist, 1983, Simon Fraser University, B.C., Canada. FELLOW of Seattle Pacific University, 1984 Sales Representative, P. and J. Gem Trading Enterprise, 1977-1978

Publications (refereed):

Russell, Dennis J. and Richard E. Norris. 1971. Ecology and taxonomy of an epibiotic diatom. Pac. Sci. 25(3): 357-367.

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Russell, D. J. The Introduction and Spread of *Acanthophora spicifera* in the Hawaiian Islands. J. of Phycology

Courses taught at Seattle Pacific University (1978-1986):

Biological Sciences (Bio 1100) 5 cr.

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General Microbiology (Bio 3351) 5 cr. with laboratory

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ECOLOGIA MARINA
Casilla 114-D Santiago, Chile

Santiago, April 21st., 1987

Dr.
George H. Balazs
Zoologist
U.S. Department of Commerce
National Marine Fisheries Service
2570 Dole Street
Honolulu, Hawaii 96822-2396
U.S.A.

Dear Dr. Balazs:

Thank you for your letter of April 3, 1987, your observation on the resistence to digestion of several algae through green turtles and your kind offer to send fecal samples. Right now we are involved in other projects related to digestion and have no capacity to receive your samples and work on them. If in the future we could do the work I will return to you for the samples.

Once again, thank you for your kindness.

Sincerely yours,

B. Santelices

Dr. BERNABE SANTELICES

BS/xag.

Sent to Dennis 5/29

INVENTORY OF ALGAE SPECIMENS

#1 - taken 05 May 86 from green turtle
carcass salvaged from beach - Pompano Beach, FL (Broward County)
cause of death unknown
38.10 cm, curved carapace length
33.02 cm, Curved carapace width

#2 - taken 09 June 86 from green turtle
carcass salvaged from beach, Lauderdale-by-the-Sea, FL (Broward County)
propeller cut to carapace
35.00 cm, curved carapace length
30.00 cm, curved carapace width

#3 - taken 15 June 86 from green turtle
carcass salvaged from beach - Hillsboro Beach, FL (Broward County)
propeller cut to carapace
approx 40 cm, curved carapace length

#4 - taken 15 June 86 from green turtle
carcass salvaged from beach - Hillsboro Beach, FL (Broward County)
propeller cut to carapace
approx. 70 cm, curved carapace length

#5 - taken 18 July 86 from green turtle
carcass salvaged from beach - Pompano Beach, FL (Broward County)
cause of death unknown
72.39 cm, curved carapace length
68.58 cm, curved carapace width

#6 - taken 07 February 87 from green turtle
carcass salvaged from beach, Lauderdale-by-the-Sea, FL (Broward County)
propeller cut to carapace
43.00 cm, curved carapace length
36.00 cm, curved carapace width

#7 - taken 19 April 87 from sand bottom
approx. 600 yards offshore (between 1st and 2nd reef areas)
Lauderdale-by-the-Sea, FL (Broward County)

#8 - taken 20 May 87 from limestone substrate
approx. 150 yards offshore on 1st reef
Lauderdale-by-the-Sea/Galt Ocean Mile, FL (Broward County)

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Dear George,

Thanks a lot for your correspondence especially your information on foraging habitat and techniques for sampling stomach contents. Great information and really helpful to us.

Well we decided to take you up on your offer to have your phycologist analyze our algae. We enclosed small specimens from all our salvaged turtles and two samples taken from the substrate. We appreciate your help with this, as it will probably allow us to locate the feeding areas for the population. If your contracted PHd can't do them all, any analysis would be helpful, especially # 5, 6, 7, 8.

We are going to begin double tagging as soon as we get back in the water. We are experiencing rough seas right now with strong easterly winds. Loggerhead nesting has begun, with a few leatherbacks, but the seas are nasty the main obstacle to double tagging

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has been the difficulty of maneuvering during shore dives without a boat. Hopefully we are going to buy a small inflatable.

Thanks again for all your help. Your support along with Barbara Schroeders and Larry Ogren has been a real motivator to continue these long dives at night.

Sincerely

Bob + Jeanne
W.

Effects of Light Intensity on the Morphology and Productivity of *Caulerpa racemosa* (Forsskal) J. Agardh

RUSSELL D. PETERSON

Reprinted from MICRONESICA, Vol. 8, Nos. 1 and 2, 1972
Printed in Japan

Effects of Light Intensity on the Morphology and Productivity of *Caulerpa racemosa* (Forsskal) J. Agardh[1]

RUSSELL D. PETERSON[2]

Abstract

Six varieties and three additional growth forms of *Caulerpa racemosa* (Forsskal) J. Ag. were found on Guam's fringing reef flat. Variation in length and number of both assimilators and rhizoids and in the spacing of ramuli with light intensity was demonstrated for the varieties *ecifera* and *lamourouxii*. Characteristics of other varieties developed on specimens collected in the field after they were placed under altered laboratory light intensities. Seasonality of *C. racemosa* was correlated with the number of midday minus tides.

Productivity data suggested the adaptation of var. *ecifera* and var. *lamourouxii* f. *requienii* to habitats of high and low light intensity, respectively. Variety *lamourouxii* f. *requienii* had a lower compensation point, larger P/R (gross photosynthesis/respiration) value, and higher net photosynthetic rate than var. *ecifera* at low light intensities. When exposed to full sunlight the P/R value and net photosynthetic rate of var. *ecifera* exceeded those of var. *lamourouxii* f. *requienii*, which dropped at that intensity. Chlorophyll a and carotenoid concentrations of field-collected var. *lamourouxii* specimens were approximately twice those of var. *ecifera*. Chlorophyll a, but not carotenoid content, was found to decrease with exposure to increasing light intensity for specimens originally classified as var. *lamourouxii*.

The relationships of morphologic and productivity factors to light intensity provide evidence for their environmental rather than genetic control. Reference to *C. racemosa* growth forms as ecophenes is suggested.

INTRODUCTION

Caulerpa racemosa (Forsskal) J. Agardh is a siphonaceous green alga exhibiting an extreme degree of variation in its growth form. It is circumtropical in distribution (Eubank, 1946) and is characterized by having a prostrate cylindrical rhizome with rhizoids below and upright assimilators bearing protuberances termed ramuli. The original descriptions of various forms of this taxon were often based on few or single specimens, and it was believed that each form represented a distinct species (Børgesen, 1907). Subsequent examinations of more extensive collections by Weber-van Bosse (1898), Svedelius (1906), Børgesen (1907), Gilbert (1942),

---

[1] Contribution No. 23, The Marine Laboratory, University of Guam. This paper represents a thesis submitted to the Graduate School of the University of Guam in partial fulfillment of the requirements for the Master of Science degree in Biology.

[2] The Marine Laboratory, University of Guam, Agana, Guam 96910.

Micronesica 8(1/2):83-98. 1972 (December).

Eubank (1946), Cribb (1958) and Taylor (1950, 1960) have led to a recognition of the gradation between these morphologic forms and normally a reduction of their taxonomic level to varieties of the single species *C. racemosa*.

Borgesen (1907) and Eubank (1946) suggest that environmental factors may control the development of *C. racemosa* varieties. The ability of transplanted specimens of *C. racemosa* to develop characteristics of other varieties has been demonstrated by Tandy (1933), although he did not hypothesize any specific environmental cause. Rehm (1969) also observed changes in the morphology of *C. racemosa* specimens brought into the laboratory and suggested that changes in ramuli shape were initiated by reduced light. The possibility of a genetic basis for separation of varieties is also suggested by Eubank (1946) and by Taylor (1950). Recommendations for more critical field and laboratory studies have appeared in the literature (Borgesen, 1907; Gilbert, 1942; Eubank, 1946).

Studies involving *C. racemosa* varieties collected during floristic surveys or observed in the field often include limited descriptions of the environmental conditions under which these specimens were collected or observed. Light intensity has been mentioned in this respect. For example, Borgesen (1907) refers to the extremely high light intensity of the reef flat habitat occupied by var. *uvifera*. Reduced light intensity can also be associated with the deep water habitat reported for var. *lamourouxii* (Borgesen, 1907; Taylor, 1950). The agitated water and bubbles formed by braking waves at the reef margin would serve to reduce the light intensity at the substrate as compared to the calmer reef flat. Borgesen (1907) has reported var. *clavifera* from such areas.

The tendency for *C. racemosa* varieties to be somewhat restricted to specific habitat situations raises the question of environmental versus genetic control of the morphologic characteristics used in their classification. The presence of intermediate forms (Weber-van Bosse, 1898; Borgesen, 1907; Eubank, 1946; Taylor, 1950, 1960) tends to weaken the argument for genetic control as does the fact that characteristics of two or more varieties are occasionally found on the same specimen (Tandy, 1934; Eubank, 1946; Taylor, 1960; Rehm and Almodovar, 1971).

Borgesen (1907) mentioned a general tendency for radial development of ramuli around the assimilator axis in shallow water versus bilateral development in deeper water for the species *C. racemosa* and *C. cupressoides* (West) J. Agardh. He also stated his belief that this tendency represents an ecological adaptation. Such an adaptation could be related to the water depth effect on light intensity and the vital requirement of light for plant growth. Round (1968) has reviewed some effects of light intensity on the morphology of algae and Daubenmire (1959) summarized such effects for higher terrestrial plants.

The phenomena of intermediate forms and multiple varietal characteristics were observed on Guam and strengthened my own speculation that many, if not all, of the *C. racemosa* varieties are the result of environmental variation within and between habitats. References to light in past literature plus my own observation that differences in light intensity occurred between many of these habitats suggested

that this environmental factor may influence the morphology of *C. racemosa*. Morphologic characteristics which have been used in identifying and describing *C. racemosa* varieties include concentration of rhizoids, length and concentration of assimilators, and especially the spacing and shape of ramuli.

The purpose of this study was to test two hypotheses: 1) that the morphology of *C. racemosa* is influenced by light intensity and 2) that characteristics of the morphology of *C. racemosa* function adaptively to ensure optimum productivity for the light intensity under which they develop.

MATERIALS AND METHODS

All specimens used during the course of this study were collected from the reef flat and margin on the eastern coast of Guam in an area extending from the University of Guam Marine Laboratory on Pago Bay 10 km south to Asanite Bay. Specimens were kept in plastic aquaria supplied with water from the Marine Laboratory seawater system.

Experiments in this study utilized specimens of the varieties *uvifera* and *lamourouxii*. One practical reason for using these varieties was that var. *uvifera* was relatively abundant on the reef flat and var. *lamourouxii*, though not always as available on the reef, was easily maintained in the laboratory. Another more important reason was that the varieties *uvifera* and *lamourouxii* were characteristically found in habitats of high and low light intensity, respectively. This allowed the consideration of pre-experimental light conditions and growth form in analyzing the effects of light intensity.

Most of the var. *uvifera* specimens on Guam had crowded, spherical ramuli conforming to Borgesen's (1907) description of this variety in the Danish West Indies and to a drawing by Weber-van Bosse (1898) of var. *uvifera* f. *intermedia* (Pl. XXXIII, Fig. 24a). The more typical form, described by Taylor (1960), has ramuli which are crowded but slightly compressed tangentially with the surface of the assimilator. This latter form was present, though rare, on Guam and all references to var. *uvifera* in this paper, unless otherwise stated, refer to the former morphologic condition.

FIELD AND TRANSPLANT OBSERVATIONS

Observations of the distribution of *C. racemosa* varieties on the reef flats of Guam were made between December, 1970 and November, 1971. Seasonal abundance and degree of exposure to light and low tide were noted. A number of specimens collected in the field were transplanted into the laboratory at various light intensities. Photographs were taken to record changes in their morphologic characteristics.

GROWTH APPARATUS EXPERIMENTS

To quantify the effects of light intensity on the morphology of *C. racemosa*, specimens were maintained in the apparatus shown in Fig. 1. This trough-like construction was made with pieces of clear plexiglas joined with epoxy glue. The partitions had double coats of white epoxy paint, and the entire outer surface was first painted white to allow a uniform white interior background. The outer surface was then painted black to further limit the entrance of light from outside the trough.

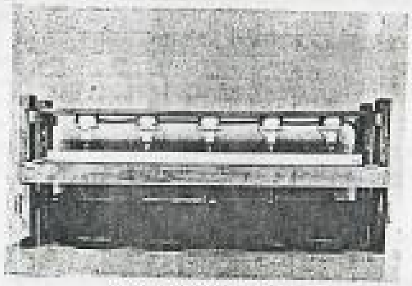

Fig. 1. Growth apparatus.

The light sources were four 40-watt cool-white fluorescent tubes and five 150-watt incandescent bulbs. To vary light intensity, 1/32 inch mesh nylon screen stapled to wooden frames was placed over the tops of sections A (eight layers of screen), B (four layers) and C (two layers). Section D had a wooden frame without screen attached. This created light intensities of 0.5, 3.5, 8.3 and 21 kilolux in sections A through D, respectively. Light intensity measurements were made at the level of the algae with a submarine photometer. The microampere readings of the submarine photometer were converted to foot candle values after comparative readings were made with a GE Type 213 light meter. Kilolux values were then determined by the conversion factor: 1 lux equals 0.093 foot candle. A 12/12 hour light/dark cycle was controlled automatically for all experiments by a timer wired into the lighting system. This light cycle closely simulated the photoperiod on Guam (U.S.D.C., 1967).

Water from the seawater system ran into an aquarium where suspended material was allowed to settle. Water was siphoned from a flask within that aquarium through plastic hoses into the influent section at the extreme left of the trough at a rate of 1.3 liters per minute. Water flowed through the trough beneath and at the sides of the partitions. These slight openings allowed some light to pass between sections but were necessary for water movement. Water flowed out of the trough over the lower end wall of the effluent section at the extreme right of the trough.

Aluminum foil was placed between the incandescent bulbs and the two fluorescent light fixtures to prevent overheating. Foil was also placed over the influent section to prevent light from passing between it and section A. Radiant heat from the lights caused an increase in water temperature, especially at the surface. This factor was reduced by the continuous flow of water, and when in use the average midday temperatures at the level of the algae were 29.8, 29.8, 30.6, and 31.2°C for sections A through D, respectively.

Before specimens were placed in the growth apparatus they were checked for foreign material which, if present, was removed. Two-pound test monofilament line was tied to the rhizome of each specimen and to pieces of embossing tape marked to identify the specimen number and section of the trough in which it was maintained. Pieces of plexiglas with grooves cut by a coping saw were attached at their ends to the sides of each section. The embossing tape was slid into these grooves to hold the specimens approximately eight cm below the water surface.

Growth which occurred during six to 10 day exposure periods was measured by the following procedures:

1. Specimens were removed from the apparatus and placed in a seawater-filled tray. A millimeter rule was used to measure rhizoid length and distance from rhizome tip, and assimilator length and distance from rhizome tip. Number of ramuli per assimilator was also determined.
2. Specimens were removed from the apparatus and placed into a seawater-filled tray at the bottom of which was a sheet of two mm ruled graph paper fused between two pieces of plexiglas. A millimeter rule was also placed next to each specimen before it was photographed with slide film. When these slides were later projected onto a screen the above-mentioned measurements were made.

Growth responses of 17 var. *uvifera* specimens freshly collected from the same shallow reef flat depression were made with a millimeter rule. Measurements were made after a six day exposure period within the growth apparatus. Data for seven other var. *uvifera* specimens which did not appear in good condition during or at the end of this period are not included. The growth responses of 26 var. *lamourouxii* specimens within the growth apparatus were also determined. These specimens had flattened blade-like assimilators with very few ramuli at the start of the experiments and were developed in the laboratory at approximately 1.6 klux from material collected two months prior at the reef margin and identified as var. *clavifera*. The rarity of this form prohibited the use of field-collected specimens. Measurements were made by millimeter rule for 14 var. *lamourouxii* specimens after seven days of growth. The remaining 12 specimens were measured photographically after 10 days of growth. Mature assimilators and rhizoids were defined as those occurring on rhizome segments more than three days old. During a preliminary study it was determined that the majority of growth occurs during that period of time. When two or more rhizoids occurred less than two mm apart the longest length was recorded.

PRODUCTIVITY AND PIGMENTS

Oxygen production and consumption of specimens freshly collected from high, var. *uvifera*, and low, var. *lamourouxii* f. *requienii*, light intensity habitats were determined over a wide range of light intensities. The intensities and sources of light were (1) 0.8 klux, fluorescent tubes 1.5 m from specimens; (2) 3.7 klux, fluorescent tubes 0.6 m from specimens; (3) 13 klux, fluorescent tubes 10 cm from specimens; (4) 50 klux, 200 watt incandescent bulbs 7.5 cm from specimens; and (5) 100 klux, natural sunlight at midday with a clear sky.

The light- and dark-bottle oxygen technique (Gaardner and Gran, 1927) was used to measure photosynthesis and respiration. Results of this type of experiment are considered to represent biological activity (Hedgepeth, 1957) or food produced (Odum, 1959). Seawater samples were analyzed for dissolved oxygen according to the alkali-azide modification of the Winkler technique (A.P.H.A., 1965). Titrations were completed within eight hours after the samples were "fixed".

Twenty freshly collected specimens were used, two of each variety at each light intensity. Prior to use, specimens were checked for epiphytic algae and invertebrates which, if present, were removed. The following sequence of steps was performed to get replicate oxygen exchange data for each specimen in both light and dark bottles.

1. Specimens were exposed to the experimental light condition for one hour before testing.
2. Each specimen was individually incubated for 60 to 75 minutes in a 300-ml BOD bottle made light-tight with two layers of aluminum foil.
3. Specimens were placed in uncovered BOD bottles for three successive 30- to 38-minute incubation periods.
4. Same as step two.
5. Wet weight of each specimen was determined as described below.

During both light and dark incubation periods BOD bottles were kept under identical light conditions. An incubation medium of continuously running seawater was used to prevent heating. Seawater used to fill the BOD bottles was first filtered through 0.45-micron membrane filters to remove plankton that could otherwise affect the results. This water was always used within 30 hours of filtering. To eliminate possible effects of periodicity of oxygen production, all trials began at approximately 1100 hours. Such a periodicity has been demonstrated for phytoplankton (Doty and Oguri, 1957).

An average of two seawater samples taken at the start of each incubation period gave an initial oxygen value. The two dark period values were subtracted from their corresponding initial values before averaging to give a measure of each specimen's respiration rate. Net photosynthesis rates were obtained from an average of the differences between each of the three light period measurements and their initial values. Adding the rates of respiration and net photosynthesis gave a gross photosynthesis value for each specimen.

Percent dry weight was determined for 10 specimens each of the varieties *uvifera* and *lamourouxii*. Wet weight was first determined for each specimen on a Mettler Model H 10 Balance after foreign material was removed and surface water had been absorbed by wrapping and blotting in absorbent tissue for 10 seconds. Specimens were then dried overnight at 100–105°C. After removal from the oven specimens were placed in a desiccator and allowed to cool for a few minutes before their dry weight values were recorded.

Chlorophyll a and carotenoid contents were measured for six specimens each of var. *lamourouxii*, collected in shaded areas of a reef flat depression, and var. *uvifera*, collected at the outer reef flat. These measurements were also made after 24 days of exposure for 12 specimens originally identified as var. *lamourouxii* which were randomly placed three each in sections A through D of the growth apparatus.

Wet weight value was recorded for each specimen before it was ground, under reduced lighting, in 100% acetone with a mortar and pestle. The acetone and plant residue, plus 100% acetone used to rinse the mortar and pestle, were poured into 50 ml centrifuge tubes. Additional acetone was added to make a total of 30 ml of 100% acetone. The tubes were covered and placed overnight in a refrigerator at 8°C. When removed they were again protected from light and allowed to return to room temperature. The percent dry weight values described above, were used to estimate the amount of water in each specimen and additional distilled water or 100% acetone was added to make a final acetone concentration of 90% for each sample. The tubes were then centrifuged at 3100 rpm for 10–15 minutes.

Samples of the acetone solution were individually placed in a cuvette to record optical density at 665, 645, 630, 510 and 480 millimicrons with a Beckman Model B spectrophotometer. The formulas developed by Richards and Thompson (1952) were used to calculate pigment concentrations. Twelve samples were also measured at 750 millimicrons as a turbidity check.

RESULTS AND DISCUSSION

FIELD AND TRANSPLANT OBSERVATIONS

Nine *Caulerpa racemosa* growth forms were found on Guam's fringing reef flat. Varietal and form names, and general habitat descriptions are given below. Observations of abundance are indicated by the terms rare, common and abundant. These are subjective estimates based on the relative number of times each form was encountered during the period of maximum *C. racemosa* growth.

1. Var. *clavifera* (Turner) Weber-van Bosse; Borgesen, 1907: p. 47, Fig. 25 and 26.

   Outer reef flat to margin, growing in tangled mats. Subjected to breaking waves, low tide exposure and high light intensity. Abundant.

2. Var. *clavifera* (Turner) Weber-van Bosse f. *reducta* Borgesen, 1907: p. 48, Fig. 27.

   Outer reef flat to margin over raised rocks and at edges of depressions. Subjected to breaking waves, low tide exposure and high light intensity. Rare.

3. Var. *uvifera* (Turner) Weber-van Bosse, 1898: Pl. XXXIII, Fig. 6.

Thick mats within 0.5 m deep, outer reef flat depressions, protected from breaking waves and low tide exposure. Isolated specimens on outer reef flat, exposed to breaking waves and low tide exposure. All exposed to high light intensity. Rare.

4. Var. *uvifera* (Turner) Weber-van Bosse f. *intermedia* Weber-van Bosse, 1898: Pl. XXXIII, Fig. 24.

Outer reef flat singly or in patches up to 3 m wide. Subjected to extreme high light intensity and low tide exposure. Occasionally in shallow reef flat depressions. Abundant.

5. Var. *macrophysa* (Kutzing) Taylor; Eubank, 1946: p. 428, Fig. 2n.

Outer reef flat, subjected to high light intensity and low tide exposure. Others within semi-shaded reef flat depressions and under rocks protected from high light intensity and low tide exposure. Common.

6. Var. *lamourouxii* (Turner) Weber-van Bosse, 1898: Pl. XXXII, Fig. 1–4 and 6.

Shaded areas within reef flat depressions. Protected from breaking waves, high light intensity and low tide exposure. Rare.

7. Var. *lamourouxii* (Turner) Weber-van Bosse f. *requienii* (Montagne) Weber-van Bosse, 1898: Pl. XXXII, Fig. 5 and 7.

Beneath ledges and thick mats of other var. *lamourouxii* thalli within reef flat depressions. Protected from breaking waves and low tide exposure. Minimum light intensities. Rare.

8. Var. *occidentalis* (J. Agardh) Børgesen, 1907: p. 49, Fig. 29.

Within slight to one m deep reef flat depressions and splash pools. Protected from breaking waves and low tide exposure. Subjected to high light intensity. Common.

9. Var. *peltata* (Lamx.) Eubank, 1946: p. 428, Fig. 2r and 5s.

Reef flat and margin. Usually within an articulated coralline algae mat on the reef flat or within slight reef flat depressions. One specimen on side of rock. Occasionally within mats of var. *clavifera*. Sometimes subjected to breaking waves and low tide exposure. Protected from high light intensity. Common.

There was a definite seasonality in the abundance of *C. racemosa* on the reef flats of Guam. Seasonality of *C. racemosa* has also been reported for Bermuda (Bernatowicz, 1952; Taylor and Bernatowicz, 1969). Development of *C. racemosa* began in October and November, and maximum growth occurred between December and May. Growth declined and specimens were increasingly difficult to find during the period June through September.

Seasonality of *C. racemosa* may be controlled by minus tides occurring at midday. Midday minus tides, whose lowest levels occurred between 0900 and 1500 hours, were absent from the latter part of October through February. They began in March and peaked at 12 per month during July and August before again declining

in number. Exposure to midday minus tides has also been suggested to explain seasonality of *Sargassum duplicatum* J. Ag (=*S. cristaefolium* C. Ag.) on Guam (Tsuda, in press).

The correlation between decreased abundance of *C. racemosa* and midday minus tides may be due to desiccation damage resulting from atmospheric exposure during periods of high light intensity. Rehm (1969) found that *C. racemosa* assimilators could survive one hour of atmospheric exposure in the shade but not one hour 15 min. Assimilators exposed to direct sunlight did not recover after a 15 min. exposure.

The possibility that low tide exposure controls seasonality is further evidenced by the fact that areas of most prolonged growth were reef flat depressions not exposed at low tide, and the reef margin area which is periodically washed by breaking waves even during the lowest tide levels. In addition, laboratory specimens never exposed to the atmosphere showed no seasonal effect and var. *chemnitzia* (Esper) Weber-van Bosse was abundant at a depth of approximately 30 m off Guam in September.

Maturity may have an effect on *C. racemosa* morphology, but it is difficult to separate its effect from that of light intensity. For example, var. *peltata* was relatively common during the developmental period of the growth season, in some areas showing an almost continuous transition from small specimens with flattened ramuli growing within a substrate of mat-like articulated coralline algae (Fig. 2) to larger specimens with hemispherical ramuli growing over the top of this substrate (Fig. 3). This latter form was classified as var. *macrophysa*. While the specimen with enlarged ramuli (Fig. 3) may be a more mature form of var. *peltata*, it is also

Fig. 2. A var. *peltata* specimen growing within an algal mat of articulated coralline algae.

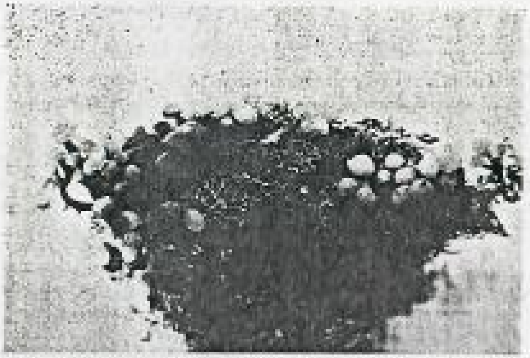

Fig. 3. A var. *macrophysa* specimen growing at the top of an algal mat of articulated coralline algae.

possible that it results from increased light intensity at the surface of the substrate. Possibly the conditions within the algal mat, including a reduced light intensity, are favorable for germinating zygotes with var. *peltata* being an early developmental form. Carrying this speculation further, a positive phototropic response of this form may eventually bring it closer to the surface of the substrate where increased light intensity results in form changes. Undisturbed *Zonaria farlowii* specimens kept at low light intensity have shown a phototropic growth response, with curving of their apical region resulting in a perpendicular orientation to the light source (Dahl, 1971).

Børgesen (1925) referred to var. *peltata* as a separate species but recognized the presence of forms transitional to *C. racemosa*. Svedelius (1906) also classified var. *peltata* as a separate species but noted swollen ramuli at the upper part of some assimilators. These reminded him of var. *clavifera* (which he also categorized as a species) except that they had a border as evidence that they were originally flat. No such border was observed on Guam specimens. Taylor (1950) in placing specimens into var. *macrophysa* referred to them as an extension of the variation shown by his var. *clavifera* specimens. If var. *clavifera* is a more mature form, developing from var. *macrophysa*, the question again arises whether such a form change results from aging or from increased light intensity. Variety *macrophysa* was observed on the surface of the outer reef flat mostly during the developmental period indicating it had not been there long. Thus, such specimens (i.e., Fig. 3) may have just reached the top of their somewhat protective coralline algae substrate and with further growth at this more exposed position would develop a var. *clavifera*-like form.

Variety *clavifera*, in occurring close to the reef margin in tangled mats, would be somewhat shaded both by especially agitated water in that area and by other var. *clavifera* thalli within these mats. Variety *uvifera*, on the other hand, was very abundant on the reef flat at some distance from the margin and occasionally near the margin during the maximum growth period. Although it often occurred in patches up to three meters in diameter, the thalli were not nearly as tightly packed as they were in the var. *clavifera* mats. It thus seems a reasonable speculation that var. *uvifera* results from growth in areas exposed to higher light intensities than genetically identical thalli which develop into var. *clavifera* at the reef margin.

A number of the specimens transplanted into the laboratory developed characteristics associated with other varieties. These form changes occurred in the areas of new apical growth at the tips of both assimilators and rhizomes. The form of this alga, once completely developed, was never observed to change.

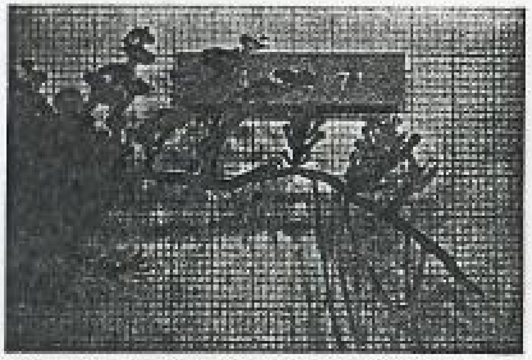

Fig. 4. Single specimen showing characteristics of var. *uvifera* (left), var. *peltata* (center), and var. *laetevirens* (right) when exposed to different light intensities.

Figure 4 is a photograph of a specimen classified as var. *uvifera* when collected. During five days of growth at 3.8 klux in a laboratory aquarium the ramuli became extremely flattened and fewer in number as is characteristic of var. *peltata*. When placed in the 21 klux section of the growth apparatus for another five days the number of new ramuli increased and they became gradually expanded to cylindrical in form, closely resembling var. *laetevirens* (Mont.) Weber-van Bosse.

During three weeks in section A of the growth apparatus, another var. *uvifera* specimen had rhizome growth without assimilator development. When the specimen was placed in an aquarium at 1.6 klux for one week, assimilators developed both from the rhizome and from two ramuli (Fig. 5). These new assimilators had

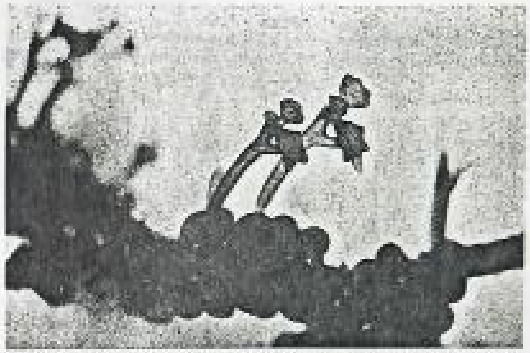

Fig. 5. Variety *uvifera* developing new assimilators from ramuli of its original growth with new ramuli characteristic of var. *exigua*.

flattened ramuli as does var. *peltata*, however, some had indented margins characteristic of var. *exigua* (Weber-van Bosse) Eubank. This same specimen was next transferred to section D with a light intensity of 21 klux. Ramuli developed at this intensity increased in number but were not crowded, and they gradually expanded to a rounded but still somewhat flattened end. This new form was identified as var. *macrophysa*.

The var. *uvifera* specimen shown in Fig. 6 was collected on the reef flat and had crowded, spherical ramuli. During approximately six weeks at 3.5 klux the newly developed assimilators were flattened with bilaterally arranged ramuli, similar to those of var. *lamourouxii*. A number of var. *clavifera* specimens made this same form change.

A var. *peltata* specimen (Fig. 7) collected from a shaded position near the reef margin was placed in section D at 21 klux for six days. During the first three days at that intensity the ramuli became slightly spherically expanded at their tips. Ramuli developed during the following three days were more expanded but somewhat flattened and like the form taken by the Fig. 5 specimen at the same intensity, this specimen was identified as var. *macrophysa*.

These obvious changes in *C. racemosa* morphology under altered light intensities serve as evidence for environmental rather than genetic control of varietal differences. The flattened ramuli and blade-like assimilators developed at reduced light intensities result in increased surface area. This may have an adaptive function at low light intensities in allowing increased light absorption in comparison with forms having enlarged ramuli.

Transplanting specimens to extreme light conditions sometimes resulted in injury. For example, four var. *lamourouxii* specimens became flaccid and colorless, except for a short segment of the rhizome near the apex, after nine days exposure to full sunlight in an outdoor aquarium. Four similarly treated var. *uvifera* specimens remained healthy. Another group of specimens placed in an outdoor aquarium screened to receive 10% sunlight responded quite differently. All four var. *la-*

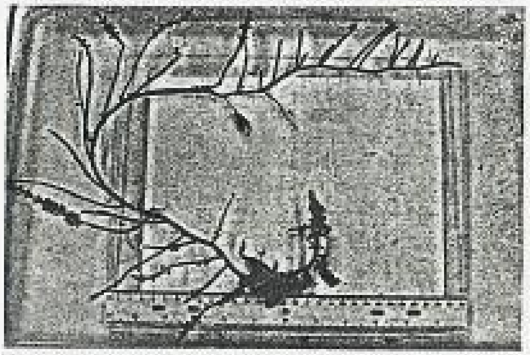

Fig. 6. Variety *uvifera* (center), changing form to that of var. *lamourouxii* when when exposed to 3.5 klux for six weeks.

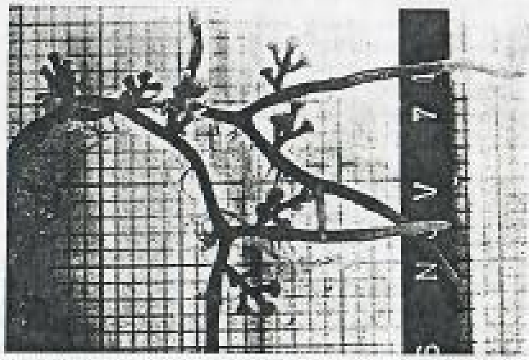

Fig. 7. Variety *peltata* (extreme left, bleached appearance) with ramuli becoming increasingly enlarged, to form var. *macrophysa*.

mourouxii specimens remained healthy whereas only one of four var. *uvifera* specimens survived. Dahl (1971) reported injury to *Zonaria farlowii* specimens collected or maintained at reduced light intensity upon exposure to full sunlight and suggested a cellular adaptation to the prevailing light intensity. *C. racemosa* may have a similar means of adaptation.

GROWTH APPARATUS EXPERIMENTS

Light intensity values were transformed logarithmically and statistical tests performed to test the effects of light intensity on all growth factors. An increase in the number of ramuli per unit length of assimilator with increasing light intensity was found to be significant for both var. *uvifera* (Fig. 8; $P < .005$) and var. *lamourouxii* (Fig. 9; $P < .025$) when tested by regression analysis. Although never presented as a ratio value, the spacing of ramuli is frequently referred to in keys and descriptions of *C. racemosa* varieties. For example, Taylor (1960) refers to

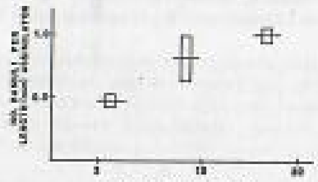

Fig. 8. Variety *uvifera*, number of ramuli per unit length of assimilator for assimilators developed on specimens maintained in sections B through D of the growth apparatus. The vertical bar represents two standard errors on either side of the mean, which is indicated by the horizontal line.

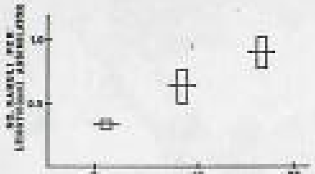

Fig. 9. Variety *lamourouxii*, number of ramuli per unit length assimilator for assimilators developed on specimens maintained in sections B through D of the growth apparatus. The vertical bar represents two standard errors on either side of the mean, which is indicated by the horizontal line.

the ramuli of var. *lamourouxii*, found in reduced light intensity habitats, as "... alternate, sub-opposite, or few and widely scattered."; var. *laetevirens*, reported from both semi-exposed and sheltered habitats (Cribb, 1958), as "... densely imbricate to widely spaced ..."; var. *clavifera*, found in high but less than maximum intensity due to agitated water and possibly self-shading within mats, as "... generally not crowded ..."; and var. *uvifera*, exposed to extreme high intensities, as "... crowded and imbricate ...". Thus, there seems to be a general tendency for varieties found in habitats of reduced light intensity to have more widely spaced ramuli. The fact that the spacing of ramuli, a characteristic used in classifying *C. racemosa* varieties, can be significantly related to light intensity for the varieties *uvifera* and *lamourouxii* indicated environmental rather than genetic control of this factor.

Assimilators did not initiate development at 0.5 klux, indicating that a minimum light intensity between 0.5 and 3.5 klux is required for initiation of assimilator development. Most of the ramuli of var. *lamourouxii* specimens developed at 3.5 klux were bilaterally arranged. Those developed at the higher intensities were generally radially arranged, as were those of the experimental var. *uvifera* specimens at all intensities. Most of the ramuli were gradually expanded from their base to a spherical tip and would be classified as var. *clavifera*, although some var. *uvifera* specimens developed more abruptly expanded ramuli at the highest intensity resulting in their classification as var. *occidentalis*.

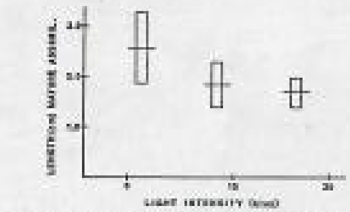

Fig. 10. Variety *uvifera*, length of mature assimilators developed on specimens maintained in sections B through D of the growth apparatus. The vertical bar represents two standard errors on either side of the mean, which is indicated by the horizontal line.

Length of assimilators did not vary significantly for either var. *uvifera* (Fig. 10) or var. *lamourouxii* (Fig. 11) when tested by analysis of variance. Both varieties did, however, show a tendency to develop shorter assimilators as light intensity increased. When analyzed separately by Student's t-test the assimilators developed at 3.5 klux were found to be significantly longer than those developed at 21 klux for var. *uvifera* ($P < .05$) but not var. *lamourouxii*. Decreased growth of terrestrial plant leaves, both coniferous (Whittaker and Garfine, 1962) and deciduous (Murray and Nichols, 1966), has also been related to increased light intensity.

Assimilator length is used by Taylor (1960) in his descriptions of *C. racemosa* varieties. For example, the varieties *clavifera*, *macrophysa* and *uvifera*, all found in relatively high light intensity habitats, are reported to have assimilator lengths of 1 to 11 cm, 3 to 6.5 cm and 1.5 to 2.5 cm, respectively. This is contrasted with the varieties *laetevirens* and *lamourouxii* whose lengths Taylor reported to be 12 to 30 cm and up to 16 cm, respectively. I have found var. *lamourouxii* only in shaded reef flat depressions on Guam. Borgesen (1907) reported var. *lamourouxii* to inhabit lagoon bottoms where the depth of water (20 to 30 m) would also reduce light intensity. Variety *laetevirens* was not found on Guam's reef flat although it developed from specimens originally classified as var. *uvifera* after being transplanted into reduced laboratory lighting. The fact that assimilator length, a morphological characteristic used in identifying *C. racemosa* varieties, can be related to light intensity is another indication that the form of this alga is under environmental rather than genetic control.

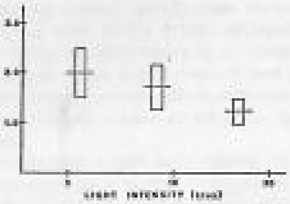

Fig. 11. Variety *lamourouxii*, length of mature assimilators developed on specimens maintained in sections B through D of the growth apparatus. The vertical bar represents two standard errors on either side of the mean, which is indicated by the horizontal line.

In analyzing the effect of light intensity on the ratio of number of assimilators developed per unit length of rhizome, a significant effect ($P < .001$) was found by analysis of variance for both var. *uvifera* (Fig. 12) and var. *lamourouxii* (Fig. 13). However, only the var. *lamourouxii* data from sections B through D showed a significant regression relationship ($P < .05$). The decrease in number of assimilators per unit of rhizome length with increases in light intensity for var. *lamourouxii* suggests an inhibitory effect and indicates its adaptation to low light intensity. Number of assimilators per unit of rhizome length was highest for var. *uvifera* at 8.3 and 21 klux, in this case suggesting adaptation to higher light intensities.

Both varieties demonstrated increased rhizoid development as light intensity increased. Variety *uvifera* had a significant regression relationship ($P < .05$) between increasing light intensity and an increase in its ratio of number of rhizoids per unit length of rhizome (Fig. 14). Length of rhizoids of var. *uvifera* did not vary

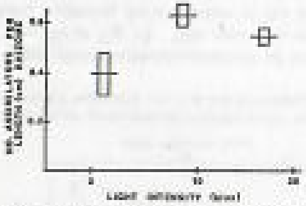

Fig. 12. Variety *uvifera*, number of assimilators per unit length of rhizome developed on specimens maintained in sections B through D of the growth apparatus. The vertical bar represents two standard errors on either side of the mean, which is indicated by the horizontal line.

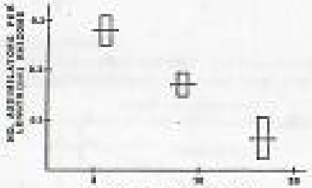

Fig. 13. Variety *lamourouxii*, number of assimilators per unit length of rhizome developed on specimens maintained in sections B through D of the growth apparatus. The vertical line represents two standard errors on either side of the mean, which is indicated by the horizontal line.

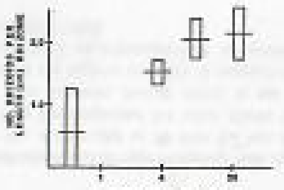

Fig. 14. Variety *uvifera*, number of rhizoids per unit length of rhizome developed on specimens maintained in sections A through D of the growth apparatus. The vertical bar represents two standard errors on either side of the mean, which is indicated by the horizontal line.

significantly between intensities. Variety *lamourouxii* (Fig. 15) did show increased rhizoid length with increasing light intensity when tested by linear regression analysis ($P < .01$), although its number of rhizoids per unit length of rhizome did not vary significantly. Because of focus and contrast problems the photographic rhizoid data for var. *lamourouxii* were not included. Although not quantified, rhizoids seemed to develop more diffuse branching at the higher intensities.

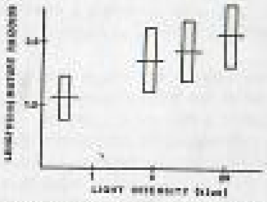

Fig. 15. Variety *lamourouxii*, length of mature rhizoids developed on specimens maintained in sections A through D of the growth apparatus. The vertical bar represents two standard errors on either side of the mean, which is indicated by the horizontal line.

Svedelius (1906), in separating the varieties *clavifera* and *uvifera* into distinct species, stated that var. *uvifera* has more highly developed rhizoids; however, I do not believe this difference is genetic. Light intensity and perhaps other environmental factors now seem a more likely explanation for varietal differences in rhizoid development.

Rhizome growth rate data showed no significant difference between varieties and no effect by light intensity over the range of intensities tested. Average rhizome growth rates for varieties *uvifera* and *lamourouxii* were 8.4 mm and 11.9 mm per day, respectively. Continued development of *C. racemosa* is dependent on rhizome growth. Perhaps other growth factors, including ramuli, assimilator and rhizoid development, are adjusted through some unknown equilibration process to ensure continued rhizome growth over a wide range of light intensities.

PRODUCTIVITY AND PIGMENTS

Net photosynthetic rate (Fig. 16) varied significantly between light intensities for both var. *uvifera* and var. *lamourouxii* f. *requienii* when tested by analysis of variance ($P < .001$). The net photosynthetic rate of var. *uvifera* dipped below the compensation point at approximately 1.6 klux whereas var. *lamourouxii* f. *requienii* required about half that intensity for compensation. At 3.7 klux var. *lamourouxii* f. *requienii* produced oxygen at three times the rate of var. *uvifera*. This gap narrowed with increasing light intensity and at 100 klux the rate for var. *uvifera* was slightly higher. Net photosynthesis increased approximately with the $\log_{10}$ of light intensity through 100 klux for var. *uvifera* and to 50 klux for var. *lamourouxii* f. *requienii*. Similar relationships have been reported for other plants (Blackman and Wilson, 1951; Marsh, 1970). The decrease, though small, in net photosynthesis of var. *lamourouxii* f. *requienii* at the highest intensity is suggestive of the photosynthetic inhibition which occurs with phytoplankton at intensities beyond their "light saturation" value (Strickland, 1960).

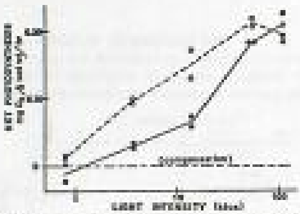

Fig. 16. Rates of net photosynthesis for var. *uvifera* (—●—) and var. *lamourouxii* f. *requienii* (--○--) at five light intensities. The point of zero oxygen exchange is the compensation intensity.

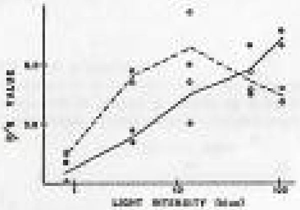

Fig. 17. P/R values (gross photosynthesis/respiration) for var. *uvifera* (—●—) and var. *lamourouxii* f. *requienii* (--○--) at five light intensities.

The P/R ratio (gross photosynthesis/respiration) of both varieties tested is plotted against light intensity in Fig. 17. This value rose steadily for var. *uvifera* and peaked at 100 klux, suggesting the adaptation of this form to high light intensities. The P/R value of var. *lamourouxii* f. *requienii* rose up to 13 klux and dropped at higher intensities, in this case suggesting adaptation to low light intensity.

Part of the reduction in the P/R value of var. *lamourouxii* f. *requienii* at 50 and 100 klux can be attributed to its higher respiration rates determined immediately after exposure to these intensities (Fig. 18). It is generally assumed that respiration remains stable under variable light conditions and that rapid increases in oxygen utilization at high light intensities result from photo-oxidation accompanied by pigment bleaching (Kinne, 1970). Regardless of whether it is increased respiration or photo-oxidation, the results again suggest the adaptation of var. *lamourouxii* f. *requienii* to low light intensity. The respiration data for var. *uvifera* were relatively stable at all light intensities, indicating that this form is not significantly affected by photo-oxidation.

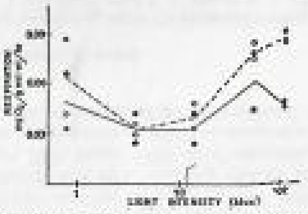

Fig. 18. Rates of respiration for var. *uvifera* (—●—) and var. *lamourouxii* f. *requienii* (--○--) measured after exposure to five light intensities.

Percent dry weight values of the varieties *uvifera* and *lamourouxii* were 4.8 and 5.5, respectively. The combined data gave a percent dry weight value of 5.2 for *C. racemosa*, similar to that reported for other *Caulerpa* species (Santos and Doty, 1971).

None of the pigment extracts tested at 750 millimicrons differed by more than 0.05 units from the optical density value of the 90% acetone standard. Most differed by a factor less than 0.01.

Chlorophyll a and carotenoid concentrations were found to be significantly higher ($P < .001$) for six specimens of var. *lamourouxii* than six specimens of var. *uvifera*, when analyzed by the t-test. Average chlorophyll a contents for var. *lamourouxii* and var. *uvifera* were 135.0 and 76.3 micrograms per gram wet weight, respectively. Corresponding carotenoid values were 4[illegible] and 24.1 MSPU per gram wet weight.

The results of 24 days of exposure to the light intensities within the growth apparatus on pigment contents are given in Fig. 19. The unusually high chlorophyll a value at 8.3 klux may have resulted from damage to the specimen or from an error in the weighing or pigment extraction technique. If this value is removed the decrease in chlorophyll a content shows a significant regression relationship with increasing light intensity ($P < .005$). The fact that chlorophyll a content varied with light intensity further indicates that characteristics which differ between *C. racemosa* varieties can result from environmental variation. The chlorophyll content of phytoplankton, both marine (Marshall, 1965) and freshwater (Sargent, 1940), and of higher terrestrial plants (Whittaker and Ga[illegible] 1965; Murray and

Nichols, 1966) has also been demonstrated to decrease in environments exposed to higher light intensities.

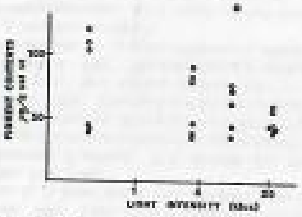

Fig. 19. Chlorophyll a (○) and carotenoid contents (●) of 12 specimens originally classified as var. *lamourouxii* after 24 days of exposure in sections A through D of the growth apparatus.

The larger chlorophyll a content of var. *lamourouxii* may be responsible for its higher photosynthetic rate at lower light intensities where photochemical processes are a limiting factor as opposed to enzymatic limitation at high light intensity (Steeman Nielson, 1962). The lower chlorophyll a content of var. *uvifera* could be considered an adaptation to high light intensity. Its low level would allow increased transmission of light which, if excessively absorbed and converted to heat, could damage the internal water balance (Daubenmire, 1959) as well as result in photo-oxidation.

Although carotenoid content was significantly greater for var. *lamourouxii* than var. *uvifera*, this factor did not vary with light intensity (Fig. 19) after 24 days of exposure in the growth apparatus. Apparently factors other than light intensity control carotenoid content.

CONCLUSIONS

Caulerpa racemosa demonstrated a remarkable ability to change its growth form under altered light conditions. Growth factors for both var. *uvifera* and var. *lamourouxii* were shown to be related to the $\log_{10}$ of light intensity. The observation that net photosynthesis similarly increased with the $\log_{10}$ of light intensity, for var. *uvifera* to 100 klux and var. *lamourouxii* to 30 klux, suggests a close relationship. However, this similarity is not necessarily causal.

Growth forms such as var. *lamourouxii* f. *requienii* and var. *peltata* appear to develop as adaptations to low light intensity. Both are flattened and present a greater surface area in proportion to size than forms occupying more exposed habitats. For this reason they should be able to use available light more effectively. The lower compensation point, larger gross P/R value and higher net photosynthetic rate at lower light intensities of var. *lamourouxii* f. *requienii* when compared to var. *uvifera* serve as evidence for such an adaptation. The observation that var. *uvifera* rose steadily in both rate of photosynthesis and P/R value as light intensity increased, whereas both of these factors dropped for var. *lamourouxii* f. *requienii* at the highest intensity, can also be considered an indication of its adaptation to high light intensity.

The increased chlorophyll a content of *C. racemosa* at low light intensity could be considered a means of adaptation for var. *lamourouxii* to occur in darkened habitats. An increased chlorophyll content would be able to absorb a greater percentage of the light available for photosynthesis. This would be especially beneficial at lower intensities where photosynthesis is limited by the rate of photochemical processes. The lower chlorophyll content of var. *uvifera* may also have an adaptive function. It allows increased transmission of light which, if absorbed and converted to heat, could damage the internal water balance or result in photo-oxidation.

Seasonality of *C. racemosa* within reef flat environments on Guam appears to be controlled by midday minus tides. The period of new development of *C. racemosa* corresponded with the cessation of midday minus tides and the period of maximum growth with their absence and low number. Minimum growth of *C. racemosa* occurred during months having the greatest number of midday minus tides.

The ability of *C. racemosa* to change growth form in altered light environments and the relationships of both morphologic and productivity factors to light intensity provide evidence for their environmental rather than genetic control. This information suggests that the classification of *C. racemosa* varieties as separate species, such as *C. peltata* (Gilbert, 1942; Taylor, 1960) and *C. lamourouxii* (Santos and Doty, 1971), is indeed in error and that these growth forms would more properly be referred to as ecophenes (ecological phenotypes) of the single species *C. racemosa*.

ACKNOWLEDGEMENTS

I thank Dr. Roy T. Tsuda for introducing me to the algae and making available much of the needed literature. I also thank Drs. J. A. Marsh, D. P. Cheney and P. J. Hoff for their suggestions and review of the manuscript. The staffs of the University of Guam Marine Laboratory and Government of Guam Water Pollution Laboratory generously allowed me use of their equipment and facilities. I am especially grateful to my wife, Marilyn, for her encouragement and invaluable assistance in the field and in the typing of this manuscript.

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George Balazs
National Marine Fisheries Service
P.O. Box 3830
Honolulu, Hawaii 96812

March 9, 1986

Dear George,

I have been looking through the algal identifications that have been made for your project and am happy with the results and organization. I have not, however, kept you informed about the credits for future sample identifications (you still have 38 vials paid for in advance).

I hope the last letter I sent you was satisfactory concerning the idenification of *Caulerpa*. All of my identifications are based on published descriptions and I can provide references to you for any species that may be challenged by another algologist (a common event).

Did I mention to you that I would be going to Guam, Ponape and Truk for the Western Society of Naturalists meetings in late June and early July 1986? The expectation of returning again to the tropical pacific thrills me. If there was ever an opportunity for employment in Hawaii or Guam, I would jump at the chance to return to the tropics.

My son, Allen, will be a freshman in college this fall and will probably go one or two years to SPU, after that only God knows where his future will lie. This is an exciting time for Sue and me since we will be "free" again. A new chapter may unfold in our lives. Excuse me for rambling a bit, but you with a young family and me completing mine just calls for some reflection.

Send me some more samples, anytime now.

Aloha,

Dennis

Sept 30, 1986

School of Natural & Mathematical Sciences

Seattle Pacific University
Seattle, Washington 98119
Phone: (206) 281-2140

Dear George,

Thank you for the article on poki. Our seaweed *Neoagardhiella gaudichaudii* is very much like limu ogo and very abundant in 20 ft of water near kelps and seagrass. maybe I should send to Tamashiro!

about the importing of snails with *Hypnea musciformis* from florida, This is anyone's guess. This disease sounds terrible. I hope you can help solve the problem. I assume *Hypnea* came from the miami area, but Lowell Funk (who received the shipment), was very reluctant to tell me anything about it. I guess miami, because that is where Clinton

Dawes is doing most of the Eucheuma work. I'll look into this more, but doubt if any better information will come forth.

Aloha,

Dennis

EPIZOITES

green from plastron at Kure = ?

check Russell I.D. letters in Algae file > ~1979 for skin/carapace scraping ID's.

check Russell letters for algae ID to add to some of stranding reports

-- Plastic cord ?

TM7

Acrochaetium gracile = Red ?

Sphacelaria furcigera = brown ?
S. novae-hollandiae

Green – ENteromorpha clathrata

See notes in JA Book + look at samples

See original 1983 Dennis ID list – Any "green" ?

7485
7565
7512
7495
7481

See true "green" ID.

Algae file

Diving

Many members have expressed interest in Scuba diving and we thought it would be a good idea to let everyone know what is happening and the general policy towards diving. MOP sstudents can participate in organized research diving. Besides being certified by an accepted scuba school, each diver must have a project proposal written and receive certification from the UH Diving Officer. MOP certification involves an extensive physical and a check-out dive to assure that each diver is competent and not a risk to themselves or others. The expensive university portion of the certification process is paid for by MOP. Without being redundant MOP dives are not recreational; there must be a serious proposal written before diving for or with MOP is considered.

We do not want to discourage members who wish to do recreational diving. UHH-MOP has had difficulty in the past with any sponsorship of diving classes and subsequent certification so we no longer sponsor such activities but we want interested members to feel free to come by the office and "talk diving".

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A Little Hilo History

This excerpt is from *Hilo Bay, A Chronological History* by Marion Kelly, Barry Nakamura, and Dorothy B. Barrere published by Bernice P. Bishop Museum Press, copyright 1981.

FOOD GATHERING IN THE BAY

Three species of edible seaweed, limu, found in Hilo Bay are known to have been gathered for food until modern times; perhaps others were also available for gathering in bygone days. The three are the limu-'ele'ele, limu-huluhuluwaena, and limu-'aki'aki (Albert Kai, pers. comm.). Limu-'ele'ele and limu-huluhuluwaena grow near shore in quiet water, in sand or mud or on small stones, and are easily gathered with bare hands; gathering of limu-'aki'aki, which has strong holdfasts, usually requires a strong swimmer with a sharp implement (Reed 1906:64). Women and children did most of the gathering of the limu along the shoreline, usually cleaning them at the site and often making a social affair of it all (Titcomb 1978:328). Until the 1930's limu-'aki'aki, called 'opihi limu locally because of its association with that mollusk, grew along the rocky shore that once existed between Reed's Bay (Kanakea) and Pier 1; the stretch has since been filled with sand and is a residential area (Albert Kai, pers. comm.)

Limpets ('opihi) have always been a source of marine food at Hilo Bay, and so have certain sea urchins (wana), shrimps ('opae), and crabs (kuhonu, 'a'ama', 'alamihi). Other mollusks and marine invertebrates were undoubtedly gathered for food and medicinal purposes in former days (Titcomb 1978:326); the invertebrates are identified and discussed in Titcomb (1978).

note: The article referred to often is *Native Use of Marine Invertebrates in Old Hawaii* by Margaret Titcomb published in Pacific Science, no.32, 1978

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How Complicated Was It?

The Library Aquarium

Do you remember the old parental speech "I know you really want the hamster dear but You have to feed it and clean the cage" or when you asked for a dog "Do you remember what happened to the hamster?" Well we would like to report a success in the aquatic pet field, the aquarium in the library. There used to be an aquarium on the second floor of the Campus Center but it's demise was due to lack of maintenance. WE ALWAYS NEED DEPENDABLE ENTHUSIASTIC PEOPLE FOR MAINTENANCE.

Enough pleas, the story behind the aquarium is interesting. Also for your trivia file, the largest aquaculture business is ornamental fish. The tank holds 85 gallons, the project has taken two years to finish, and the cost is estimated at $1500.00. The monies have come from Sea Grant, Student Government, and Marine Option. Arranging for the funding and materials took the most time, construction and installation only took about two months. The base was built by a professional aquarium builder (Stockly in Kona). A special note and thanks should go to Dennis Epperly, MOP Student Coordinator, who built the tank and put the final finish on the base. He also is responsible for the installation, acquiring the fish, feeding, and maintenance but he could use some help. He has said he is interested in holding a workshop on aquarium building, maintenance,and troubleshooting with the added resource of input from some local pet shop owners. If you are interested come by the office and let us know.

The tank has both biological (coral) and mechanical (ultraviolet sterilizer) filters and the fish are fed live guppies and some commercial flake pet food. MOP keeps a live guppy tank which is restocked usually by Dennis who says he catches them some place near Onekahakaha. It's bad fishing etiquette to give the exact location so we won't.

The marine life in the tank is diverse and here's the roll call: 1 eel, 1 humuhumu, 1 Christmas Wrasse, 1 trumpet fish, 1 reef triggerfish, 2 Philippine damselfish, 1 Triton's Trumpet shell (a carnivorous mollusk that paralyzes it's prey with a acid fluid), 5 cone shells (that are venomous), 1 slate pencil urchin, 3 sea cucumbers, 2 barber pole shrimp (if they haven't been eaten), 1 sea anemone, and some Nudibranchs or Spanish Dancers.

Keeping a salt water aquarium is very difficult (more so than fresh water types) but Marine Option thinks the effort is worthwhile and it's always nice when entering the library to see a few people looking and pointing.

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11-5-91 A5 Star-Bulletin

Study of seaweed growth off Maui gets more funds

By Lila Fujimoto
Maui Correspondent

WAILUKU — A University of Hawaii researcher expects to have a clearer view of what's causing the seaweed growth threatening coral reefs off West Maui with an additional $6,000 to study the problem.

The money from the university's Sea Grant College Program will allow further testing of waters off the island's resort area, said Steve Dollar, assistant researcher at the university's Hawaii Institute of Marine Biology.

"I'm pretty sure we can get a pretty good idea of what's going on," said Dollar, who will do more testing as part of a statewide water pollution study.

Divers have complained of heavy algae growth this year in the ocean off Kaanapali and Honokowai. But a Maui County Council committee's attempts to find funding to study the problem were blocked by questions of who has responsibility for coastal waters.

Dollar said the additional money will allow monitoring of West Maui waters to measure the effects of runoff on water quality. In recent weeks, with little runoff and algae anchored to reefs on the ocean bottom, the water has been clear, Dollar said.

But he said the algae growth is doing the worst damage to coral reefs that he has seen since sewage was pumped in Oahu's Kaneohe Bay.

"It's an environmental threat," Dollar said. "It's killing the coral reefs."

Dollar, who collected water samples off Kaanapali in 1989 after large amounts of algae bloomed, said he has ruled out golf courses and a sewer treatment plant as causes because the growth was absent last year.

He said he hopes the Legislature will support a request for money to do more studies. Rep. Rosalyn Baker, who represents West Maui, Molokai and Lanai, has said she will introduce a bill next session to fund the research.

9-13-91 HSB

Reed asks EPA to check growth of algae off Maui

By Lila Fujimoto
Maui Correspondent

WAILUKU — The growth of green seaweed in West Maui waters is prompting state Sen. Rick Reed to ask a federal agency to determine whether the state and Maui County are complying with the Clean Water Act.

In a letter this week to the director of the Environmental Protection Agency's water management division in San Francisco, Reed says the algae bloom may be a sign of water pollution from pesticides.

"Although state government and Department of Health officials are uncertain as to the cause of the algae growth, the best guess is that it is due to runoff from pesticide and fertilizer use on ag lands, or sewage, or a combination of the two," Reed says in the letter.

He asks the agency to conduct studies in the ocean off West Maui.

The algae, which first appeared three or four years ago, is so thick in some places that experts have said it could threaten coral reefs as well as discourage tourists who come to Maui to dive in the island's clear waters.

A Maui County Council committee last month asked Mayor Linda Crockett Lingle to find $14,880 for a study to document the algae growth and study its cause and solutions.

But Lingle said the state, not the county, is responsible for the waters off West Maui.

"I believe we should ask the state to be responsible for this survey," she wrote in a letter to Councilman Patrick Kawano, who heads the Council's Public Works Committee.

Fears raised by rapid algae growth off Maui

By Lila Fujimoto
Maui Correspondent

WAILUKU — For Ursela Keuper Bennett and others diving in the ocean off West Maui, this has been one of the cloudiest of years, with carpets of green seaweed obscuring the normally clear waters.

"This algae is the equivalent of throwing up in space — you get to see it all the time," said Keuper Bennett, a Toronto resident who has spent part of every summer for the past 15 years diving off West Maui.

"It was knee deep, then it became where we were just covered with it. I don't expect to see this. This is Maui."

Keuper Bennett and others Tuesday urged Maui County Council members to do something about the green slime that could harm the island's reefs as well as threaten an industry that attracts thousands of visitors each year.

Members of the Council's Public Works Committee agreed to ask Mayor Linda Crockett Lingle to find $14,880 for a study to document the algae growth and study its cause and solutions.

If the mayor does not respond in two weeks, the Council will try to find the money, said committee Chairman Patrick Kawano.

"We are trying to get at this problem," said Council Chairman Howard Kihune, who represents West Maui. "For the past three or four years, it's been very evident. But it's grown so much now."

At Tuesday's meeting, experts speculated that the island's numerous drainage canals could be discharging excessive nutrient-rich water into the ocean, spurring the algae bloom.

The state has found high concentrations of nitrogen in water samples taken at Mahinahina and Wahikuli in West Maui, said Eugene Akazawa of the state Department of Health's clean water branch.

State sampling also showed high amounts of ground water running into the ocean in some areas, Akazawa said. "I don't have the answer as far as tracing what the exact source is," he said.

After algae bloomed in waters off Kaanapali in 1989, the University of Hawaii's Institute of Marine Biology collected water samples to test for nutrients that might be caused by golf course runoff or a sewage treatment plant in the area, said Steven Dollar, an institute researcher.

But the samples showed no unusual nutrient levels, Dollar said, and algae did not bloom off Kaanapali last year.

Keuper Bennett said algae bloomed in waters off Honokowai both last year and in 1989. She called the area between Kaanapali and Kapalua "algae central."

After seeing her videotape of the growth at Honokowai, Dollar said the West Maui outbreak was worse than any he has seen elsewhere in the state.

"I don't think there's any question that this is not a natural phenomenon," he said. "It seems there's inevitably some cause due to activities on land."

William Magruder, a marine botanist for Bishop Museum, proposed a $14,880 five-week study to examine where the seaweed is showing up and why.

The solutions must come soon or Maui's tourist industry will be at risk, said Los Angeles businessman Monroe Hemmerdinger.

"People won't come here to dive in algae-filled slime," said Hemmerdinger, who said he has noticed the change in water quality in the past six months alone.

"What I have seen in the last 10 days has been alarming," he said. "The coral reef is dying. I have seen slime-filled waters and fish that look unhealthy and huge coral heads that are brown and dead."

Yes Humm

Pam

VOLUME 15 JUNE NUMBERS 1-2

ON THE GROWTH RATE OF THE RED ALGA, *HYPNEA MUSCIFORMIS*, IN THE CARIBBEAN SEA[1]

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ABSTRACT: It is shown that small plants of *Hypnea musciformis*, a red alga that is abundant in the Caribbean Sea and of economic value for its cell wall polysaccharides (κ-carrageenan), will double in weight in two days at depths of 150 to 900 mm in inshore waters around the islands of the Caribbean. It is proposed that this species can be grown economically in vats along shore through which water is pumped, and that half the crop can be harvested every few dags.

INTRODUCTION

An exceptionally rapid growth rate in *Hypnea musciformis* (Wulfen) Lamouroux, an abundant red alga in the West Indies, Caribbean Sea, Gulf of Mexico, and along the southeastern coast of the United States to Beaufort, North Carolina, was reported by Causey ~~et al.~~ (1946) in North Carolina. A similar growth rate for *Gracilaria foliifera* (C. Agardh) Borgesen was reported for North Carolina and the northern Florida Gulf coast by Kim and Humm (1964). *Hypnea musciformis* is a species of economic value for its cell wall polysaccharide, kappa-carrageenan, but wild stocks of this plant apparently are insufficient to meet the requirements of carrageenan producers. For this reason, cultivation of *Hypnea* may become profitable as soon as large-scale procedures have been developed.

Previous data on growth rates of *Hypnea musciformis* have been obtained in areas of relatively high fertility, Beaufort Harbor in North Carolina and Alligator Harbor in north Florida. In order to determine its growth rate in the relatively low nutrient waters of the Caribbean Sea, the writers made some observations in Tague Bay, north coast of St. Croix, American Virgin Islands, when there was a brief opportunity during August 1973, while they were in residence at the West Indies Laboratory of Fairleigh Dickinson University.

PROCEDURES

Hypnea musciformis was collected in shallow water attached to leaves of the seagrass, *Thalassia testudinum*. Fragments of plants weighing from 4 to 7 grams were tied to a line at 150 mm intervals by means of soft twine. The line was then attached to the pier of the West Indies Laboratory of Fairleigh Dickinson University. A float

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[1] Contribution #19, West Indies Laboratory, Fairleigh Dickinson University, St. Croix, U.S.V.I.

Carib. J. Sci., 15 (1-2). June 1975.

was attached to the line so that one plant was about 150 mm beneath the water surface and others at 150 mm intervals to a depth of 900 mm. A small weight was attached to the bottom of the line so that the line remained in a vertical position and each plant at a more or less constant depth.

Salinity of the water during the experiments was 34 to 35$^{0}/_{00}$, temperature 28-29° C. Water clarity was such that bottom was readily visible in depths of 2 M. Means of determining light penetration and nutrient content were not available.

The experimental plants were brought to the laboratory each day for a period of nine days and weighed, after free water had been shaken from them for a few seconds. They were re-tied to the line and placed back in the sea with only a few minutes exposure to the air.

RESULTS

Table 1 is a composite of two experiments involving 12 plants at six different depths on the vertical line over two nine-day periods. The total wet weight of each plant is given for each day of the two nine-day experiments. Figure 1 is a graphic record of the weight gains of the two best plants, those with the least grazing or fragmentation, or none.

It is evident from these preliminary data that the growth rate of *Hypnea* in island coastal waters in the Caribbean Sea is almost as high as in coastal waters of Florida and North Carolina.

DISCUSSION

Since no effort was made to avoid or compensate for reduction in rate of growth as a result of grazing and fragmentation, these data do not show the full growth potential of *Hypnea*. Among the 88 times that a plant was weighed other than the initial weight determination, weight loss (6) or no gain (4) was recorded ten times. On many other weight determinations the gain was so little as to indicate clearly that part of the plant had been lost.

Several species of algae-eating fish were common around the West Indies Laboratory pier where the experiment was conducted, and grazing appeared to be progressively greater with depth. In addition to the accidental loss of branches by fragmentation that occurred while the plants were on the lines, some occurred in connection with the daily weighing and re-tying.

The data indicate that in the clear waters of the Caribbean there is only a slight reduction in growth rate with each 150 mm increment of depth, and that the difference between 150 mm and 900 mm may be less than 10%.

Aside from losses by fragmentation and grazing, there is a progressive decrease in growth rate per unit weight of the plants as they increase in size. We attribute this to shading of the interior of the plant by the outer branches and to the less favorable environment of the interior part of the plant for exchange of materials with the surrounding water. In small plants these inhibiting conditions do not obtain.

In addition to the observed progressive decrease in growth rate per unit weight as the plants increase in size, there is a peculiarity in the growth increment per day in these experimental plants. The overall average weight increase per plant per day was two grams (1.998), including loss from grazing and fragmentation, with the distinct exception of the second 24-hour period.

During the second 24-hour period the average weight gain per plant was only 0.21 gram with a range among the 11 experimental plants of 0.1 to 0.6 gram (one plant was lost). This is about one-tenth the growth increment of the plants during all other 24-hour periods, including the first.

Organisms that are moved to a different environment that is equally favorable, or more so, normally go through a lag phase or period of adjustment before resuming a normal growth rate. These plants, however, exhibited the highest growth rate during the first 24-hours. If grazing and fragmentation could have been prevented, apparently the average plant would have increased in weight by more than 100% during this time. The second 24-hour period, however, was characteristic of a lag phase with weight gains of only one-tenth as much.

The only apparent environmental changes were light intensity and constancy and water movement around the plants. They were collected from the experimental area where they grew near the bottom attached to seagrass leaves in about 600 mm depth at mean sea level. They were partially shaded by the seagrass leaves and by other algae. The depth changed with tide from 150 to 300 mm per day, depending upon moon phase. In tying them to lines so that they were suspended in mid-water at a constant depth, they received more constant and continuous light. The up and down movement as a result of wave action on the float increased water movement around each plant in comparison to the natural environment.

It is tempting to speculate that the condition of the enzyme systems of these plants was such as to promote a rapid growth rate during the first 24-hours of the experiment that was partly at the expense of stored carbohydrates and not entirely a result of photosynthesis during the first day. During the second day, with an abnormally low carbohydrate reserve, a lag phase occurred. These experiments should be repeated under circumstances that permit an analysis of the plants for carbohydrate, protein, and the wet weight/dry weight ratio.

CONCLUSIONS

It appears to us that successful cultivation of *Hypnea* and other benthic algae in the Caribbean Sea can best be done in tanks or vats along shore into which sea water is pumped, perhaps by windmills, in such a manner as to keep the plants in motion so that clumping does not occur, exposure to light is even, and water motion permits rapid exchange of materials. With such a device, harvesting of half the plants in the tanks may be possible every other day during the summer months, or every few days, as doubling in weight should occur that often. It may be necessary, however, to break plants in two in order to prevent growth rate reduction with increase in size if the maximum growth rate is to be maintained in the tanks. The surface waters of the Caribbean Sea, at least along the island beaches, are sufficiently fertile to enable small *Hypnea musciformis* plants to double in weight in two days.

50%/day wgt increase

J. Phycol. **25,** 192–196 (1989)

NOTE

HYPNEA MUSCIFORMIS (RHODOPHYTA): ECOLOGICAL INFLUENCE ON GROWTH[1]

Rocilda P. F. Schenkman[2]

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ABSTRACT

Seasonal variations in growth, biomass, phenology, and phycocolloid content were studied in a population of Hypnea musciformis *(Wulfen in Jacquin) Lamouroux on the coast of the state of São Paulo.* H. musciformis *grows epiphytically on* Sargassaum cymosum *C. Agardh in a zone ca. 0.5 m wide at the lower spring tide. It also epiphytizes, but to a lesser extent,* Laurencia scoparia *J. Agardh and occasionally* Bryothamnion seaforthii *(Turner) Kützing and* Acanthophora spicifera *(Vahl) Børgesen. Seawater temperatures vary from 19° C in winter to 29° C in summer.*

Variations in H. musciformis *biomass (32–190 g dry weight·m^{-2}), were controlled by several factors, notably seawater temperature, diurnal lower spring tides on sunny days accompanied by calm seas soon followed by rough water, and by grazing. especially by Gammaridea (amphipods) and* Aplysia *(sea-hares). Fertile tetrasporophytes were present all year, their frequency varying from 22–99%. Cystocarpic plants were rare; male thalli were not found. Carrageenan yields varied from 48–66% of dry weight. Lowest values of carrageenan occurred when seawater temperatures were high (26–29° C) and biomass was low or, when biomass was high but frequency of fertile tetrasporophytes was highest. Mean field growth in plastic containers initiated from branches of* H. musciformis *(ca. 0.1 g) over a 28-day period was 0.48, 0.88, 1.30, and 1.53 g fresh weight, respectively, from summer to spring.*

Key index words: carrageenan; epiphytism; Hypnea; *phenology;* Sargassum; *seasonal growth; seasonal variation*

Hypnea spp. are well known sources of carrageenan and other compounds of pharmaceutical interest. *Hypnea* is eaten in the Philippines, used as a vermifuge in Indonesia and has been used in the U.S.A.'s phycocolloid industry since 1945–46 (Levring et al. 1969). More recently, carrageenan from *H. musciformis* was shown to have anti-inflammatory activity against rat hind-paw edema induced by commercial carrageenan (Solimabi 1980). Halogenated pyrrolopyrimidine analogs from *H. valentiae* are potent muscle relaxants, induce hypothermia when injected into mice, and may be the most potent and specific known adenosine kinase inhibitors (Davies et al. 1984). Low-molecular-weight peptides from *H. japonica* agglutinate animal and human erythrocytes, as well as mouse tumor cells (Hori et al. 1986). *H. musciformis* produces an extraordinarily high concentration of dimethylsulphide (79 μg·g dry wt^{-1} of algae), an important atmospheric compound emitted by the ocean (Whelan et al. 1982, Bates et al. 1987), and 1-methyl-1,3-cyclopentadiene [0.11 μg·g dry wt^{-1} of algae (Whelan et al. 1982)], a powerful Diels-Alder reagent that should interest phytochemists engaged in exploring biosyntheses, including those for pheromones.

About 20 years ago, Brazil began to export *H. musciformis* (Oliveira F. 1981). Nevertheless, *H. musciformis* was considered a lesser-known carrageenophyte (Woodward 1966). Several aspects of *Hypnea* spp., mostly in India and Hawaii, have since been studied and reviewed (Mshigeni 1978).

In Brazil *H. musciformis* is the most abundant carrageenophyte, occurring from the states of Maranhão (5° lat. S) to Rio Grande do Sul (30° lat. S). Despite economic potential, there are no studies in Brazil concerning this genus aside from species descriptions in floristic surveys. The work presented here includes phenological observations, analysis of biomass, and growth experiments in the field on the coast of the state of São Paulo.

The study site was at the north end of Praia Grande, Municipality of Ubatuba, São Paulo, Brazil (Fig. 1). The shore at mean high-tide consists of gneiss boulders, which support an algal and animal community typical of Brazil's southeast coast. The boulders are smaller seawards and are moderately exposed to lateral wave impacts. Seawater temperature was measured with a mercury thermometer at each visit and varied from 19° C (July 1979) to 29° C (February 1980). Global solar radiation varied from 230–239 (April–July 1979) to 436 cal·cm^{-2}·min^{-1} (January 1979). The solar radiation was measured with a bimetallic actinograph, and tidal levels were measured with a tide gauge, both fixed 6 km from the study site. Salinity varies little, ranging between 34.0–35.6‰ (Tararam and Wakabara 1981). Nutrients in a nearby area vary more within a month than seasonally: nitrate from 0.005–0.102 μg-at·L^{-1} during winter and 0.011–0.126 μg-at·L^{-1} in the summer; phosphate from 0.027–1.010 μg-at·L^{-1} in the winter and 0.031–1.103 μg-at·L^{-1} in the summer (Teixeira 1979).

H. musciformis occurred as an epiphyte on *Sargassum cymosum* C. Ag., less commonly on *Laurencia scoparia* J. Ag., and sporadically on *Bryothamnion seaforthii* (Tur.) Kütz and *Acanthophora spicifera*

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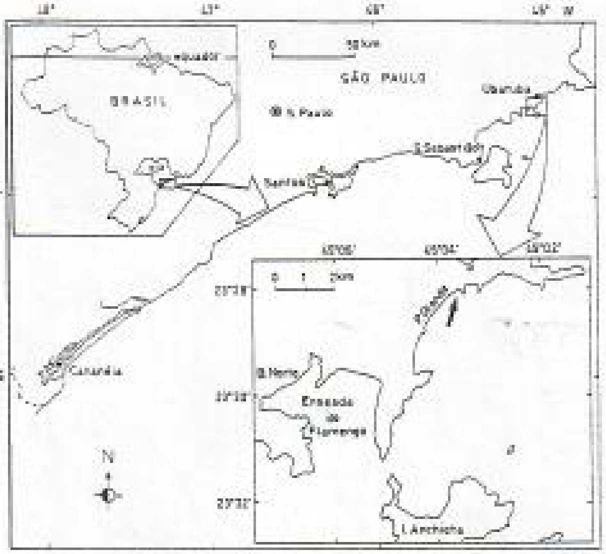

FIG. 1. Location of study area (arrows), coast of state of São Paulo.

(Vahl) Børg. The plants were 4–8 cm high, irregularly branched, sometimes with short spinescent branchlets. Hamate organs occurred at apices of well-developed branches enabling attachment to the host plants; rhizoidal cells may project from the surface of the hamate organ where it is in contact with the host.

The size of the sampled area, minimum number of samples, and sampling methods for biomass studies were chosen according to Goldsmith and Harrison (1976). Random samplings of areas of occurrence of *H. musciformis* (restricted-random) were initially done monthly and later bimonthly from September 1978 to July 1980 using a 20 cm square (area = 400 cm^2). Ten samples were collected at each visit for biomass studies. In the laboratory, 10 plants of *H. musciformis* from each sample, totaling 100 specimens per month, were examined for reproductive structures. *Sargassum* and *Hypnea* were dried at 60° C to constant weight (percentage of water in thallus = 84.6 ± 0.3%, n = 10).

Phycocolloid yields were calculated from dry material previously washed in fresh water, then reduced to powder in a blender (Santos and Doty 1975) except that the pH was adjusted to 7.4–8.0 instead of 8.5–9.0 because a more alkaline pH yields a darker product, presumably by a Maillard reaction. The mixture was heated at 93° C for 5 h. Acetone was used instead of isopropanol to precipitate the colloid (acetone, cheaper locally, produces the same yields).

Field growth experiments conducted at the Centro de Biologia Marinha, Universidade de São Paulo, Municipality of São Sebastião, ca. 60 km south of the sampling area. Fifteen branches of *H. musciformis* collected in Praia Grande, 0.12–0.16 g fresh weight each, were place individually in perforated polystyrene containers (8.5 cm high, 6.0 cm diam) with perforated caps. Such containers were chosen after testing various types. They have the advantage that thalli could be easily removed, weighed, and replaced without fragmentation, which permitted continuous work with the same material. Also, in these containers, thalli were not lost by water currents, waves, or predation by herbivores. Compared with mesh-bag receptacles, the polystyrene containers had the disadvantage of restricted space and low flow that may have led, as thalli grew, to nutrient-limiting conditions. The containers were tied to a cord at zero tide level ca. 2 m from the bottom and 100 m from water line. Branches and containers were brushed clean of a fouling layer, the branches weighed, then put back in the sea once a week, for 4 weeks. These experiments were done in each season.

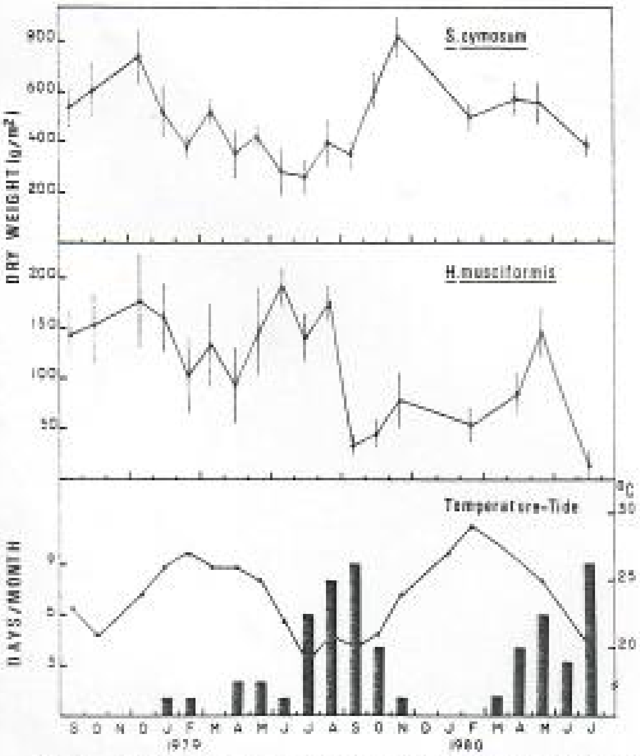

FIG. 2. Seasonal fluctuation of biomass (vertical lines = 95% confidence interval), seawater temperature, and number of days of occurrence of diurnal low (<0.0) spring tide (bars) between 0800 and 1700.

H. musciformis was present throughout the year, its biomass varying from 32–190 $g \cdot m^{-2}$ dry weight (Fig. 2). Values were lower when the temperature reached the 26–29° C maxima; higher values were found at the coolest temperatures (19–25° C). Two dramatic decreases in biomass occurred during September 1979 and July 1980 and were related to the occurrence of diurnal lower spring tides (Fig. 2). Occurrence of *H. musciformis* throughout the period of observation (23 months) parallels the situation in India (Rama Rao 1977) where it is found all year, but not so in Massachusetts (Conover 1958) or Florida (Benz et al. 1979) where *H. musciformis* is conspicuously seasonal in occurrence.

This study suggests that fluctuation of *Hypnea* biomass is influenced by several factors, notably seawater temperature, lower diurnal spring tides, availability of substrate (host plants) and possibly predation by herbivores.

Although temperature has an important influence on *H. musciformis* biomass, it does not counter the effects of low tides. Exposure to air during lower spring tides on sunny days and calm sea (even in winter) results in thallus bleaching and attendant loss of biomass especially if followed by rough-water periods, which are frequent in winter and affect especially plants with soft thalli such as *H. musciformis*. Lawson (1957) and Mshigeni (1976a, 1977) showed for Ghana and Hawaii, respectively, that the abun-

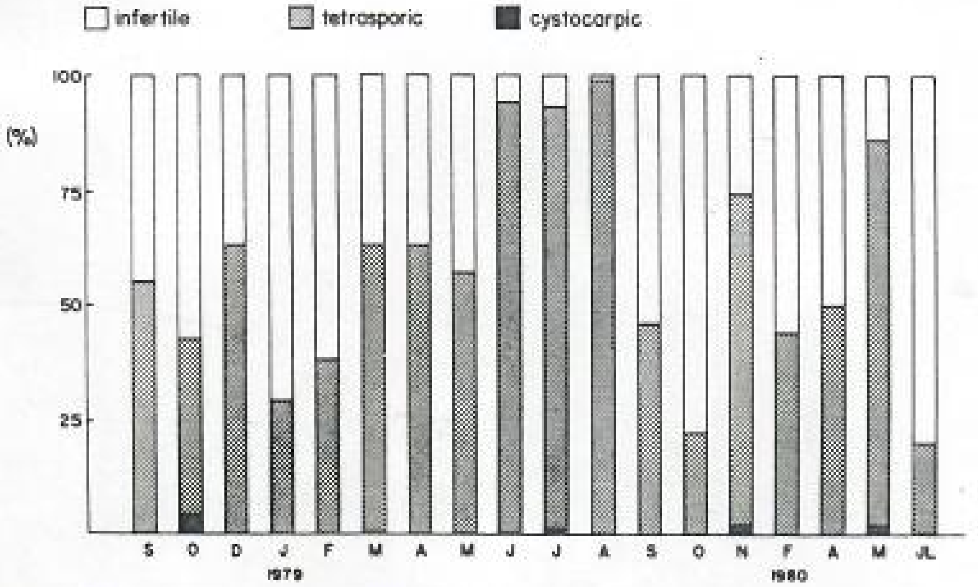

FIG. 3. Percentage of fertile tetrasporophytes, fertile female gametophytes, and infertile *H. musciformis* plants.

dance of *Hypnea* spp. during the year is correlated with seasonal changes in the time of low spring tides.

Biomass of *Sargassum cymosum*, the most common substrate, (Fig. 2) varied from 267–802 g·m^{-2} dry weight, maximum values occurring in late spring (November and December), minimum values during winter (July). *S. cymosum* was the dominant species in this area. It is abundant throughout the year but loses branches from the end of summer until midwinter.

Laurencia scoparia was also present throughout the year with a frequency varying from 20–100% of the samples. It was the alternate host species for *H. musciformis* in late autumn and winter when *Sargassum* biomass was low. *Bryothamnion seaforthii* and *Acanthophora spicifera*, also rare alternate hosts for *H. musciformis*, had frequencies varying from 0–40% of the samples during the year.

Since *H. musciformis* is intimately associated with *S. cymosum* as substrate, factors affecting *Sargassum* biomass affect *Hypnea*. Throughout the observation period, correlations emerged between *S. cymosum* and *H. musciformis* biomass (September 1978–April 1979, $r = 0.91$, $P = 0.001$; September 1979–April 1980, $r = 0.69$, $P = 0.1$). Absence of correlation between May–August 1979 may be due to the faster growth of *H. musciformis* compared with *S. cymosum* and the use of *Laurencia scoparia* as an alternative host species.

The nudibranch mollusk *Aplysia* (sea-hare) and Gammaridea (amphipods) were observed but not quantified in this work. Tararam and Wakabara (1981) showed that the frequency of gammarid herbivores in this community increased from spring to summer, then declined. Winkler and Dawson (1963) showed that *H. valentiae* is the most common seaweed found in the crop and pellets of *Aplysia californica*. Perhaps amphipods and *Aplysia* are significant grazers in this *H. musciformis* population.

Fertile tetrasporophytes occurred throughout the year, varying from 20–99% of the *H. musciformis* population (Fig. 3). The highest frequencies of tetrasporophytes with many stichidia and spores were observed during the months with high biomass (June–August 1979; May 1980). Low frequencies of fertile tetrasporophytes were in summer and in months following the dramatic decrease of biomass (October 1979, July 1980); in this case only a few, old stichidia in the oldest portion of the thalli were observed. Cystocarpic specimens were rare, found only four times (October 1978, July and November 1979, May 1980) during 23 months of regular observation; the relative frequency was always <4%. Plants with spermatangia were not found.

Literature references to male plants of *Hypnea* spp. are rare. Only Tanaka (1941), studying 14 species of *Hypnea* from Japan, described male structures for three species, but not for *H. musciformis*. Rarity of male plants of *Hypnea* spp. in nature has been documented (Rama Rao 1970, 1977, Mshigeni 1976b). At the São Paulo coast, as in India, no male plants were found: fertile tetrasporophytes predominated.

Why are fertile gametophytes rare despite the high production of tetraspores in several genera of Rhodophyta? Is it due to the apomeiotic division of the sporangia or to a greater ability of diploid plants to propagate vegetatively compared to the haploid phase (Dixon 1973, Searles 1980), or neither? I found (Schenkman 1986) that tetraspores of *H. musciformis* gave rise in vitro to male and female fertile gametophytes; meiosis seemed normal in the tetrasporangia, which may mean that apomeiosis probably does not explain the population studied. In the present

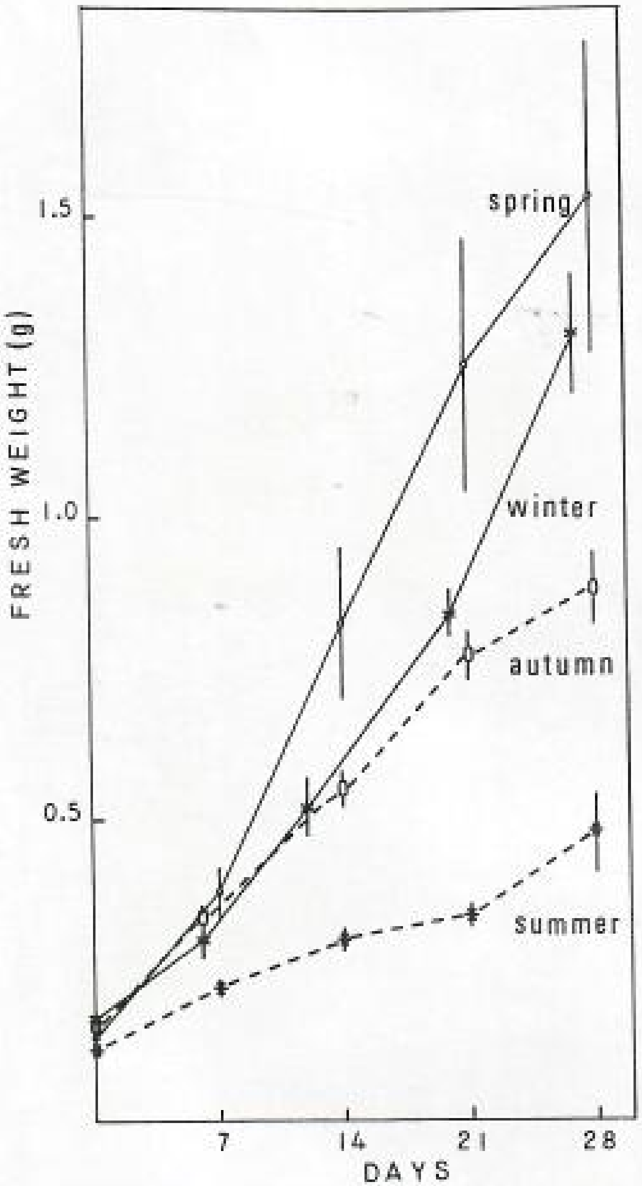

FIG. 4. Seasonal experimental field-growth of *H. musciformis* (vertical lines = 95% confidence interval).

study, the *H. musciformis* population appears to be maintained mainly by vegetative reproduction due to its epiphytic habit, aided by possession of the hamate attachment organs. Regeneration of *H. musciformis* plants, probably from rhizoidal cells formed from the interior of hamate organs, was observed in the laboratory on *S. cymosum* branches or, less frequently, in culture vessels where *H. musciformis* had attached by itself. I did not compare the ability of diploid plants (tetrasporophytes) and haploid plants (gametophytes) to propagate vegetatively.

Mean growth from explants of *H. musciformis* weighing 0.12–0.16 g fresh weight each over a 28-day period was 0.48 ± 0.07 g, 0.88 ± 0.06 g, 1.30 ± 0.10 g, and 1.53 ± 0.26 g fresh weight in the summer, autumn, winter, and spring, respectively (Fig. 4). The growth of *H. musciformis* was better in winter and spring when the temperature was <25° C. Changes in thallus morphology included a color change from reddish to yellowish, and most lateral branches elongated during experiments, with loss of the spinescent aspect of the thallus—a phenotypic plasticity to be considered in taxonomic studies.

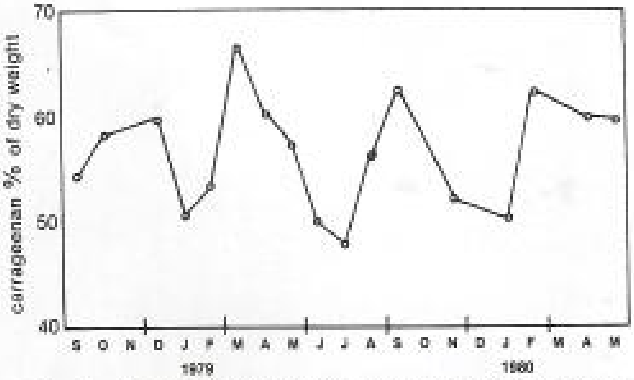

FIG. 5. Seasonal fluctuation in carrageenan yield of *H. musciformis.*

Phycocolloid content varied from 48–66% of the dry weight (Fig. 5) with maximum yields in autumn (March–April) and spring (September–October). Minimum values were in summer (January) when *H. musciformis* biomass declines and also in winter (July) where, despite high biomass, the percentage of fertile plants is highest. Reported phycocolloid content in *H. musciformis* varies from 16–48% dry weight (Humm and Williams 1948, Rama Rao 1970, Haines 1975, Guist et al. 1982, Friedlander and Zelikovitch 1984). This variation is probably attributable not only to strain differences but different extraction procedures.

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2/17/92

northstar-at-tahoe®

GEORGE – ENCLOSED IS MY ALOHA REPORT FOR THIS SEASON. HOPE THE INFO. IS OF SOME HELP.

INCIDENTALLY, YOU KNOW ABOUT THE ALGAE PROBLEM OVER HERE AND THE STUDY BEING ORGANIZED TO DETERMINE THE CAUSE(S). I HAVE PHOTOS OF THE HEDGE-ROWS OF ALGAE ON THE BEACH HERE IN JAN. '91 + '92. IT SEEMS TO OCCUR IN OUR NEARSHORE TIDEPOOLS FOLLOWING OR DURING HIGH TIDE HEAVY SURF PERIODS. AT LEAST 90% (+) IS THE RED → ALGA HYPNEA CERVICORNIS ?? (HUNA IN HAWAIIAN) – MAYBE YOU CAN PASS THIS ALONG TO THOSE CONCERNED IN THE STUDY.

over

I HAVEN'T FOREGOTTEN
THE "TURTLE WATCHIN'"
LETTER-TO-THE-EDITOR OR
ARTICLE. WHEN COMPLETED
I WILL SEND YOU A
DRAFT FIRST & GO FROM
THERE. I AM MORE
LIKE YA DESERT TORTOISE
THAN YA SWIFT(?) SEA
TURTLE sometimes,
especially WHEN
"POLYNESIAN PARALYSIS"
SETS IN. TO WHIT:

ME KA PUMEHANA —

ALOHA O'E —

BUD HEACOX

B. Heacox
227 San Miguel Way
Sacramento, CA 95819-1931

STOMACH CONTENT ANALYSIS OF STRANDED JUVENILE AND ADULT GREEN TURTLES IN BROWARD AND PALM BEACH COUNTIES, FLORIDA

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Jeanne L. Wershoven
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Strandings of sea turtles occur on the beaches of Broward and Palm Beach counties throughout the year. Strandings are reported to Florida DNR, county environmental agencies, or to a telephone hotline monitored by the authors. SCUBA divers also report sightings of dead sea turtles on the reefs.

Broward and Palm Beach counties constitute the northern two-thirds of the highly developed Miami-Fort Lauderdale-West Palm conurbation of southeastern Florida. The coasts are highly developed, with most of the beachfront property in private ownership. The offshore areas of these counties consist of relict limestone reefs which support large recreational and commercial fishing and diving activities.

METHODS

Since 1986 a stranding hotline, first sponsored by Broward County Audubon Society, has been effective in gathering reports from Broward County. In 1990, this effort was expanded to include Palm Beach County. Stranding reports are investigated by volunteers, with necropsies performed when feasible, either in the field on the adults, or at our home on the juveniles. Live animals are taken to rehabilitation facilities.

Necropsies are performed in accordance with techniques outlined in Wolke and George (1981). Gross examination of organs are undertaken. The gastro-intestinal tract is thoroughly examined for identifiable food particles and for evidence of plastics or other foreign material. Due to the nature of injuries to most animals, many strandings are not intact. Food matter is often obtained from the esophagus if the lower organs are missing.

Stomach samples are preserved in 10% sea water-buffered formalin. Algae samples have been identified by Dennis Russell of Seattle Pacific University and Steve Blair of Dade County (Florida) Department of Environmental Resource Management. Specimens of sea grasses have been examined by Steve Blair and Paul Mikkelsen of Palm Beach County Department of Environmental Resource Management.

RESULTS

Of the over 100 strandings reported to us since 1986, 66 have been green turtles, 56 juveniles and 10 adults. A curved carapace measurement of 80 cm separates the juveniles from the adult category. The 10 adults were represented by 6 males and 4 females, and were reported in 1989 and 1990. Four of the 6 adults stranded in 1990 were reported in Palm Beach County. In 1990, a record 19 juvenile strandings were reported, 4 of which were from Palm Beach county. These figures do not represent total strandings, only those reported to the authors.

Juvenile strandings are reported year-round, with an increase in the spring and summer months. This may be indicative of more efficient reporting due to the presence of nest survey patrols. The decrease during the winter months also corresponds to the lowered number of captures made by the authors during SCUBA surveys in the Fort Lauderdale area for the same months (Wershoven and Wershoven, 1989). Adults have been reported only during the nesting season.

The curved carapace lengths of the juveniles ranged from approximately 8.0 cm to 72.4 cm, with a mean of 45.1 cm and a median of 43.0 cm. Stranded adult greens ranged in size from 98.5 cm to 113.5 cm, with a mean of 104.8 cm and a median of 105.45 cm.

Boat propellers were responsible for the most injuries to stranded green turtles, with 34 of 56 juveniles and 4 of 10 adults exhibiting severe cuts usually on the rear of the carapace. Entanglement in fishing line and other human artifacts has been the apparent cause of death for 8 juveniles and 2 adults. Evidence of possible shark attacks has been observed on 1 adult and 4 juveniles. One juvenile stranded just north of a fishing pier with a fishing hook in its mouth, its skull fractured, and its throat slit. The remaining strandings exhibited no apparent cause of death. No evidence of papilloma was observed on any stranded turtles.

Necropsies were performed on 42 of the 56 juvenile green turtles. Three of these turtles yielded no stomach or esophageal contents. Most of the specimens contained a mix of algae and/or sea grasses, only 11 stomachs contained a single species of food material. No plastics or foreign matter, other than fishing hooks and line, were found in the gastro-intestinal tracts of any of the necropsied juvenile turtles.

The most common food items by frequency of occurrence were the Rhodophytic algae of the family Gelidiaceae. The contents of 12 specimens were wholly comprised of these algae. Sea grasses were the second most abundant food element, completely constituting 4 samples, and occurring with Gelidiaceae and Gracilaria in 12 others. Of the remaining 15 genera of algae represented in the specimens, the red algae Gracilaria was most common.

The ingested algae were found to be representative of the offshore floral communities proximate to the location of the stranding. Although the offshore area is devoid of significant sea grass beds, floating rafts of grasses are blown in from other areas during periods of high winds. One juvenile green stranded on the shore of Lake Worth had been feeding primarily on sea grasses from the lake, Halodule wrightii and Halophila johnsonii.

Necropsies were performed on 8 of the 10 stranded adult greens. Eggs were present in the oviducts of 2 of the 3 females examined. Examination of the gastro-intestinal tract yielded only small amounts of food matter; two of the animals had empty guts. Four turtles had been feeding upon Sargassum natans; two exclusively, two had augmented their diet with hydroids and red algae of Gelidium and Gracilaria species. One adult presented a sample of Halimeda with unidentified sponge material; Hypnea was found with hydroid stems in the remaining specimen. No plastics or other foreign matter was found.

CONCLUSION

Strandings of green turtles, especially juveniles, occur with some frequency along the coasts of Broward and Palm Beach counties. Juvenile green turtles are known to utilize the offshore areas of Broward County as a developmental habitat (Wershoven and Wershoven, 1989), and have been frequently observed by divers along nearshore hardbottom areas of Palm Beach County. The paucity of food matter in the stomachs of stranded adult green turtles and the absence of sightings of adults in the offshore area during non-nesting months indicate that these turtles may be utilizing a different habitat and are migrating to South Florida beaches during the nesting season.

The presence of Rhodophytic algae within the gastrointestinal tracts of stranded juvenile green turtles, sightings of turtles feeding upon these plants and the abundance of these food items along the hardbottom areas of Broward and Palm Beach County underscore the importance of this habitat to an endangered species. At the present time there are several threats to the integrity of these nearshore reef areas: massive freshwater discharges during periods of heavy rainfall may result in the proliferation of algal species disruptive to growth and maintenance of food items preferred by green turtles. Beach renourishment projects result in frequent overloading of the nearshore environment with silts and clays not only during projects, but for years afterward. The disruptive activities of recreational boaters, while not a direct threat to algal growth, are clearly a cause of mortality. Similarly, commercial fishing activities that result in sea turtle deaths need to be regulated or modified. Efforts to control and monitor these threats should be implemented.

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A cytochemical study of the wound-healing protein in *Bryopsis hypnoides*

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Abstract

A cytochemical study of the wound-healing material in the alga *Bryopsis hypnoides* was conducted at both the light and electron microscope levels. The results of the tests indicate that the wound-healing material is a proteinaceous substance which is not complexed with a polysaccharide. That is, it does not seem to be a glycoprotein or a mucopolysaccharide. In addition the structures which were interpreted earlier as representing stages in the development of the wound-healing protein (Burr and West, 1971) show similarity in their reactivity in the various tests. This lends further support to the proposal that they are ontogenetically related.

Introduction

The marine coenocytic alga *Bryopsis hypnoides* Lamouroux is a simple, siphonous, pinnately branched chlorophyte. It is devoid of crosswalls except at the bases of older branches and gametangia. Being constructed of only a relatively small number of large multinucleate units, it might be surmized that damage to the plant would have serious consequences. However, the alga has evolved a mechanism for dealing with such crises. If the thallus is injured either by incision or puncture, there is a rapid formation of a hyaline septum at the site of the wound (Haberlandt, 1929; Burr and West, 1971). This septum, or wound wall, is produced from large masses of flocculent material which are released from the vacuole upon injury. The material accumulates at the opening and quickly condenses to form a sealing layer. At the ultrastructural level this transformation is seen as a conversion of a highly disperse, granular-fibrillar substance into a dense, compacted matrix.

Our earlier paper (Burr and West, 1971) traced the complex developmental pathway by which this wound-healing material is synthesized and examined the formation of the wound wall. An abbreviated summary is provided in Figures 1 to 6 inclusive. The first developmental stage is represented by vesicles of rough endoplasmic reticulum (Figure 1) which increase in size and accumulate a dense material (Figure 2). A large, highly subdivided granule (the protein body) is formed within each vesicle from this material (Figure 3). The protein body is released into the vacuole where it loses its bounding membrane and begins to break down (Figure 4), first into thick filaments which have a microtubule-like core and a flocculent coat (Figure 5) and finally into a fine granular-fibrillar material (Figure 6). It is this latter material which condenses to form the wound wall.

Previous studies on the subject of wound-healing in plants have been largely limited to higher plants. In addition most of the work has been concerned with later stages in the recovery process such as dedifferentiation and regeneration. Some documentation already exists for the cytological events leading to the restoration of vascular connections (Eschrich, 1953; Sinnott and Bloch, 1945) and to the formation of protective barriers (Bloch, 1941, 1952; Lipetz, 1970) but the physiological bases for these changes remain poorly understood (Lipetz, 1970). Our studies in *Bryopsis* have focused on the primary cellular events following wounding rather than on the longer-term consequences. Regeneration has been examined in some detail by Haberlandt (1929).

The participation of a 'clotting' substance in wound-healing as seen in *Bryopsis* may be an isolated phenomenon limited to *Bryopsis* and a few other coenocytic algae. On the other hand, as we mentioned earlier (Burr and West, 1971), the process bears some interesting parallels to the effects of injury seen in sieve tubes. The typical electron micrograph of a sieve plate shows that the sieve pores are filled with P-protein and that the sides of the pores are covered with callose (*e.g.* Cronshaw and Anderson, 1969). Most investigators tend to regard the presence of both P-protein and callose as artifacts resulting from injury to the tissue during specimen preparation (Evert and Derr, 1964; Evert *et al.*, 1971; Cronshaw and Anderson, 1969; Ie, 1966; Milburn, 1971, 1972). Callose is found in cells other than sieve elements and in some of these its deposition appears to be associated only with injury (Currier and Strugger, 1956; Nims *et al.*, 1967; Ainst and Williams, 1971). Whether or not P-protein is also involved in the sealing mechanism in higher plants is still unclear (Milburn, 1971, 1972; Walker, 1972). However, if P-protein does function in this capacity then the system would constitute a strong analogy to the wound-healing process seen in *Bryopsis*. Although no callose seems to be present in *Bryopsis*, the formation of a new polysaccharide wall just internal to the wound septum shortly after 'clotting' (Burr and West, 1971) might be considered the equivalent of callose deposition. In appearance and staining this wall does not differ from the lateral walls of the thallus (Burr and West, 1971) and is presumably also composed of xylan (Iriki *et al.*, 1960; Frei and Preston, 1968).

Figure 1 (*top, left*) The first stage in the development of the protein bodies are small, rough endoplasmic reticulum vesicles (ERV). Double arrow points to an unswollen ER cisterna. The ribosomes on the membranes serve as a convenient marker for the early stages. The material in Figures 1 to 6 was fixed in glutaraldehyde-osmium, and poststained by uranyl acetate and lead citrate. × 42,500.

Figure 2 (*top, right*) ERV at a later stage showing the accumulation of a homogeneous, slightly electron dense substance. × 30,600.

Figure 3 (*centre, left*) Mature protein body. Note the presence of ribosomes on the limiting membrane. × 18,800.

Figure 4 (*centre, right*) Protein body in the vacuole (actually a vacuolar embayment) undergoing breakdown. × 30,600.

Figure 5 (*bottom, left*) Filaments resulting from the degradation of the stage seen in Figure 4. The filaments consists of a microtubule-like core surrounded by a granular-fibrillar material. These are located in the central vacuole. × 18,800.

Figure 6 (*bottom, right*) The granular-fibrillar substance produced from the degeneration of the filaments. × 36,000.

Because the wound-healing material in *Bryopsis* originates within cisternae of rough endoplasmic reticulum, it seemed plausible to surmize that it was largely, or entirely, proteinaceous. This was borne out by our initial tests with aniline blue black and Coomassie brilliant blue (Burr and West, 1971). Since then we have conducted additional studies in order to characterize the wound-healing material more precisely. In particular, it was desirable to learn whether the material was a protein-polysaccharide complex, such as a glycoprotein or a mucopolysaccharide. Also, some proof of compositional similarity between the different stages of development would be expected if indeed they were ontogenetically related. To this end various cytochemical tests were conducted at both the light and electron microscope levels.

Material and methods

The material used in these studies was fixed in glutaraldehyde-osmium and embedded in Epon (Burr and West, 1970). In addition, some of the plants were fixed only in glutaraldehyde. Semi-thin sections for light microscopy were cut at 0·25–0·5 μm with glass knives. Whole pieces of the thallus, either fresh or fixed in glutaraldehyde, and cleaned wall preparations were also used in the light microscope tests. The cleaned walls were prepared by placing the thalli in distilled water and homogenizing for a few seconds in a Waring blender then rinsing several times with distilled water. The combination of breakage and osmotic shock proved sufficient to produce walls that were free of cytoplasm. The wall preparations were especially beneficial in the study of the basal septa.

In all the ultracytochemical tests the thin sections of glutaraldehyde-osmium or glutaraldehyde-only fixed material were floated successively on each of the solutions, transfer being accomplished by a platinum or chromium loop. At the end of each procedure the sections were picked up on copper mesh grids, which had generally been precoated with Formvar.

Figure 7 (*top*) PA-AgP test. Only the starch grains react strongly following incubation in aqueous AgP (pH 6·4). The less electron dense ring around each starch grain is an artifact produced by the shrinkage of the grain during dehydration. The small clear spots in the chloroplast are lipid globules (L) which have been dissolved by the treatment. Because the section has not been poststained, all components lack the usual contrast. Note that none of the stages of development of the protein material show any reactivity — endoplasmic reticulum vesicles (ERV), protein bodies (PB) and filaments (F). There is some contamination of scattered silver grains on the section. This is non-covalently-bound silver which was left by insufficient rinsing following staining. The material in Figures 7 to 10 was fixed in glutaraldehyde-osmium. B — endogenous bacteria. N — nucleus. S — starch. V — vacuole. W — cell wall. × 4,500.

Figure 8 (*bottom, left*) PA-AgP test, pH 9·2. No detectable staining of ERV or F. The filaments, it should be mentioned, are actually in an evagination of the vacuole, and not in the cytoplasmic matrix. × 22,500.

Figure 9 (*bottom, centre*) PA-AgP test, pH 9·2. A degenerating protein body in the vacuole. No staining by the silver proteinate. × 16,600.

Figure 10 (*bottom, right*) PA-AgP test, pH 9·2. The cell wall and the cuticle show slightly better reactivity at this pH than at pH 6·4. The fine silver grains are evenly distributed throughout the wall. C — cuticle. × 41,300.

Polysaccharides

For the light microscope localization of polysaccharides, semi-thin sections were stained by the periodic acid-Schiff's reagent procedure outlined by Jensen (1962). Controls were also run in which the periodic acid step was omitted. Acid and sulphated polysaccharides were reacted for by the methods of Parker and Diboll (1966) using pieces of fresh or glutaraldehyde-only fixed thalli and cleaned wall preparations. Acid polysaccharides stain with Alcian yellow at pH 2·5 and sulphated polysaccharides with Alcian blue at pH 0·5. The suggestions of Scott and Mowry (1970) were incorporated into our procedures.

Two tests for the ultrastructural detection of carbohydrates were used, the periodic acid-silver proteinate test (PA-AgP) (Thiéry, 1967) and the periodic acid-silver methenamine procedure (PA-AgM) (Rambourg, 1967; Thiéry, 1967). Both are modifications of the periodic acid-Schiff's test (PAS) used in light microscopy (Rambourg, 1967); Thiéry, 1967). The PAS test is believed to localize carbohydrates with 1,2-glycol groups. In the Schiff test the diol is first oxidized to a dialdehyde with periodic acid, then reacted with the Schiff reagent to yield the highly coloured fuchsin. In the ultrastructural tests, the Schiff reagents are silver solutions, which deposit metallic silver at the reactive sites. In the silver proteinate test the dialdehyde is first complexed to a thiol-containing compound, either thiosemicarbizide (TSC) or thiocarbohydrazide (TCH) and then reacted with the silver proteinate (Thiéry, 1967). According to Jewell and Saxton (1970), silver proteinate is a mixture of silver nitrate and peptones. Incubation in TSC and TCH was varied as suggested by Thiéry (1967). Periods of 24, 48, and 72 hr were tried to ensure coupling of the thiol compound with the dialdehyde.

Thiéry's procedures (1967) were used for both tests with two modifications: sections were always treated initially with 2% sodium bisulphite for 30 min at room temperature or with saturated dimedone for 2 hr at 60°C (Pease, 1968) to block free aldehydes. In addition, we adopted Rambourg's suggestion that the staining period in the silver methenamine be performed twice in succession

Figure 11 (*top*) PA-AgP control, pH 6·4. If the periodic acid step is omitted, the starch grains are unreactive. The ERV's, PB's, and F's appear as they did in the test material. The lipid globules in the chloroplasts (L) are also eroded here as they were in the test sections. The mature protein body (PB) seen at the bottom of the micrograph lacks the cleavage fissures which usually subdivide the granule at this stage. The material in Figures 11 to 14 was fixed in glutaraldehyde-osmium. ERV — endoplasmic reticulum vesicle. F — filaments. S — starch. V — vacuole. W — cell wall. × 7,000.

Figure 12 (*bottom, left*) PA-AgP control, pH 6·4. Two developing protein bodies. No staining is evident in the matrix. × 22,500.

Figure 13 (*bottom, centre*) PA-AgP control, pH 6·4. Filaments in a vacuolar evagination. The faintly contrasted small granules that are seen among the filaments are part of their structure and do not represent silver grains (*cf.* Figure 6). × 22,500.

Figure 14 (*bottom, right*) PA-AgP control, pH 6·4. Higher magnification of the cell wall (W) and cuticle (C) to show the lack of staining. × 22,500.

(Rambourg *et al.*, 1969). Periodic oxidation was not performed in the control sections.

Jewell and Saxton's highly alkaline silver proteinate method (1970) was also tested, as those authors contend that staining sensitivity can be improved considerably by raising the pH of the solution to 9·2. They believe that under these alkaline conditions even a few reactive sites will result in a rapid accretion of silver.

Proteins

Swift (1966) first demonstrated that silver methenamine could be used as an ultrastructural stain for proteins. The reagent apparently reacts with the sulphydryl groups of cystine (Swift, 1966, 1968). Our treatment of the material followed Swift's schedule (1968) except that the sections were always pretreated with sodium bisulphite or dimedone and that the alcoholic solutions of benzyl thiol and iodoacetate were replaced by solutions of dithiothreitol and iodoacetate in 0·2 M borate buffer at pH 8·0. In addition, the alkylation step was begun on ice and subsequently allowed to warm to room temperature. The details of these modifications and an evaluation are covered in another paper (Burr, 1972).

The presence of proteins at the ultrastructural level may also be shown by extraction with various proteases. Glutaraldehyde-osmium fixed, Epon-embedded material was first treated with hydrogen peroxide to remove the osmium, which otherwise can inhibit protease activity (Douglas *et al.*, 1970). Pepsin (porcine stomach, 3X crystallized, Schwarz/Mann), pronase (Sigma), and protease (bovine pancreas, Schwarz/Mann) were employed using the methods of Douglas *et al.*, (1970). Incubation in 0·5% trypsin (bovine pancreas, 2X crystallized, salt free, Schwarz/Mann) in 0·02 M Tris-HCl buffer, pH 8, at 37 °C for 1 hr was also tested. For each of the treatments controls were run in which the enzyme being tested was omitted from the incubation medium.

Figure 15 (*top, left*) PA-AgM test. The membranes are obscured by the deposition of silver. However, it is still possible to distinguish a young protein body (ERV) in the centre of the picture which exhibits no staining which can be attributable to PA oxidation (*cf.* Figure 16). The material in Figures 15 to 18 was fixed in glutaraldehyde-osmium. ERV — endoplasmic reticulum vesicle. × 21,500.

Figure 16 (*top, right*) PA-AgM control. The result is similar to the test material. In control sections the starch grains (S) have a strongly reacting periphery. B — endogenous bacteria. × 22,500.

Figure 17 (*centre, left*) PA-AgM test. Developing protein body, cleavage just beginning. No notable staining by silver. Most of the electron density seen is due to osmium and not to silver. The latter can be distinguished as coarse granules deposited on the section. (*cf.* Figure 3). × 22,500.

Figure 18 (*centre, right*) PA-AgM control. Developing protein body, probably older than that shown in Figure 17 as the matrix becomes more electron dense in the older protein bodies. As in Figure 17, the electron density observed is due principally to osmium and not to silver precipitation (see caption for Figure 17). × 22,500.

Figure 19 (*bottom, left*) PA-AgM test. Glutaraldehyde only. Cytoplasm and filaments (F) in a vacuolar evagination which are unspecifically stained (*cf.* Figure 20). × 22,500.

Figure 20 (*bottom, right*) PA-AgM control. Glutaraldehyde-only. Unspecific staining of filaments (F) in a vacuolar evagination and surrounding parietal cytoplasm. V — vacuole. × 27,000.

Results and discussion

Periodic acid-Schiff's reagent (PAS)*

In semi-thin sections subjected to the PAS test, the starch grains were strongly reactive while the cell wall stained only a pale pink. Other components of the cell, including the protein bodies and filamentous material were negative. The amorphous matrix, which is present in most basal septa and which is also composed of the wound-healing material, showed no reactivity. No staining of any cellular constituent was detected in the controls.

Acid and sulphated polysaccharides*

There is no staining in either the thallus pieces or the wall preparations following treatment with Alcian yellow or Alcian blue for periods varying from 30 min to 2hr. (*Because the wound-healing material was always negative in these tests, and because of the costs of colour reproduction, Figures to illustrate these results have not been included).

Periodic acid-silver proteinate (PA-AgP)

Figures 7 to 10 show the results obtained in the PA-AgP test. The components of the cytoplasm lack their usual contrast because the sections have not been poststained with uranyl or lead salts (cf. Figures 1 to 6). None of the structures we have interpreted as being developmental stages of the wound-healing material — the endoplasmic reticulum vesicles with their homogenous matrix (ERV) (Figure 8), the fragmented protein bodies both within the cytoplasm and within the vacuole (PB) (Figures 7 and 9) or the filamentous elements (F) (Figures 7 and 8) — show any reactivity after the silver stain has been applied.

Staining of the cell wall is always very light but does appear to be enhanced if the pH of the silver proteinate solution is raised to pH 9·2 (Figure 10) as reported by Jewell and Saxton (1970). As usually employed silver proteinate is an aqueous solution with a pH of 6·4 (Jewell and Saxton, 1970; Thiéry, 1967). The low level of staining in the wall was not unexpected as Iriki *et al.* (1960) have shown that the walls of *Bryopsis* are composed primarily of xylan which is 1,3-linked. A 1,3-linkage would mean that the xylose chains would lack adjacent hydroxy

Figure 21 (*top, left*) AgM-C test. Prote [illegible] le than the cytoplasm. Also note strong staining of the cell wall. The material in Figures [illegible] glutaraldehyde-osmium. × 27,000.

Figure 22 (*top, right*) AgM-C control. Much [illegible] protein bodies (ERV) and the cytoplasm. Virtually all the staining in the cell wall (W) has [illegible]

Figure 23 (*centre, left*) AgM-C test. Mature [illegible] massive deposition of silver has formed a solid layer over the entire protein body. Cl [illegible] be detected. × 9,600.

Figure 24 (*centre, right*) AgM-C control. [illegible] in the cytoplasm. Staining has been greatly reduced although not eliminated. Th [illegible] plete alkylation. × 20,000.

Figure 25 (*bottom, left*) AgM-C test. Filame [illegible] with AgM is as marked as it is with the mature protein bodies, resulting in a thi [illegible] ments. × 16,000.

Figure 26 (*bottom, right*) AgM-C control. [illegible] ation. The affinity of the filaments to the AgM is far less than in the test sections. [illegible] 13,800.

groups which are necessary for detection with Schiff-type reagents. The slight staining of the wall that is observed in the PAS and PA-AgP tests is probably due to the groups at the ends of the chains or to minor polysaccharide components (Frei and Preston, 1964). Raising the pH, however, has no effect on any of the stages in the development of the wound-healing substance (Figures 8 and 9). Neither does increasing the length of exposure to TSC and TCH to up to 72 hr alter the results. The control sections are always free of any reactivity (Figures 11, 12, 13, and 14). Even the starch grains (Figure 11) and the cell wall (Figures 11 and 14) are unstained.

Periodic acid-silver methenamine (PA-AgP)

In both the glutaraldehyde-osmium (Figures 15 to 18) and the glutaraldehyde-only fixed material (Figures 19 and 20), the cytoplasm exhibits a rather pronounced staining in both the test sections and the controls. This non-specific background staining has been variously attributed to glutaraldehyde (Rambourg, 1967; Tramezzani *et al.*, 1964), the acrolein contaminant in glutaraldehyde (Rambourg, 1967), sulphydryls (Pickett-Heaps, 1967), or other reducing compounds pre-existing in the tissue (Rambourg, 1967; 1971). Periodic acid oxidation has been reported as greatly reducing the nonspecificity observed in glutaraldehyde-osmium fixed material. It is contended that this is because of the removal of osmium (Marinozzi, 1961; Pickett-Heaps, 1967). In our tests this background staining does not appear to be caused by the presence of osmium as it is also noted in the glutaraldehyde-only fixed material (Figures 19 and 20). It is quite probable that most of the background is caused by sulphydryl groups, as proposed by Pickett-Heaps (1967). In assessing our results, Rambourg's criterion (1967) was adopted: '. . . only those staining reactions which appear after periodic acid oxidation, but do not appear in nonoxidized controls, should be considered specific'. Employing this as a standard, our tests find no detectable differences in the wound-healing material at any stage of development in the test sections *versus* the controls (Figures 15 to 20).

Silver methenamine staining of cystine-containing proteins (AgM-C)

When silver methenamine is employed as a stain for localizing cystine-containing proteins, a heavy layer of silver is observed over the protein bodies, filaments, and the granular-fibrillar material (Figures 21, 23, 25, 27 and 28). The deposition is so heavy over the mature protein bodies and the filaments that it is impossible to resolve individual grains except at the edges of the structures. There is also a pronounced staining of the pyrenoids (Burr, 1972) and the cell

Figure 27 (*top*) AgM test. A dispersing mass of granular-fibrillar material (GF) in the vacuole with a filament (F) in the centre, all heavily stained with silver. At the top of the figure are more filaments which have not yet begun to disintegrate. The material in Figures 27 and 28 was fixed in glutaraldehyde-osmium. × 3,500.

Figure 28 (*bottom*) AgM-C test. Detail of the deposition of silver grains on the granular-fibrillar material. × 31,500 wall (Figure 21 and Burr, 1972). In the control sections, which have been alkylated before incubation in the silver stain, reactivity is greatly reduced, although not completely eliminated (Figures 22, 24, and 26). This is probably due to incomplete alkylation. Silver grains are still present over the cytoplasm with slightly more over the proteinaceous material. There are not, however, the solid plates of silver seen in the test sections. The difference in the cell wall, on the other hand, is striking. While the wall is highly reactive in the tests (Figure 21), in the controls the wall is usually completely free of silver (Burr, 1972) or sometimes a few grains are noted (Figure 22). Gotelli and Cleland (1968) have shown that a hydroxyproline-rich protein fraction is associated with wall preparations of *Bryopsis*. It is possibly this wall protein that is being stained.

Enzymatic digestion of proteins

Only pepsin showed good proteolytic activity. Under the conditions employed pronase, protease, and trypsin had no noticeable effect on the material. Even with pepsin the wound-healing substance could only be successfully extracted from the mature protein bodies (Figure 31) and the filaments (Figure 33). At the earlier stages in development when the material is beginning to accumulate within the endoplasmic reticulum cisternae, it is resistant to digestion (Figure 29). However, so are other cytoplasmic components which are known to contain protein such as mitochondria, endophytic bacteria, and ribosomes. The granular-fibrillar material which results from the degradation of the filaments (and sometimes also directly from the mature protein bodies [Burr and West, 1971]) is also pepsin-resistant (Figure 31). A number of reasons could account for the resistance of these two stages, which we know contain protein from the results of our AgM-C tests. (The stains for light microscopy cannot be used for these particular stages because of their small size.) Inextractibility could be attributable to imperviousness of the epoxy resin or to small amounts of osmium remaining within the section which could cause enzyme inactivation (Douglas *et al.*, 1970). Some osmium undoubtedly does remain even after extended treatment with hydrogen peroxide and accounts for the electron density seen in the

Figure 29 *(top, left)* Pepsin treatment. Young protein bodies are not noticeably affected by incubation in pepsin. The material in Figures 29 to 34 was fixed in glutaraldehyde-osmium and poststained by uranyl acetate and lead citrate. × 33,750.

Figure 30 *(top, right)* Pepsin control. The matrix of the young protein body and the surrounding cytoplasm are unaltered by the control medium. × 39,000.

Figure 31 *(centre, left)* Pepsin treatment. A mature protein body in the vacuole surrounded by granular-fibrillar material (GF). The protein body has been almost completely digested while the granular-fibrillar material remains intact. × 10,000.

Figure 32 *(centre, right)* Pepsin control. Mature protein body in the vacuole. No change in electron density. × 24,000.

Figure 33 *(bottom, left)* Pepsin treatment. Digestion of the filaments. × 10,500.

Figure 34 *(bottom, right)* Pepsin control. The filaments are unaltered by incubation in the medium lacking the enzyme. × 16,000.

sections. The use of glutaraldehyde-only fixed material had been considered but was rejected because the earliest and latest stages in development are impossible to identify with surety in such preparations. We cannot completely discount the possibility that the protein at these stages is associated with some other compound which is preventing it from being readily extracted. At least our present evidence indicates that if such exists it could not be a carbohydrate containing acidic, sulphate, or 1,2-glycol groups. The greater ability of pepsin to extract proteins from fixed and embedded material is probably related to its high activity, good stability, and rather broad spectrum of activity (Ryle, 1970).

Taken together, the results we have obtained with cytochemical procedures for both light and electron microscopy indicate that the wound-healing material is a highly proteinaceous substance, perhaps pure protein, which does not seem to be complexed with a polysaccharide. Isolation and characterization of the material will show whether or not this can be confirmed. The similarity in composition of the various stages indicated by these tests also lends some support to the previously proposed pathway for the formation of the wound-healing protein (Burr and West, 1971).

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CHEMICAL COMPOSITION OF THE WOUND PLUG AND ENTIRE PLANTS FOR SPECIES OF THE COENOCYTIC GREEN ALGA, *CAULERPA*

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Abstract: Levels of ash, NaOH-soluble protein, lipid, and TCA-soluble carbohydrate were determined for seven species of the green alga *Caulerpa*, for both the entire plant and wound plugs produced by cutting the rhizomes or blades. Insoluble carbohydrate was estimated by subtraction. Wound plugs had higher ash levels and a distinctly higher level of total carbohydrate. Both entire plants and wound plugs had low levels of lipid and protein. Carbohydrate is the major component of the wound plug in *Caulerpa* in contrast to protein reported for *Bryopsis*.

INTRODUCTION

Wound healing in coenocytic algae has been described (Haberlandt, 1929), but except for *Bryopsis* (Burr & West, 1971; Burr & Evert, 1972) no detailed studies on the chemical and developmental cytology of wound healing have been published. In *Caulerpa*, a highly differentiated coenocytic green alga, a unique and rapid wound healing mechanism has evolved which prevents excessive cytoplasmic loss. In about 15–30 sec after wounding the rhizome, blades, or rhizoids, a mass of gelatinous material rapidly accumulates at the cut end and develops into a compact, fibrillar matrix (Dawes & Rhamstine, 1967; Lohr, 1975).

The present investigation indicates that carbohydrate is the major component of the wound plug which is formed at cut ends of *Caulerpa* based on examination of 7 species representing 6 of the 12 recognized sections (Calvert, Dawes & Borowitzka, 1976). The composition of wound plugs and entire plants is compared in terms of total protein, soluble and insoluble carbohydrate, lipid, and ash. The species examined are: *C. ashmeadii* Harvey, *C. racemosa* (Foskål) J. Agardh, *C. prolifera* (Forskål) Lamouroux, *C. sertularioides* (Gmelin) Harvey, *C. paspaloides* (Bory) Greville, *C. lanuginosa* J. Agardh, and *C. cupressoides* (West) C. Agardh.

MATERIALS AND METHODS

Plants were collected from a number of sites along the west coast of Florida and the Florida Keys (Calvert *et al.*, 1976) in the fall. Wounding by cutting the rhizome and blades was carried out within 1–2 h after collection using cleaned and apparently healthy plants. Wound plugs were collected after 4–8 h by gently scraping off the wounded end with forceps. Entire plants and wound plugs were dried in a vacuum desiccator or in an oven (60 °C for 8 h) and stored in capped vials. Previous analyses established that there was no detectable change in organic levels when dried in an oven or in a vacuum desiccator. Samples were fine ground in a micromill tissue grinder immediately before analysis.

All analyses were carried out with at least two samples and a minimum of two replicates per analysis. A third analysis was carried out if there was a variation of more than 10% between replicates or between samples. Protein was determined after extraction for a minimum of 48 h with 1 N NaOH using the Folin reagent and a standard curve was run using albumen (Lowry *et al.*, 1951). Soluble carbohydrate was determined after extraction with 5% hot TCA (85 °C, for 4 h) according to the phenol–sulfuric acid method (Dubois *et al.*, 1956) and a standard curve was run using glycogen (Dubois *et al.*, 1956). Total lipid content was determined gravimetrically (Freeman *et al.*, 1957; Sperry & Brand, 1955). The inorganic fraction was determined by weight difference before and after treatment in a muffle furnace at 500 °C for 4 h. Longer periods or higher temperatures were avoided in ashing so that carbonate was not removed (Paine, 1971). Insoluble carbohydrate was estimated by subtraction of ash, protein, and TCA-soluble carbohydrate from the dry weight.

RESULTS

Ash levels for entire plants ranged from 28.4–61.4% and for wound plugs from 34.9–70.1% (Table I). With the exception of *C. racemosa*, ash in the entire plant was 60% or less of the dry weight. In all species, the ash content of the wound plug was consistently greater than the inorganic component of the entire plant.

Extracted protein levels ranged from 3.4–8.3% of dry wt and 6.9–12.4% of organic wt for the entire plant (Table I). A consistently lower level of protein was found in the wound plug for all species (0.6–3.0% of dry wt and 1.6–8.5% of organic wt).

Except for *C. cupressoides*, wound plugs had levels of soluble carbohydrate per g organic wt higher than that found in the entire plant (Table I). The range for wound plugs was 7.2–28.6% of dry wt and 23.0–43.9% of organic wt. Levels of soluble carbohydrate in entire plants ranged from 8.7–20.1% of dry wt and 22.5–30.8% of organic wt. When determined by subtraction, insoluble carbohydrate was high, both in entire plants (22.5–38.4% of dry wt; 51.3–63.2% of organic wt) and wound plugs (17.5–25.6% of dry wt; 36.2–66.2% of organic wt). Only *C. prolifera* showed a lower level of insoluble carbohydrate (organic wt basis) in the wound plug when compared to the entire plant.

In most species, the lipid content of the wound plug was less than that found in the entire plant (Table I). Entire plant lipid content ranged from 2.6–9.8% of dry wt and 5.2–15.2% of organic wt. Wound plug lipid levels ranged from 0.6–2.3% of dry wt and 1.9–5.7% of organic wt except for *C. prolifera*. Wound plugs collected from

TABLE I

Comparison of entire plant and wound plug material (%) in *Caulerpa* species; * insufficient wound plug material to carry out analysis on second sample; A, dry wt; B, organic wt.

	Ash	Lipid		Soluble carbohydrate		Insoluble carbohydrate		Protein	
Species	A	A	B	A	B	A	B	A	B
C. ashmeadii									
Entire plant	53.0	3.2	6.8	10.7	22.8	29.7	63.2	3.4	7.2
Wound plug	65.6	1.2	3.5	9.1	26.4	22.4	65.2	1.7	4.9
C. racemosa									
Entire plant	61.4	2.6	6.8	8.7	22.5	22.5	58.3	4.8	12.4
Wound plug	64.9	0.9	2.6	8.1	23.0	23.4	65.8	2.7	8.5
C. prolifera									
Entire plant	28.4	9.8	13.7	16.7	23.3	38.4	53.6	6.8	9.4
Wound plug	34.9	9.9	15.2	28.6	43.9	23.6	36.2	3.0	4.6
C. sertularioides									
Entire plant	39.6	6.1	10.1	17.9	29.5	32.1	53.1	4.4	7.3
Wound plug	62.7	1.3	3.5	13.7	36.8	21.4	57.5	0.8	2.2
C. paspaloides									
Entire plant	34.7	3.4	5.2	20.1	30.8	33.5	51.3	8.3	12.8
Wound plug	66.2	0.9	2.5	13.4	39.6	17.5	51.7	2.1	6.2
C. lanuginosa									
Entire plant	46.1	8.2	15.2	10.6	19.6	31.5	58.4	3.7	6.9
Wound plug	60.6	2.3	5.7	10.9	27.7	25.6	65.0	0.6	1.6
*C. cupressoides**									
Entire plant	34.3	3.8	5.7	10.7	30.0	34.7	52.8	7.5	11.5
Wound plug	70.1	0.6	1.9	7.2	24.1	19.8	66.2	2.3	7.8
Average of species									
Entire plant	42.5	5.3	9.1	14.9	25.5	31.7	55.8	5.6	9.7
Wound plug	60.7	2.4	5.0	13.0	31.7	22.0	58.2	1.9	5.1

C. prolifera had three times the highest lipid levels when compared with the other species (9.9% of dry wt; 15.2% of organic wt).

DISCUSSION

Average levels of ash, NaOH-soluble protein, lipid, and TCA-soluble carbohydrate for the seven species of *Caulerpa* examined differ in the entire plant and wound plug (Table I). Instead of protein, as found in the wound plug of *Bryopsis* (Burr & West, 1971; Burr & Evert, 1972), the dominant component in *Caulerpa* wound plugs is carbohydrate. The entire plant had higher average levels of lipid and protein while the wound plug had distinctly higher levels of ash. The levels of TCA-soluble and estimated insoluble carbohydrate varied among species with no pattern evident. There was a dominance, however, of insoluble carbohydrate over soluble carbohydrate in the entire plant for all seven species and in the wound plug for six species (excluding *C. prolifera*). Since wound plug formation occurs in sea water, it appears that the higher ash level may be due to the incorporation of salts into the wound plug. Of all the species, *C. prolifera* produced the most firm, compact and rapidly formed wound plug while the other six species with larger rhizomes produced softer, more gelatinous plugs.

Based on previous studies, we suggest that the insoluble carbohydrate forms the fibrillar network in the wound plug of *Caulerpa*, visible under the electron microscope (Dawes & Rhamstine, 1967). Using chromatographic procedures, Lohr (1975) demonstrated that in *C. prolifera*, the dominant sugar is glucose. Although the major structural (fibrillar) cell wall component in *Caulerpa* is a β 1,3-linked xylan (Mackie & Percival, 1959; Iriki *et al.*, 1960; Frei & Preston, 1964) an insoluble β 1,3-linked glucan is present in the cell wall and is also a product of photosynthesis (Howard, Gaylor & Grant, 1975; Howard, Grant & Foch, 1976; Howard, Wright & Grant, 1977) in *C. simpliciuscula*. It was suggested by Howard *et al.* (1977) that the soluble β 1,3-linked glucan is converted to an insoluble form in the wound plug. Our findings would support this suggestion. Further studies are now needed to establish the chemical sequence in wound plug formation in *Caulerpa* and related green algal coenocytes.

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DISTRIBUTION OF THE ALGAE BELONGING TO THE GENUS *CAULERPA* IN FRENCH POLYNESIA (ATOLL OF TAKAPOTO AND ISLAND OF MOOREA)

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ABSTRACT

Eleven species of algae belonging to the genus *Caulerpa* were found during several field trips in the atoll of Takapoto and in the island of Moorea: *C. bikinensis*, *C. serrulata* var. *pectinata*, *C. serrulata* var. *serrulata*, *C. pickeringii*, *C. racemosa* var. *clavifera*, *C. racemosa* var. *peltata*, *C. sertularioides* f. *brevipes*, *C. seuratii*, *C. urvilliana*, *C. verticillata*, *C. webbiana*. Only two species live in both sites: *C. pickeringii* and *C. racemosa* var. *peltata*.

In the atoll of Takapoto *C. bikinensis* forms very dense populations covering all the substratum from 40 m to more than 70 m on the outer slope. The biomass reaches 5 kg/m^2 (wet weight). Several scuba dives allowed us to check the presence of this *Caulerpa* all around the atoll.

INTRODUCTION

The genus *Caulerpa* (Caulerpales, Chlorophyta) includes more than 100 species which are widely distributed in all warm seas. A number of investigations dealing with their distribution show the particular biogeographical interest of the above genus. In French Polynesia, Montagne (1845), Weber van Boss (1910) and Setchell (1926) mentioned the presence of the following taxa: *C. cupressoides* (West) J. Ag.; *C. freycinetii* var. *typica* Weber van Bosse; *C. freycinetii* var. *de boryana* (J. Ag.) Weber van Bosse; *C. mamillosa* Montagne; *C. peltata* Lamouroux; *C. pickeringii* Harvey and Bailey; *C. racemosa* var. *chemnitzia* (Esper) Lamouroux; *C. racemosa* var. *clavifera* (Turner) Weber van Bosse; *C. racemosa* var. *laetevirens* (Montagne) Weber van Bosse; *C. sedoides* (R. Brn) C. Ag.; *C. sertularioides* (Gmelin) Howe; *C. seuratii* Weber van Bosse; *C. taxifolia* (Vahl) C. Ag.; *C. urvilliana* Montagne and *C. webbiana* Montagne. Since that time, two taxonomic reviews updating Boergesen (1932), Eubank (1946) led to some modifications in the above list: *C. peltata* = *C. racemosa* var. *peltata* (Lamouroux) Eubank and *C. freycinetii* = *C. serrulata* (Forsskal) J. Ag. emend. Boergesen.

The field trip that we made in French Polynesia (summer, 1978) allowed us to complement the inventory of *Caulerpa* in this part of the South Pacific Ocean and to better define the biotopes of the species found.

METHODS

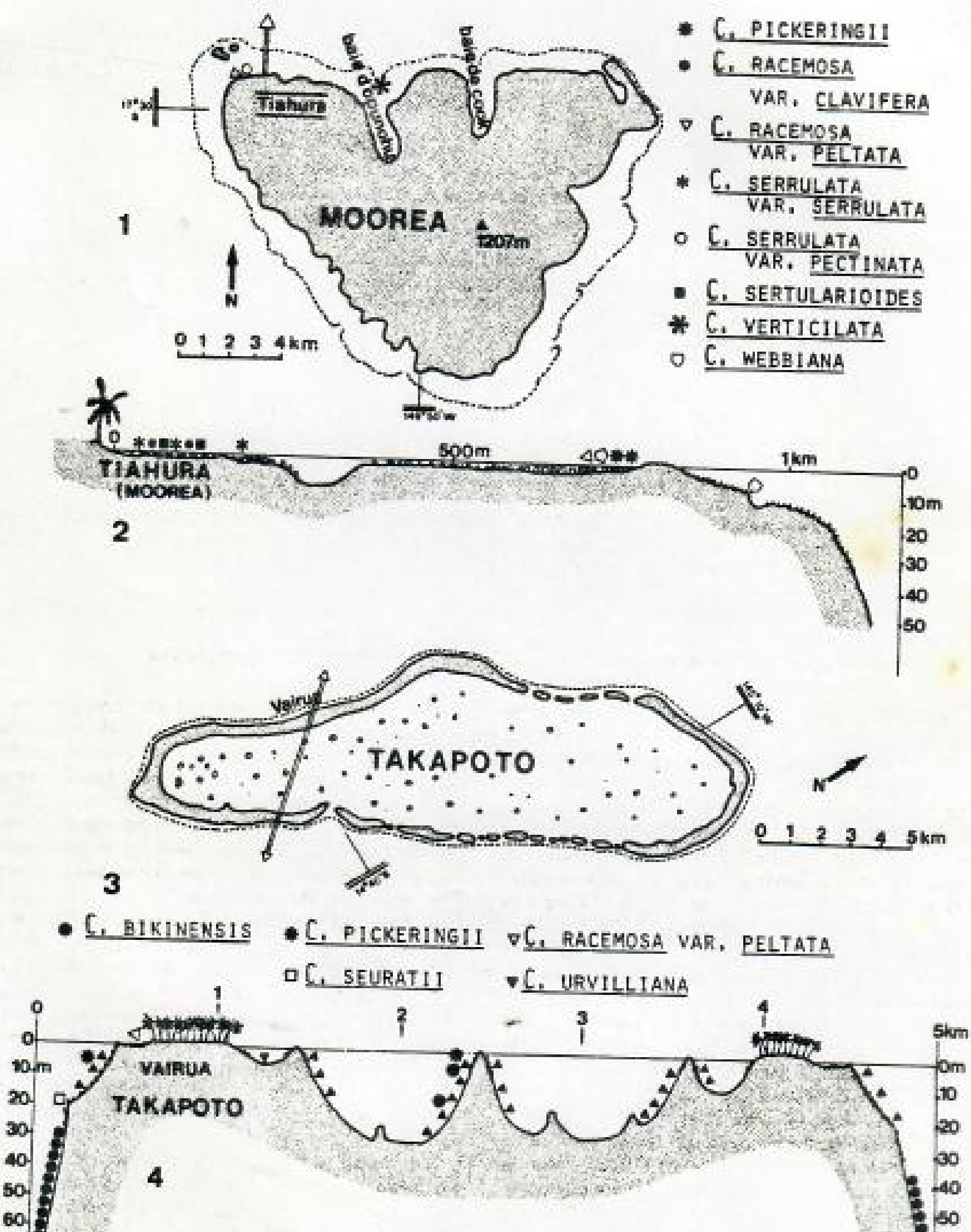
The different species listed herein have been sampled in the atoll of Takapoto and in the island of Moorea. The atoll of Takapoto (Figs. 3-4) belongs to the King George islands and is located in the northeastern part of the Tuamotu archipelago. It is characterized by an almost closed lagoon that communicates with the open sea by a few spillways (hoas) which flow intermittently in both directions. The island of Moorea faces Tahiti. It is a volcanic island circled by coral reefs (Figs. 1-2). Places we investigated were limited to the northwestern part of the island. Our survey was made by scuba diving. Logistic support was supplied by the "Antenne du Muséum National d'Histoire Naturelle et des Hautes Etudes" in Moorea, and by the "Service de la Pêche" in Takapoto.

RESULTS

We found a total of 11 species or varieties of *Caulerpa* in Moorea and Takapoto. Their exact location and the different characteristics of their biotopes are given here.

CAULERPA BIKINENSIS TAYLOR
Ref.: Taylor (1950. Pp. 66-67, pl. 33)

This species belongs to the section Sedoideae J. Agardh and to the group Claviferae which in-



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Figures 1-4. 1. Map of the island of Moorea. 2. Cross-section of the coral reef of Tiahura in Moorea. 3. Map of the atoll of Takapoto. 4. Cross-section of the atoll of Takapoto.

cludes *C. racemosa* and its various forms and varieties, amongst them specimens looking like *C. bikinensis*.

We found it in Takapoto on the outer slope as well as in the lagoon. On the outer slope, it forms a very dense vegetation (rate of coverage exceeding 75%) from a depth of 35 m. At a depth of 70 m, the vegetation is still very dense and extends downwards as far as is possible to see. *C. bikinensis* appears to form around the atoll an almost continuous belt. We checked its presence in several places distributed along the east coast (exposed to the prevailing wind and the resulting surf) and along the west coast (sheltered). Jaubert and Vasseur (pers. comm.) found it in July, 1974 and August, 1975. The biomass, measured from samples of 1 m^2, taken at depths of 40 to 45 m, reaches 5 kg/m^2 (wet weight).

In the lagoon, we found the above species in the dark microcavities of some pinnacles, within the 1 to 7 m depth range.

The presence of *C. bikinensis* is noted by Taylor (1950) in the Rongelap and Bikini atolls (North Pacific); by Dawson (1956) in the atoll of Eniwetok and Abbott (1961) in the atoll of Ifaluk (North Pacific).

The morphology of the thalli we sampled on the outer slope (Fig. 5) corresponds exactly to the characteristics given by Taylor. The size of the different parts of the thalli are slightly bigger than for the specimens from Bikini and Rongelap. Stolons: 50 cm to 1 m in length and 1 cm in diameter. Rhizoids: 15 cm in length and 5 mm in diameter at their insertion point on the stolon. The maximum length of fronds is 15 cm and their diameter varies from 2 to 3 mm. Most of the ramuli are alternate and distichous. Their size can reach 1.2 cm in length and 1 cm in width. Most of them end by a flat or slightly convex disc which suggests similarities with *C. racemosa* var. *chemnitzia*. Thalli sampled in the lagoon (Fig. 6) are significantly slender (diameter of stolons: 2 to 4 mm, length of fronds smaller than 10 cm).

Specimens of *C. bikinensis* which live on the outer slope of the atoll of Takapoto have more amyloplasts than chloroplasts.

CAULERPA PICKERINGII HARVEY AND BAILEY

Ref.: Harvey and Bailey (1851, p. 373); Weber van Bosse (1898, Pp. 272-273, pl. 21, Figs. 7 and 8)

We sampled this species in Moorea and Takapoto. In the lagoon of Takapoto, *C. pickeringii* is frequent in the 0.3 to 5 m depth range. On the outer slope, it grows deeper (7 to 20 m) and seems fairly infrequent. In Moorea, *C. pickeringii* proliferates on the algal ridge between 0.1 to 0.5 m, namely in front of Tiahura. Its geographical distribution seems to be limited to Polynesia (Denizot 1967) where it has been found by Harvey and Bailey (1851) and Weber van Bosse (1898), whereas it is not mentioned by Setchell (1926) in his list of the algae of Tahiti.

This species has been mistaken or identified as another species by several authors, either as *C. elongata* of Tanzania (Jaasund 1976-1977) or *C. webbiana* of Hawaii (*C. webbiana* var. *pickeringii* Eubank 1946). We consider that both species are different from *C. pickeringii* and have a much wider geographical distribution: Indian Ocean and Pacific Ocean for the former, Atlantic Ocean, Indian Ocean and Pacific Ocean for the latter. Due to these uncertainties the presence of *C. pickeringii* in the Caroline Islands, noted by Trono (1968) must be verified (only one thallus sampled).

In Takapoto, *C. pickeringii* (Figs. 11-12) is identical to the specimens sampled by Harvey and Bailey in Wilson Island and drawn by Weber van Bosse (1898, pl. 21, Fig. 7).

In Moorea, the stolons of *C. pickeringii* (Figs. 13-14) are most often masked by ramuli which form a very dense cover (2 to 5 cm in length; 3 to 4 mm in diameter). This form is identical to the ones described by Weber van Bosse (1898, pl. 2, Fig. 8) from a specimen sampled in Tahiti by Vernier.

Microscopic observations show that *C. pickeringii* of Takapoto differ from specimens of Moorea by the size and the shape of the ramuli. Ramuli of specimens from Takapoto fit the previous descriptions of Weber van Bosse (1898, pl. 21, Fig. 7): 0.5 to 0.6 μm in length, 130 μm in diameter at the lower part and 65 μm at the extremity, 3 dichotomies and angular extremities ended by a mucron. Ramuli of specimens from Moorea are slightly longer (0.65 to 0.7 mm) and slender (100 μm at lower part and 50 μm at the extremity). They exhibit 4 to 5 dichotomies and it is interesting to point out that their extremities are rounded (with a mucron).

The ramuli of both types of *C. pickeringii* form a uniform covering on fronds and stolons. Thus they look like *Codium*. It is not the case of *C. elongata* and *C. webbiana* whose stolons are partly stripped or carry only a few ramuli. This is to our opinion a very basic character which makes the difference between the above two species.

CAULERPA RACEMOSA VAR. *CLAVIFERA* (TURNER) WEBER VAN BOSSE

Ref.: Weber van Bosse (1898, p. 361, pl. 33, Fig. 5)

We found the above *Caulerpa* in Moorea. It is frequent on the backreef flats in very shallow waters where it is always exposed to full sunlight. *C. sertularioides* and *C. serrulata* var. *serrulata* grow in the same biotope.

C. racemosa var. *clavifera* is widely distributed in the warm seas (Atlantic, Indian and Pacific Oceans). It was sampled in Tahiti by Setchell (1926, p. 85). In Moorea we sampled a number of specimens (Fig. 15). All of them are characterized by the small size of the different parts of the thallus (a diameter of 1 mm for rhizomes; fronds are 2 cm high; between 1 and 3 mm in diameter for ramuli).

CAULERPA RACEMOSA VAR. *PELTATA* (LAMOUROUX) EUBANK

Ref.: Eubank (1946, p. 421)

In Takapoto the above *Caulerpa* grows in the outer reef flat and on the outer slope (3 m) in the most sheltered side of the atoll. In Moorea we found it on the fringing reef and on the reef front (1 m). It always grows in the shadow of microcavities. Setchell found it in Tahiti and called it *C. peltata* (1926, p. 86).

Characteristics of the specimens (Fig. 16) we sampled are: stolons measuring 1 to 1.5 mm in diameter; disc-like ramuli measuring 2 to 5 mm in diameter. Several specimens sampled in Moorea exhibit, on the same stolon, fronds typical of the var. *peltata* and others typical of the var. *clavifera*. This confirms the observations of Peterson (1972) concerning the growth forms resulting from light variations (var. *peltata* when irradiance is weak; var. *clavifera* when irradiance is strong).

CAULERPA SERRULATA VAR. *SERRULATA* (WEBER VAN BOSSE) TSENG (= VAR. *TYPICA*)

Ref.: Weber van Bosse (1898, Pp. 312-313, pl. 25, Figs. 4-5 = *C. freycinetii* var. *typica*)

The above species is frequent on the fringing reef of Moorea in sheltered and very shallow waters. It is always exposed to full sunlight. Setchell (1926, p. 85) found it in Tahiti and described it under the name of *C. freycinetii* var. *typica* (Weber van Bosse). *C. serrulata* var. *serrulata* has a very wide geographic distribution (Atlantic, Indian and Pacific Oceans). This alga exhibits different growth forms (Figs. 7-8) according to light variations. In fully illuminated biotopes, fronds less than 1.5 cm in length are smaller than in shaded biotopes (6 cm for the less lighted ones).

Several times, we found it in a reproductive stage (August 4, 5, 6, 13, 19 and 21, 1978). Reproductive papillae measure 1.5 to 2.5 mm in length and 180 to 200 μm in diameter. Thalli are monoecious, the male gametes (without stigma) are 8 μm in length while the female gametes are 10 μm in length. We observed several fusions which produced zygotes. This confirms the observations of Goldstein and Morrall (1970) and Price (1972) who described the complete life history of this alga.

CAULERPA SERRULATA VAR. *PECTINATA* (WEBER VAN BOSSE) NOV. COMB. = *C. FREYCINETII* VAR. *PECTINATA* WEBER VAN BOSSE

Reef.: Weber van Bosse (1898, Pp. 516 and 517, pl. 26, Figs. 4, 5, 6)

We found only one group of specimens under a pile wharf (depth 0.5 to 1m). This variety has been described by Weber van Bosse (1898) from specimens sampled by Mazé and Schramm in the West Indies (Guadeloupe). Mazé and Schramm noted it (1870, p. 78) under the name of *C. pectinata* Kützing. Meanwhile, Weber van Bosse (1898) pointed out that *C. serrulata* var. *pectinata* was completely different from *C. pectinata* described by Kützing (1849, p. 495 and 1857: Tab. 5, Fig. 4) which is nearer *C. crassifolia* (C. Ag.) J. Ag.

The characteristics of the thalli (Figs. 9-10) are: stolons 1.5 to 2 mm in diameter; fronds 1.5 to 5 cm in length. Fronds are very typical, they are laterally and regularly dentate and most of the time are divided. Their extremities are sometimes twisted, a character which allows us to link this variety to the *serrulata* species.

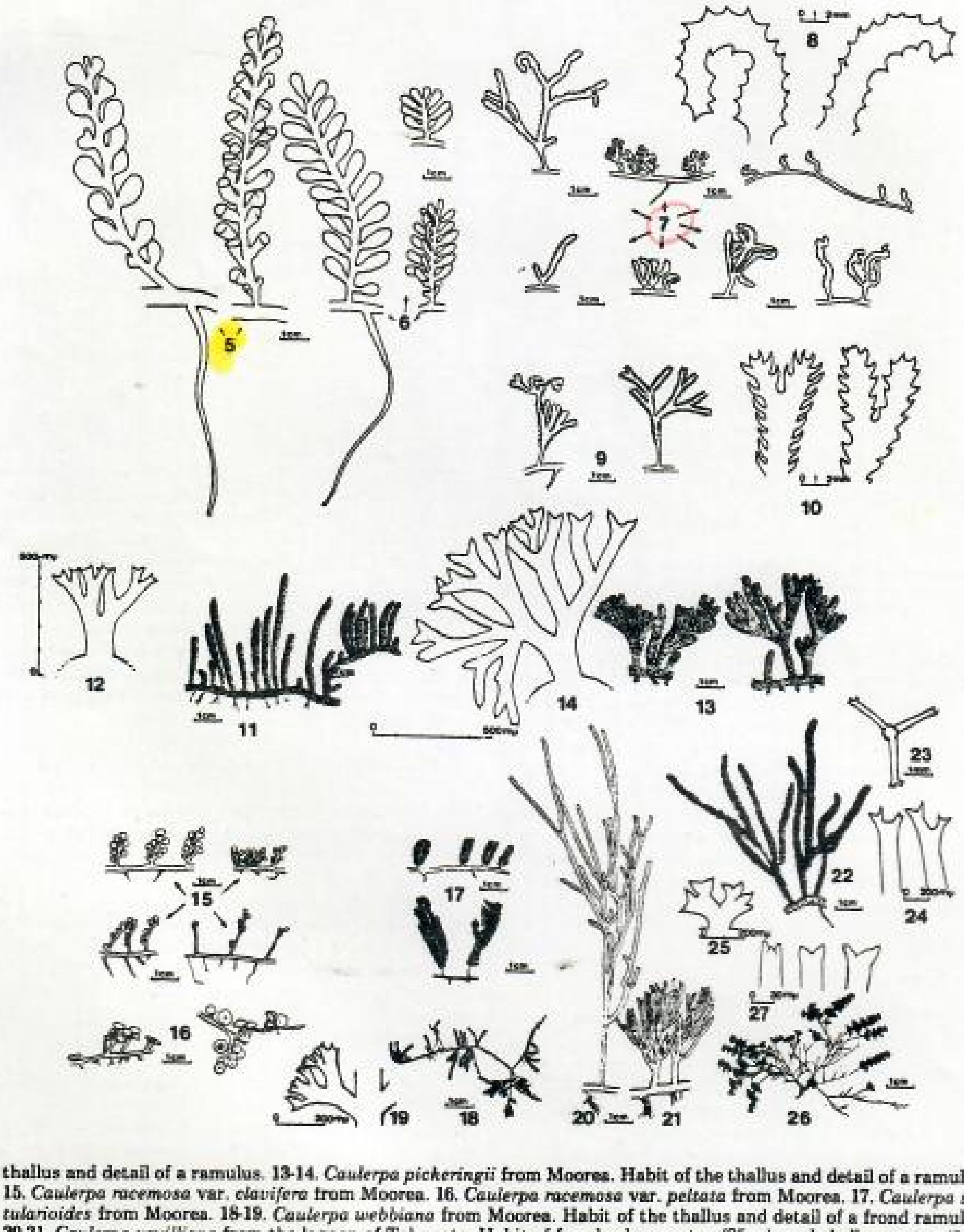
CAULERPA SERTULARIOÏDES (GMELIN) HOWE F. *BREVIPES* (J. AGARDH) SVEDELIUS

Ref.: Weber Van Bosse (1898, p. 294-295), (= *C. plumaris* f. *brevipes*)-Svedelius (1906, Pp. 114-115, Figs. 7, 8 p. 114)— Howe (1905, p. 576)

This species is frequent in Moorea on the back fringing reef in the 0-1.5 m depth range. Stolons tend to cover dense vegetations of *Halimeda incrassata* which was observed in Polynesia by Grunow (1868, p. 34 = *C. plumaris*) and by Setchell (1926, p. 84). It is widely distributed in the Indian and Pacific Oceans.

Stolons of the specimens we sampled are sometimes divided and can reach a length of 40 cm

Figures 5-27. 5. *Caulerpa bikinensis* (outer slope of Takapoto, 40 m). 6. *Caulerpa bikinensis* (lagoon of Takapoto, 7 m and 1 m). 7. Different forms of fronds of *Caulerpa serrulata* var. *serrulata* from Moorea. 8. Marginal serrations of fronds of *Caulerpa serrulata* var. *serrulata*. 9. Fronds of *Caulerpa serrulata* var. *pectinata* from Moorea. 10. Marginal serrations of fronds of *Caulerpa serrulata* var. *pectinata*. 11-12. *Caulerpa pickeringii* from Takapoto. Habit of the



thallus and detail of a ramulus. 13-14. *Caulerpa pickeringii* from Moorea. Habit of the thallus and detail of a ramulus. 15. *Caulerpa racemosa* var. *clavifera* from Moorea. 16. *Caulerpa racemosa* var. *peltata* from Moorea. 17. *Caulerpa sertularioides* from Moorea. 18-19. *Caulerpa webbiana* from Moorea. Habit of the thallus and detail of a frond ramulus. 20-21. *Caulerpa urvilliana* from the lagoon of Takapoto. Habit of fronds: deep water (25 m), and shallow water (3 m) (the smaller one). 22-25. *Caulerpa seuratii* from the outer slope of Takapoto. Habit of the thallus (Fig. 22); transverse section of a frond showing the triseriate insertion of pinnules (Fig. 23); dichotomous or trichotomous extremities of pinnules (Fig. 24); stolon ramulus (Fig. 25). 26-27. *Caulerpa verticillata*. Habit of a thallus and detail of the extremities of pinnules.

with a diameter of 0.8 to 1 mm. Fronds are small (Fig. 17), less than 5 cm. In the most lighted places (emerged at low tides) its size does not exceed 2 cm. The diameter of vertical axis varies from the base (0.8 mm) to the extremity (0.5 mm). Pinnules occur from the base of fronds (3 mm maximum between stolon and first pinnules).

We found reproductive specimens on the 20th of August, 1978. Pinnules bear reproduction papillae of 1.8 to 2.4 mm in length and 150 μm in diameter at the level of their bases and 90 μm at their extremities. Thalli are monoecious. Male gametes are located at the extremities of pinnules while female gametes are at their bases. Our observations agree with those of Goldstein and Morrall (1970).

CAULERPA SEURATII (WEBER VAN BOSSE)

Ref.: Weber van Bosse (1910, pl. 1, Figs. 5-9 and pl. 2, Fig. 1)

This species lives on the outer slope of Takapoto, between 17 and 25 m. About 20 thalli were found by diving in July, 1978. Jaubert sampled it in 1975 in the same place where it was very abundant. All the thalli (Fig. 22) belong to the forma *major* Weber van Bosse (1910). *C. seuratii* seems to be endemic to French Polynesia and it has not been published since the species was discovered by Seurat (in Weber van Bosse 1910). The thalli we sampled have stolons more than 40 cm long and 1 to 4 mm in diameter. They carry short ramuli (Fig. 25), 1 to 1.3 mm in length and 250 to 300 μm in diameter, which are characterized by 2 or 3 dichotomies and by mucrones at their extremities. Fronds are 1 to 7 cm long and are sometimes divided. They carry pinnules (Fig. 24) of 300 μm in diameter which are always disposed in three parallel lines along the axis (Fig. 23). The extremity of each pinnule is distichous or tristichous and ends with mucrones.

CAULERPA URVILLIANA MONTAGNE

Ref.: Montagne (1845, Pp. 21-22); Weber van Bosse (1898, p. 318-322; pl. 7: Figs. 7-11).

We sampled this species in Takapoto. It is frequent in the lagoon from 1 to 28 m. We found it also on the outer slope between 3 and 25 m.

Its geographical distribution is limited to tropical and pan-tropical areas of the Pacific Ocean. It was noted in Polynesia by Weber van Bosse (1910) and Denizot (1967 and 1972).

A number of sampled specimens exhibit several very different light-dependent growth forms (Figs. 20-21). Biometric investigations and light measurement allowed us to specify the relationship existing between irradiance and the different growth forms (Jaubert and Meinesz 1981).

CAULERPA VERTICILLATA J. AGARDH.

Ref.: Weber van Bosse (1898, Pp. 267-268, pl. 20, Figs. 7-10) — Svedelius (1906, Pp. 108-109, Fig. 1, p. 109) — Boergesen (1907, Pp. 355-357, Fig. 1, p. 356 and Fig. 2, p. 357)

This species was collected in Moorea in the reef pass of Papetoaii (Bay of Opunohu) under the wreck of Kersaint at a depth of 30 m, where it was growing on *Halimeda discoidea* Decaisne thalli. *C. verticillata* is widely distributed in the Atlantic, Indian and Pacific Oceans, but has never been found before in Polynesia. Stolons are divided and reach more than 15 cm in length and 225 to 375 μm in diameter. They carry ramified rhizoids whose lengths are very irregular. Vertical axes are 1 to 3 cm long and their diameter decreases from the lower (220-225 μm) to the upper part (70-85 μm). They carry superposed tufts of ramuli (Fig. 26) which are, most of the time, composed of 2 or 3 series of verticils; each verticil including 3 or 4 ramuli. Ramuli are 2.5 to 3 mm long and their diameter is 55-65 μm at the lower part, and 25-30 μm at the extremity. Each ramule is divided (3 to 5 dichotomies) and ends by 2 and sometimes 3 indentations (Fig. 27). All the ramuli of a series of superposed verticils are regularly distributed and form a tuft whose upper part reaches the lower part of the following tuft.

CAULERPA WEBBIANA MONTAGNE

Ref.: Montagne (1838, Pp. 146-151, pl. 6, Figs. 1 to 7). Weber van Bosse (1898, Pp. 269-271, pl. 21, Figs. 1 to 4)

We sampled specimens of this species in Moorea, in front of Tiahura in microcavities of the algal ridge and of the outer slope at a depth of 4 m. It is widely distributed in the Atlantic, Indian and Pacific Oceans and has been sampled in Tahiti by Setchell (1926, p. 84) and in Tahiti and Fakarava by Eubank (1946).

Our specimens (Fig. 18) correspond to the typical form described by Montagne (1838). Stolons can reach a length of 7 cm and their diameter varies between 400 and 500 μm. They carry a number of irregular rhizoids measuring 9 μm at their extremities. Vertical axes are 0.5 to 1 cm long and 200-300 μm in diameter. They carry short dichotomously branched ramuli (Fig. 19) and can reach a length of 300 μm for a diameter of 80 μm at the lower part and 30 μm at the upper part. Ramuli end by one or several mucrones exhibiting various shapes.

CONCLUSION

Among the 11 species or varieties of *Caulerpa* we found in Moorea or Takapoto, 3 are new for Polynesia: *C. bikinensis, C. verticillata* and *C. serrulata* var. *pectinata* nov. comb.

We found two species endemic in Polynesia: *C. seuratii* and *C. pickeringii,* but we underline that the last one has been confused or considered as being similar to *C. elongata* or *C. webbiana* which are widely distributed in tropical waters.

The distribution of the genus *Caulerpa* in French Polynesia shows that there are big differences in vegetation from one island to another. Thus among the 11 specimens sampled in Moorea and Takapoto, only two species were found in both places: *C. pickeringii* and *C. racemosa* var. *peltata.*

Two species are quantitatively important in the reef ecosystem: *C. bikinensis* and *C. urvilliana. C. bikinensis* forms a very dense belt from the depth of 35 m down to more than 70 m on the outer slope of the atoll of Takapoto. *C. urvilliana* is the most abundant macrobenthic alga of the lagoon of Takapoto.

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HAWAIIAN REPRESENTATIVES OF THE GENUS CAULERPA

BY

LOIS L. EUBANK

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ALTHOUGH the Hawaiian Archipelago has been known to the world of science since its discovery by Captain Cook in 1778, little work has been done on the marine algae of the region. A few publications may be mentioned as representative of the present state of knowledge, but on the whole they are little more than obsolete lists of some of the commoner seaweeds: Chamberlain (1880) recorded 112 species; Tilden (1901) listed less than 100 forms; Lemmermann (1905) included in his paper approximately 70 species based upon material collected by Shauinsland and upon the records of some of the earlier writers, such as Dickie (1876, 1877), De Toni (1889), Reinbold (1899) and others; Reed (1907) listed some 50 economic seaweeds; Rock (1913) published a catalogue which was an expansion of Reed's list; MacCaughey (1918) in addition to recording 115 species (compiled in part from the lists of Lemmermann and others), added notes of an ecological nature; Neal (1930) published the results of some successional studies of marine algae and gave an annotated list of 60 plants; and Howe (1934) gave an account, including the description of a few new species, of a small collection brought home by Galtsoff. Several additional references might be added, but knowledge of the flora is based primarily upon the papers cited above.

While at the University of Hawaii during 1941–1942, I undertook a study of certain Hawaiian marine algae. Collections were made on the Island of Oahu over a period of ten months. The present account is based on a study of the material of *Caulerpa* collected at that time, as well as that in the collections of Papenfuss, Reed, Rock, and Setchell which are deposited in the herbarium of the University of California.[*]

Previous records of *Caulerpa* in Hawaii included but eight names: *C. Chemnitzia* (Chamberlain, 1880), *C. clavifera* (Chamberlain, 1880), *C. asplenioides* (Chamberlain, 1880), *C. taxifolia* (Chamberlain, 1880; Tilden, 1901; MacCaughey, 1918), *C. pinnata* (Lemmermann, 1905; MacCaughey, 1918), *C. racemosa* var. *laetevirens* (Lemmermann, 1905; MacCaughey, 1918), *C. Pickeringii* (Neal, 1930), and *C. serrulata* (Howe, 1934). Their present status is discussed in this paper, along with an expansion of the list to include several new records for the Archipelago.

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GENUS CAULERPA

Caulerpa Lamouroux 1809, p. 136.

Thallus nonseptate, coenocytic, from 2 mm. to a meter in length, differentiated into a prostrate, branched rhizome and erect assimilative fronds, which usually consist of an axis beset with ramuli of diverse form; internally thallus traversed by trabeculae, which are extensions of the cell wall; reproduction vegetative and sexual, the latter anisogamous; gametes are liberated through papillae that develop on the frond, or occasionally on the rhizome.

Caulerpa is a genus exhibiting a high degree of polymorphism. In Hawaii, this is best exemplified by *C. ambigua*, *C. serrulata*, and *C. racemosa*. Before such a group can be attacked from the genetic, physiologic, or ecologic viewpoint, a taxonomic organization of the material seems an essential starting point. Whether the plasticity demonstrated by the species is due to a high genetic diversity within the population, to the effect of ecological factors, or to an interacting of both in the isolation of ecotypes, is not yet understood. Tandy (1934, p. 63) who has done some experimental culturing of members of the genus says, "The characters relied on for [specific] separations [within the genus] can be varied by transplant experiments on a single ramulus . . . There is evidence that *C. peltata* Lamx. and *C. fastigiata* Mont. are only forms of *C. racemosa*." Accordingly, we must admit that the boundaries which delimit certain "species" of *Caulerpa*, and the categories within such species, are undefinable at present.

KEY TO HAWAIIAN SPECIES

1. Fronds lacking ramuli. . . . . .5. *C. serrulata*
1. Fronds beset with ramuli. . . . .(2)
  2. Ramuli differentiated into pedicel and inflated apex. . . . .(3)
  2. Ramuli not pedicellate; filiform or tapering toward apex. . . . .(4)
3. Ramuli with constriction between pedicel and globose apex. . . . . .6. *C. lentillifera*
3. Ramuli lacking constriction, apex globose to plano-convex. . . . . .7. *C. racemosa*
  4. Ramuli distinctly mucronate. . . . .(5)
  4. Ramuli obtuse to acuminate, rarely, if at all, mucronate. . . . .(6)
5. Ramuli distichously arranged, simple, laterally compressed, sickle-shaped. . 4. *C. taxifolia*
5. Ramuli verticillately arranged, furcate, terete. . . . . .2. *C. Webbiana*
  6. Plant minute, 4 mm. to 2.0 cm. tall; ramuli in distichous or verticillate arrangement, simple or branched, obtuse, and reduced or lacking toward base of frond. . . . . .1. *C. ambigua*
  6. Plant 2.5 to 4 cm. tall; ramuli strictly distichously arranged, simple throughout, obtuse to acuminate, continuous to base of frond. . . . . .3. *C. sertularioides*

1. *C. ambigua* Okam.

C. ambigua Okamura 1897, p. 4, pl. 1, fig. 8–12.
C. Vickersiae Børgesen 1911, p. 129, fig. 2.

Plant minute, 4 mm. to 2.0 cm. tall, filiform; fronds erect, branched, supporting simple to quadridichotomous, cylindrical, verticillately, or distichously arranged ramuli; habit feathery to dense.

Caulerpa ambigua (pl. 22; fig. 2, *a*, *b*) is very minute and easily overlooked in the field. The majority of specimens were secured with the aid of hand lens and forceps from the protected surfaces of pieces of coral and rocks brought into the laboratory. Some were obtained from the sides of shallow pools. In the process of collecting, the delicate rhizomes were broken and the specimens are fragmentary. For this reason it is impossible to determine the exact nature of the entire plants. The several specimens collected show a diversity in form. A few fronds display a completely distichous arrangement of unbranched ramuli (pl. 22, *a*). In a second type of distichous frond, the ramuli are practically all dichotomous (pl. 22, *b*, *c*). Intermediate stages between these two were found. Other specimens possess verticillately arranged ramuli which branch profusely (pl. 22, *d–f*). Most plants show zonations which are believed to be the limits of periodic growth. *C. ambigua* is cited here for the first time from the Hawaiian Islands.

Some difficulty was encountered in the identification of *C. ambigua* owing to the belief of certain writers that the species embraces two distinct entities: *C. ambigua* Okam. and *C. Vickersiae* Børgs. A brief review of the history of the two may be of interest. In 1897 Okamura described a plant under the name *C. ambigua*, which he characterized as lacking a prostrate rhizome and possessing articulated rachises and ramuli. In the same paper he also pointed to the similarity between *C. ambigua* and *C. Okamurai* Weber-van Bosse (in Okamura, 1897), which has constricted (or at least pedicellate) ramuli. The articulations he thought of such significance that he placed his plants in the section Sedoideae of the genus, which includes only articulated members. Okamura (1897, pl. 1, fig. 3–12) illustrated *C. ambigua* with a series of ten figures. His figures 4, 6, and 7 show the constricted character, while figures 9 and 10 seemingly represent a plant which lacks articulations and is quite distinct from *C. Okamurai*. None of the ramuli in his figures are branched.

In her monograph of *Caulerpa* in 1898 (p. 388) Weber-van Bosse used the phrase "indistinctement articulé" in discussing *C. ambigua*, while in her Siboga paper of 1913 (p. 97), she pointed out that her *C. ambigua* material was more closely related to *C. elongata*, a member of the section Bryoideae, than to members of the section Sedoideae.

Vickers (1908, pl. 37, fig. 3–6), illustrated *C. ambigua* from Barbados as being distinctly nonconstricted, that is, similar to the nonarticulated plants of Okamura (1897, pl. 1, fig. 9, 10), but in no way resembling his constricted specimens (*op. cit.*, pl. 1, fig. 4, 6, 7).

Børgesen (1911, p. 129, fig. 2), in reporting his West Indian material, was led to the conclusion that Okamura had referred two species to *C. ambigua*, since his West Indian specimens agreed well with the figures of Vickers and with those of the nonarticulated plants of Okamura (1897, pl. 1, fig. 9, 10), but were unlike the latter's articulated specimens. Børgesen, therefore, proposed the name *C. Vickersiae* (originally *C. Vickersii*) for the plants without constrictions.

Taylor in 1928 (p. 104) accepted the specific name *C. Vickersiae* of Børgesen, and added a new variety, *C. Vickersiae* var. *luxurians*. In a later paper (1933, p. 396) he described a second variety, *C. Vickersiae* var. *furcifolia*, of the same species.

Okamura (1931, p. 101), in answering Børgesen's argument, says "... that in my plant two species are not confused may be seen from my Icones [1915] Pl. 139, figure 6, which is a portion of figure 3. In that figure, there may be some irregularities in the arrangement of ramenta having somewhat alternate manner, instead of regularly distichous, and moreover, some of ramenta have constricted base. Figure 5 may be taken as an extreme one of such a frond. Considering on the one hand, that *Caulerpa* is a plant taking several different forms in one and the same species, and on the other, that figure 3 which has not well-defined surculus is not different from figure 1 and 2, I think that my determination is justified."

In Yamada's report of the algae from the Ryukyu Islands (1934, p. 64) *C. Vickersiae* Børgs. is given as a synonym of *C. ambigua* without comment. Among his algae he found four specimens which he referred to *C. ambigua*, three of which were representative of the figure and the description of *C. Vickersiae* as given by Børgesen (1911, p. 129, fig. 2), while in a fourth specimen the ramuli are multiseriately arranged in a dense and irregular fashion.

The Hawaiian material was found to be unquestionably related to Okamura's Japanese plants as well as to Børgesen's *C. Vickersiae*. The diversity of form (pl. 22, and intermediate stages) approximates the range of conditions indicated by Okamura.

I have had occasion to examine some of Okamura's material of *C. ambigua* from Japan (Herb. Univ. Calif. 688774 from Nemoto [Bōshū] distributed as No. 95 in his Exsiccatae of Japanese Algae). All the characters illustrated in his original account of the species (1897, fig. 3–12) are well represented in the above collection. Several additional specimens furnish convincing connecting links between the misunderstood oriental plants and those from the other parts of the world discussed in the foregoing historical account. On the basis of these observations, which have been supplemented by examination of the Hawaiian plants, I conclude that but one species is involved, the proper designation of which is *C. ambigua*, and that *C. Vickersiae* should be considered a synonym of it. Such polymorphism within a species is not unusual for this genus. Okamura probably overstressed the importance of what appeared to be "spurius articulations" in a specimen which he later referred to (1931) as showing an extreme rather than a typical development.

In the material from Hawaii very slight constrictions were occasionally observed at the bases of some of the ramuli. No significance is assigned to them, however, as they are not a typical feature of the plants, and are very obscure at best, showing no regularity of occurrence. Weber-van Bosse's (1898, p. 388) statement "indistinctement-articulé" is open to the interpretation that there may have been very slight, although indistinct, constrictions in her plants. Furthermore, Taylor includes as a part of his original description of *C. Vickersiae* var. *furcifolia* (1933, p. 397): "very slightly contracted toward ramular bases." These admissions leave little basis for a split of Okamura's original *C. ambigua* into two species.

Before the absence of a prostrate rhizome can be relied upon as a distinguishing feature of *C. ambigua*, it must be remembered that in such a delicate and minute plant, the chance of destroying the rhizome in collecting is great. The specimens of Okamura and of Dodge, Dodge, and Thomas (Herb. Univ. Calif 688774 and 437251 respectively) have rhizomes which are partly prostrate.

The practice of supplying varietal designations to closely related plants within a species has been common in the genus *Caulerpa*, and is adhered to here, although these varieties are, for the most part, not clearly definable, being connected by transitional forms.

Type locality: Ogasawarajima (Bonin Islands).

Geographic distribution: Ogasawarajima (Bonin Islands); Nemoto (Bōshū); Ryukyu Islands; Kōtōsho (Botel Tobago); Virgin Islands; Barbados; Florida; Puerto Limon, Costa Rica; Oahu, Hawaiian Islands.

KEY TO HAWAIIAN VARIETIES

1. Fronds beset with simple to twice-dichotomous, distichously arranged ramuli; plant usually loose and feathery.......................................(2)
1. Fronds beset with densely furcate, multiseriately arranged ramuli; plant often heavily imbricate.......................................*d.* var. *furcifolia*
    2. Short simple ramuli alternating with longer dichotomously branched ramuli on a single frond.......................................*a.* var. *Vickersiae*
    2. Ramuli usually unbranched throughout.......................................*b.* var. *simplex*
    2. Ramuli usually once to twice-dichotomously branched throughout....*c.* var. *dichotoma*

1*a*. *C. ambigua* var. **Vickersiae** (Børgs.) comb. nov.

C. Vickersii (corrected to *C. Vickersiae*) Børgesen 1911, p. 129, fig. 2.

Fronds beset with zones of distichously arranged ramuli; short unbranched ramuli alternating with longer dichotomous ramuli.

Members of this variety are considered to be intermediate between *C. ambigua* var. *simplex* and *C. ambigua* var. *dichotoma* (cf. pl. 22, *a–c*). The figures of Børgesen (1911, p. 129, fig. 2, as *C. Vickersiae*) and of Yamada (1934, p. 64, fig. 33, as *C. ambigua*) are illustrative of this variety.

Børgesen (1913, p. 121), in discussing the characteristic periodic zonation in *C. ambigua*, implied that the shorter unbranched ramuli toward the apical region of each zone were less vigorous stages, and that the basal ramuli of each zone represented a more developed condition in which they became noticeably longer and dichotomized.

Collection: Papenfuss 10508, Feb. 15, 1942, Waikiki, reef in front of the Halekulani Hotel, Oahu.

Type locality: Virgin Islands.

Geographic distribution: Virgin Islands; Ryukyu Islands; Oahu, Hawaiian Islands.

1*b*. *C. ambigua* var. **simplex** var. nov.

C. ambigua var. **simplex** var. nov.

Planta disticha, frondes cum ramulis simplicibus.

Frond beset with distichously arranged unbranched ramuli, dichotomous ramuli rare to completely lacking.

Plate 22, *a*, illustrates the nature of the terete, filiform, unbranched ramuli which characterize *C. ambigua* var. *simplex*. Several of the plants of Okamura from Nemoto (Herb. Univ. Calif. 688774) and the material of Dodge, Dodge, and Thomas from Costa Rica (Herb. Univ. Calif. 437251) fit well into this category. The plants illustrated by Okamura in 1897 (figs. 7–10) are also representative of this variety. *Caulerpa ambigua* var. *simplex* might be considered to be in keeping with Taylor's *C. Vickersiae* var. *luxurians*[1] but as its members are not necessarily luxuriant in form, as is true of Okamura's plants from Nemoto, it seems best at present to regard them as separate entities.

That *C. ambigua* var. *simplex* is distinct from *C. ambigua* var. *Vickersiae* becomes more convincing in the light of the interpretation of Børgesen (with reference to *C. Vickersiae*) concerning the difference in form of the ramuli of a given frond. He suggested that the first-formed ramuli of each growth period of *C. Vickersiae* tend to be longer and to dichotomize. The striking feature of *C. ambigua* var. *simplex* is that the first-formed ramuli maintain an unbranched condition almost without exception.

Collections: Papenfuss 10507*a*, *type*, and 10507*b–f*, Feb. 15, 1942, Waikiki reef in front of the Halekulani Hotel, Oahu; Okamura (Herb. Univ. Calif. 688774), Nemoto; Dodge, Dodge, and Thomas (Herb. Univ. Calif. 437251), Costa Rica.

Type locality: Waikiki, Oahu, Hawaiian Islands.

Geographic distribution: Ogasawarajima (Bonin Islands); Nemoto (Bôshû); Costa Rica; Oahu, Hawaiian Islands.

1c. *C. ambigua* var. **dichotoma** var. nov.

C. ambigua var. **dichotoma** var. nov.

Planta disticha, frondes cum ramulis dichotomis.

Fronds beset with distichously arranged ramuli, which are usually once to twice dichotomous.

Variety *dichotoma* is proposed for certain Hawaiian plants (pl. 22, *b*, *c*) in which the ramuli throughout the frond are dichotomously forked. If the morphology of the ramuli is indicative of genetic variation in *C. ambigua*, as seems possible among varieties in certain other species of this genus, the simple or forked character of the ramuli of the varieties of *C. ambigua* would then serve to distinguish *C. ambigua* var. *dichotoma* from both *C. ambigua* var. *simplex* and *C. ambigua* var. *Vickersiae*.

Collections: Eubank 627, *type*, and 628, May 2, 1942, Marine Biological Laboratory at Waikiki, Oahu.

Type locality: Waikiki, Oahu, Hawaiian Islands.

Geographic distribution: Oahu, Hawaiian Islands.

1d. *C. ambigua* var. **furcifolia** (Taylor) comb. nov.

C. Vickersiae var. *furcifolia* Taylor, 1933, p. 396.

Fronds beset with densely furcate, multiseriately arranged ramuli; plant usually heavily imbricate.

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[1] A new combination of *C. Vickersiae* var. *luxurians* is apropos at this time even though no Hawaiian plants have been assigned to it: *C. ambigua* var. **luxurians** (Taylor) comb. nov. Taylor, 1928, p. 104.

The Hawaiian plants of *C. ambigua* var. *furcifolia,* as illustrated in plate 22, *d–f,* are well within the limits of the category as set forth by Taylor. There are obvious local variations in gross structure. Some plants possess distichously arranged simple ramuli at their terminal portions, becoming whorled and densely furcate toward the base of the frond (pl. 22, *e*). Except for the fact that the ramuli are forked, the habit of the plant in plate 22, *e,* is not far removed from Okamura's plants from Ogasawarajima, in which the ramuli are simple (1897, fig. 3, 4, 6). Several of his plants from Nemoto (Herb. Univ. Calif. 688774) actually have furcate ramuli. Other Hawaiian specimens show the multiseriate condition along the entire frond axis, and all ramuli appear to be forked (pl. 22, *f*). Yamada (1934, p. 65) states that the verticillate plants seem to branch much more profusely than the distichous. He gathered a specimen from Ryukyu which presumably fits into variety *furcifolia.* A duplicate of the material of Dodge *et al.* from Costa Rica (Herb. Univ. Calif. 437251) upon which *C. Vickersiae* var. *furcifolia* Taylor is based, completely lacks forked ramuli, thus showing that *C. ambigua* var. *simplex* as well as *C. ambigua* var. *furcifolia* occurs in Costa Rica.

Collections: Eubank 612–625, May 2, 1942, Marine Biological Laboratory at Waikiki; Eubank 626, May 16, 1942, reef at foot of Puu O Hulu, near Nanakuli, Oahu.

Type locality: Costa Rica.

Geographic distribution: Costa Rica; Ryukyu; Nemoto (Bōshū); Oahu, Hawaiian Islands.

2. *C. Webbiana* Mont.

C. Webbiana Montagne 1838, p. 129, pl. 6.

Plant small, .5 to 2 cm. tall; fronds erect, simple or branched, densely beset with whorls of furcate, mucronate ramuli, 4 or 5 per node; prostrate rhizome usually naked.

In Hawaii, *C. Webbiana* (fig. 1, *d–f*; 2, *c*) is not infrequently found on small rocks in shallow water or in tidal pools in coral reefs. Because of its small size it is often overlooked. At first glance it resembles a greatly dwarfed *Codium,* having a dark green spongy appearance. The ramuli of the frond come off in whorls of four typically (occasionally in whorls of five) as shown in figure 1, *e,* and are intricately branched near their apices, with one or two sharply definable hyaline mucrons at the tip of each ramular dichotomy. The only previous record of this plant from Hawaii was given by Neal (1930, p. 46) with the following comment: "Setchell determined this *Caulerpa* as very near to *pickeringii,* at least probably in the same group."

Collections: Papenfuss 10511, March 29, 1942, Halona; Papenfuss 10513 and Eubank 509, April 14, 1942, Waikiki reef, Honolulu; Papenfuss 10512 and Eubank 508, May 16, 1942, reef at foot of Puu O Hulu near Nanakuli; Eubank (leg. Conger) 562, Oct. 24, 1942 and 547, Nov. 1942, Waikiki reef at Marine Biological Laboratory, Honolulu; Herb. Univ. Calif. 622065 (leg. Neal), Oahu, Hawaiian Islands.

Type locality: Canary Islands.

Geographic distribution: Canary Islands; Barbados; Virgin Islands; Florida; Japan; Oahu, Hawaiian Islands.

2a. *C. Webbiana* var. **Pickeringii** (Harvey and Bailey) comb. nov.

C. Pickeringii Harvey and Bailey 1851, p. 373.

Plant 3 mm. to 4 cm. tall; creeping rhizome and erect fronds covered with whorls of furcate ramuli, 6–12 at each node; ramular apices mucronate.

So far as is known, this variety (fig. 1, *a–c*) is restricted to the islands of the South Pacific and does not occur in Hawaii. In the course of examining the herbarium material at the University of California, I came across a specimen from Oahu collected and identified by Neal as *C. Pickeringii* (Herb. Univ. Calif. 622065, referred above to *C. Webbiana*). Its striking resemblance to *C. Webbiana* prompted further study of *C. Pickeringii*. In most respects Montagne's (1838, p. 129) description of *C. Webbiana* and that of Harvey and Bailey of *C. Pickeringii* are similar. Examination of part of the original material of *C. Pickeringii* has shown the plant to differ from *C. Webbiana* in the following features: (1) The rhizome of *C. Pickeringii* is densely pubescent with ramular scales which are morphologically identical to the ramuli of the fronds, whereas the rhizome of *C. Webbiana* is usually glabrous. (2) *C. Pickeringii* is stouter, the rhizome and the fronds ranging from 2 to 2.5 mm. in thickness and the ramuli reaching a length of 1 mm. The Hawaiian material of *C. Webbiana* is noticeably smaller, the rhizome never exceeding 740 microns in thickness and the ramuli averaging about 600–740 microns in length. (3) In *C. Pickeringii* the ramuli occur in whorls of six to twelve (or possibly more) on the frond axis, whereas *C. Webbiana* usually has but 4 ramuli per node (cf. fig. 1, *c*, *e*).

The name *C. Pickeringii* has been overlooked by a majority of workers, including J. Agardh (1872, pp. 1–44) and De Toni (1889, pp. 441–487). Weber-van Bosse (1898, p. 272) apparently saw part of the Harvey and Bailey material but retained *C. Pickeringii* as an independent species. In a later paper (1910, p. 8) she suggested that *C. Pickeringii* is phylogenetically more advanced than *C. Webbiana*, and placed the two species side by side in the section Bryoideae. In my opinion, however, the differences between *C. Webbiana* and *C. Pickeringii* are not of sufficient magnitude to justify separation into distinct species and it seems best to reduce the latter to a variety of the former. Actually the habit of the two plants, as well as the ramular arrangement and the general morphology, appear to be identical. Even though this plant does not occur in Hawaii, a new combination seems justified at this time.

Collections: Eua, Tonga, Herb. Univ. Calif. 667642; Tahiti, Herb. Univ. Calif. 667634; Fakarava Atoll, Paumota Group, Herb. Univ. Calif. 667635; Wilson Islands, Paumota Group, Herb. Univ. Calif. 677620 (part of the original material of Harvey and Bailey of *C. Pickeringii*).

Type locality: Wilson Islands, Paumota Group.

Geographic distribution: South Pacific Islands including: Tonga; Tahiti; Wilson Islands and Fakarava Atoll, Paumota Group.

3. *C. sertularioides* (Gmel.) Howe

C. sertularioides (Gmel.) Howe 1905, p. 576.

Plant 2.5 to 4 cm. tall, yellow-green, feathery; frond axis cylindrical, erect; ramuli distichous, opposite, terete, simple, obtuse to acuminate.

This is a beautiful and delicate plant (fig. 2, *d*, *e*) found in warm quiet waters of tidal pools or creeping along the sandy sea floor. The species is recorded here for the first time from the Hawaiian Islands.

Collections: Papenfuss 10514, Sept. 3, 1940; Papenfuss 10518, Dec. 15, 1940; Papenfuss 10515, Mar. 30, 1941; Papenfuss 10517, Dec. 15, 1941; Papenfuss 10516 (leg. Conger); Eubank 511, Jan. 12, 1942; Eubank 514, Mar. 1, 1942—all from Hanauma Bay. Eubank 512, Nov. 2, 1941, near Makapuu Point, Oahu.

Type locality: Gmelin (1768) gave no definite locality other than "in corallis americanis."

Geographic distribution: In all tropical seas.

4. *C. taxifolia* (Vahl) Ag.

C. taxifolia (Vahl) Agardh 1822, p. 435.

Plant 3 to 10 cm. tall; frond axis erect, laterally compressed; ramuli about 4 mm. long, distichous, opposite, laterally compressed, simple, usually tending to be sickle-shaped, apices mucronate.

In Hawaii, *C. taxifolia* (fig. 2, *f*, *g*) is locally abundant in quiet water or in tidal pools. A comparison of the descriptions and of specimens of *C. taxifolia* and *C. crassifolia* (Ag.) J. Agardh (1872, p. 13) suggests that the two may be cospecific. In *C. crassifolia* the thallus seems to be of larger stature and the pinnae flatter and broader, but these are features of minor importance. The name *C. taxifolia* is retained for the Hawaiian plants since the material conforms to the descriptions of this species and since, in any event, this name antedates *C. crassifolia*.

C. taxifolia was first reported from Hawaii by Chamberlain (1880), later by Tilden (1901) and MacCaughey (1918). Chamberlain also listed *C. asplenioides* Grev., which name, according to De Toni (1889, p. 452), is synonymous with *C. taxifolia*. Through the kindness of Dr. David H. Linder, I have had the opportunity of examining the Chamberlain material of *C. asplenioides* and *C. taxifolia* in Herbarium Farlow. The specimens of both were found to be of the latter species. The determination of Tilden's specimen of *C. taxifolia* in her *Exsiccatae of American Algae* (1901, Century V, No. 452) has been verified also.

Lemmermann (1905), and later MacCaughey (1918), who took the record from Lemmermann, report *C. pinnata* (L.) Weber-van Bosse from Laysan on the northern extremity of the Hawaiian Archipelago. Since this name was employed by Weber-van Bosse for the plant currently known as *C. crassifolia*, an entity which, to my knowledge, has not been found in Hawaii, and since the latter resembles *C. taxifolia*, a quite common species from Hawaiian shores, it seems probable that the record in question applies to *C. taxifolia*.

Collections: Papenfuss 10520, Oct. 4, 1941, Puuloa Beach (leg. Kawahara), deep-water form; Papenfuss 10523, Nov. 2, 1941, near Makapuu Point; Papenfuss 10519, Nov. 23, 1941, Nanakuli reef; Papenfuss 10521 and Eubank 510, May 16, 1942, reef at foot of Puu O Hulu near Nanakuli, Oahu.

Type locality: St. Croix, Virgin Islands.

Geographic distribution: Virgin Islands; Barbados; Ceylon; Oahu, Hawaiian Islands.

5. *C. serrulata* var. *typica* (Weber-van Bosse) Tseng.

C. serrulata (Forsskål) J. Agardh emend. Børgs. var. *typica* (Weber-van Bosse) Tseng. Børgesen 1932, p. 5; Tseng 1936, p. 178, pl. 1.

Plant 1 to 4 cm. tall; fronds laterally compressed with mucronate serrations along the margins, sessile, usually twisted, asymmetrical, lacking ramuli but often with proliferations; texture membranaceous.

The nature of the marginal serrations of the frond, the frequent presence of twisted laminae, and the absence of pedicels distinguish variety *typica* from the other varieties of *C. serrulata*. The following three forms of this variety are represented in the material seen from Hawaii (fig. 2, *h–j*).

Howe (1934) reported *C. serrulata* from the Pearl and Hermes Reef which is at the northwesternmost tip of the Hawaiian chain.

Type locality: Red Sea.

Geographic distribution: Red Sea; Ceylon; Mariana Islands; Hainan; Ryukyu Islands; Formosa; Tonga; Philippine Islands; Oahu and Pearl and Hermes Reef, Hawaiian Islands.

5*a*. *C. serrulata* var. *typica* f. *lata* (Weber-van Bosse) Tseng.

C. serrulata var. *typica* f. *lata* (Weber-van Bosse) Tseng. 1936, p. 178, pl. 1.

Twisted fronds with distinct mucronate serrations along infolded margins.

Collections: Papenfuss 10524, Aug. 21, 1941; 10525, May 2, 1942, Waikiki reef at Marine Biological Laboratory, Honolulu. Papenfuss 10522, Dec. 15, 1940; 10510, Sept. 30, 1940; Papenfuss and Eubank (leg. Conger) 516, Sept. 20, 1942,—all from Hanauma Bay, Oahu. Herb. Univ. Calif. 622028 (leg. Rock), Oahu, Hawaiian Islands.

5*b*. *C. serrulata* var. *typica* f. **angusta** (Weber-van Bosse) comb. nov.

C. Freycinetii var. *typica* f. *angusta* Weber-van Bosse 1898, p. 313, pl. 25, fig. 4*b*.

Twisted fronds with distinct mucronate serrations along free margins, lacking serrations along infolded parts.

Collections: Papenfuss 10526, April 11, 1941, foot of Koko Head; Papenfuss 10574, Nov. 30, 1941, Black Point, Oahu; Herb. Univ. Calif. 622027 (leg. Reed), Hawaiian Islands.

5*c*. *C. serrulata* var. *typica* f. *serrulata* (Weber-van Bosse) Gilbert

C. serrulata var. *typica* f. *serrulata* (Weber-van Bosse) Gilbert 1942, p. 15.

Fronds scarcely twisted, if at all; marginal serrations regular throughout frond.

Collections: Papenfuss 10541, Mar. 1, 1942, Hanauma Bay; Eubank (leg. Conger) 545, Nov. 1942, Waikiki reef at Marine Biological Laboratory; Eubank 562, October 11, 1942, Marine Biological Laboratory, Waikiki, Oahu, Hawaiian Islands.

6. *C. lentillifera* J. Ag.

C. lentillifera J. Agardh 1837, p. 173.

Plant 1 to 3 cm. tall; fronds erect, cylindrical, with pedicellate, unbranched, globose ramuli, which are inserted verticillately; ramuli about 2 mm. long, constricted at point of attachment of pedicel to terminal head.

The fronds of *C. lentillifera* (fig. 2, *k, l*) resemble bunches of grapes. The constricted character of the ramuli is a constant feature of the species, and is the only character distinguishing this species from members of the *C. racemosa* group. It is conceivable that it is only a variety of *C. racemosa*. This is the first record of the occurrence of *C. lentillifera* in Hawaii.

Collection: Eubank 528, Jan. 12, 1942, Hanauma Bay, Oahu.
Type locality: Red Sea.
Geographic distribution: Red Sea; Madagascar; Ceylon; Oahu, Hawaiian Islands.

7. *C. racemosa* (Forsskål) J. Ag.

C. racemosa (Forsskål) J. Agardh 1872, p. 35.

Plant 2 mm. to 4 cm. tall; fronds beset with pedicellate ramuli, which may be globose, obovate, plano-convex, or discoid; ramuli at times occurring singly on the rhizome.

C. racemosa (fig. 2, *m–w*) is a composite of overlapping varietal types which differ from one another only in degree. Members of this species are common in tidal pools in the littoral zone, on reefs, or in quiet waters of the upper sublittoral belt. But two varieties of *C. racemosa* have been reported from the Hawaiian Islands. Lemmermann (1905), and later MacCaughey (1918) accredit *C. racemosa* var. *laetevirens* to Laysan. I have seen no specimen which can definitely be referred to var. *laetevirens*. *C. clavifera*, now known as *C. racemosa* var. *clavifera*, was reported from Oahu by Chamberlain (1880) but was not secured by me, nor were Hawaiian representatives of this variety found in the various other collections examined.

Geographic distribution: Throughout all tropical seas.

KEY TO HAWAIIAN VARIETIES

1. Ramular apex a flattened or cup-shaped disc, peltately attached to pedicel............(2)
1. Ramular apex clavate, pyriform, globose or plano-convex, usually pedicellate.........(3)
    2. Ramular disc with smooth margin; plant 1–3 cm. tall..................7*d*. var. *peltata* [illegible]
    2. Ramular disc scalloped or dentate at margin; plant 2 mm. to 1 cm. tall..7*e*. var. *exigua* [illegible]
3. Ramuli often occurring singly on rhizome, apex spherical.........................(4)
3. Ramuli on distinct assimilative fronds, apex pyriform to plano-convex..............(5)
    4. Plant minute, rarely taller than 6 mm.; ramular apex about 1–2 mm. in diameter ....7*a*. var. *microphysa*
    4. Plant stouter, 2 cm. or more tall; ramular apex 4–6 mm. in diameter .... 7*b*. var. *macrophysa*
5. Frond 5 mm. to 1 cm. tall; plant densely imbricate; ramular apex minute, 1 to 1.5 mm. in diameter, ranging on a single frond from pyriform to plano-convex in shape ....7*f*. var. *imbricata* not nearly so 6^2
5. Frond 1.5 to 3 cm. tall; ramular apex rather constantly plano-convex in shape, 1.5–2 mm. diameter.......................................7*c*. var. *turbinata* 6^2

7*a*. *C. racemosa* var. *microphysa* (Weber-van Bosse) Taylor

C. racemosa var. *microphysa* (Weber-van Bosse) Taylor 1928, p. 102, pl. 12, fig. 14; pl. 13, fig. 11.

Fronds 4 to 6 mm. high, thinly scattered on the stolon, branched or simple, giving rise to a loose arrangement of spherical, pedicellate ramuli, the latter often occurring singly on rhizome; ramular apex 1 to 2 mm. in diameter.

This variety (fig. 2, *m*) has not previously been reported from Hawaii.

Collection: Eubank 529, May 16, 1942, reef at foot of Puu O Hulu, Oahu.
Type locality: Macassar.
Geographical distribution: All tropical seas.

7b. *C. racemosa* var. *macrophysa* (Kütz.) Taylor

C. racemosa var. *macrophysa* (Kütz.) Taylor 1928, p. 101, pl. 12, fig. 3; pl. 13, fig. 9.

Fronds 2 or more cm. high, thinly scattered on the stolon, giving rise to a loose arrangement of spherical, pedicellate ramuli, the latter often occurring singly on the rhizome; ramular apices 4–6 mm. in diameter.

Caulerpa racemosa var. *macrophysa* (fig. 2, *n*) is reported here for the first time from the Hawaiian Islands. When Kützing (1857, p. 6) created the specific name *Chauvinia macrophysa* he described the ramuli as "inflatis obovatis subglobosis." Weber-van Bosse (1898, p. 361), who reduced the entity to a form of *Caulerpa racemosa* var. *clavifera*, characterized the ramuli as globose and 4 to 5 mm. in diameter. She also introduced a new form, *C. racemosa* var. *clavifera* f. *microphysa*, which is similar to *C. racemosa* var. *clavifera* f. *macrophysa* but with the globose ramuli measuring at most only 1 mm. in diameter. Taylor (1928, p. 101) records both forms from Florida, but elevates them to varietal rank under *C. racemosa*. However, his description of *C. racemosa* var. *macrophysa* is not in agreement with that of Kützing, for he describes the ramuli as convex or nearly so, distinguishing it from *C. racemosa* var. *microphysa*, in which the ramuli are spherical. Setchell apparently noticed this discrepency for he made a marginal note in his copy of Taylor's work to the effect that Taylor's plant was not based on the criteria employed by Kützing. In accord with Taylor, I regard these entities as varieties of *C. racemosa*, although it may be noted that the Hawaiian representatives of both varieties possess globose to subglobose ramular apices.

Collections: Papenfuss 10527, Sept. 3, 1940; Papenfuss 10528, Dec. 15, 1940, both from Hanauma Bay, Oahu.
Type locality: Coast of Central America.
Geographic distribution: All tropical seas.

7c. *C. racemosa* var. **turbinata** (J. Ag.) comb. nov.

C. clavifera var. *turbinata* J. Agardh 1837, p. 173.
C. racemosa var. *Chemnitzia* Weber-van Bosse 1898, p. 370.

Fronds 1.5–4 cm. high, erect, densely beset with plano-convex, trumpet-shaped ramuli approximately 2 mm. in length.

This variety (fig. 2, *o–q*) is very common in Hawaii, occurring at practically every locality visited. Close examination shows a diversity of ramular shapes to exist on a single plant or even upon a single frond. Figure 2, *t*, illustrates a common condition in which the diversity within a single plant is so great that the specimen can not be placed in any single taxonomic category. Clavate, "uviferoid," "laeteviroid," and peltate ramuli are all present. In some less extreme specimens, for the sake of convenience, classification is based upon the ramular type which is most prevalent on a given plant. Occasionally, proliferating ramuli were found (fig. 2, *q*) in which an outgrowth had developed from the apical margin and had become extended into another ramulus, which in turn produced proliferations.

For almost half a century *C. racemosa* var. *turbinata* (J. Ag.) has been known as *C. racemosa* var. *Chemnitzia* (Esper) Weber-van Bosse. Weber-van Bosse (1898, p. 370), in making the varietal combination from *Fucus Chemnitzia* Esper (1798), included as a synonym the *C. clavifera* var. *turbinata* of J. Agardh (1837); and from J. Agardh's description it is clear that *C. racemosa* var. *turbinata* actually is synonymous with *C. racemosa* var. *Chemnitzia*. According to Article 58 of the International Rules of Botanical Nomenclature (1935) the former has priority.

C. racemosa var. *turbinata* was reported from Hawaii by Chamberlain (1880) as *C. Chemnitzia*.

Collections: Papenfuss 10530, Sept. 3, 1940; Papenfuss 10529, Dec. 15, 1941; Papenfuss 10531, Jan. 12, 1942 (leg. Conger); Eubank 530, Mar. 1, 1942—all from Hanauma Bay. Papenfuss 10543, Nov. 23, 1941, Nanakuli; Papenfuss 10540, May 2, 1942, Waikiki reef in front of Marine Biological Laboratory, Honolulu; Eubank 560, May 31, 1942, Kahala reef; Herb. Univ. Calif. 622057 (leg. Reed), Koloa Point, Oahu.

Type locality: Red Sea.

Geographic distribution: All tropical seas.

7*d*. *C. racemosa* var. **peltata** (Lamx.) comb. nov.

C. peltata Lamouroux 1809, p. 145.

Plant 1 to 3 cm. high; fronds producing pedicellate, peltate ramuli with cup-shaped or flattened apical discs, 3 to 5 mm. in diameter; margins without serrations, frequent proliferations originating either from center or from margin of apical disc.

The Hawaiian material (fig. 2, *r*, *s*) presents convincing evidence that *C. peltata* is not a distinct species and should be reduced to a variety of *C. racemosa*. Numerous collections, such as that illustrated in figure 2, *t*, demonstrate transitional stages which often occur on a single plant. Even plants which at first glance appeared to be entirely peltate, proved upon examination to possess various erratic ramuli which were identical with the characteristic trumpet-shaped ramuli of *C. racemosa* var. *turbinata* or the clavate structure of *C. racemosa* var. *clavifera*. The removal of *C. peltata* to varietal rank under *C. racemosa* is further supported by the following suggestions of early and modern workers in the field:

Lamouroux (1809, p. 145) commented: "Cette espèce se rapproche singulièrement de la précédente [*C. Chemnitzia*]." He made reference to the diversity of ramular types found on a single plant, but attributed it to desiccation and suggested that it would not be found in living plants.

Zanardini (1858, p. 287) concluded that *C. peltata* Lamx. should be a variety of *C. Chemnitzia* because of its striking resemblance to it.

Weber-van Bosse (1898, p. 374) discussed the problem somewhat more fully: "Le *C. peltata* Lamx. a été considéré par Turner, Decaisne et Zanardini, comme une variété du *C. Chemnitzia*, et Harvey l'a déterminé pour *C. clavifera* var. *platydisca* [nomen nudum]. J'ai tâché de démontrer qu'il est plus logique de regarder le *C. Chemnitzia* comme une variété intermédiaire entre le *C. racemosa* et le *C. peltata;* ce dernier, vu les nombreux cas où la forme des ramules est constante, et les variétés qu'il fait naître, peut être considéré comme sous-espèce distincte. Je dis sous-espèce, car on rencontre de nouveau trop souvent des échantillons à ramules peltés et en massue, pour qu'on puisse fermer les yeux à l'étroit lien de parenté qui unit le *C. peltata* ou *C. racemosa.*"

Børgesen (1925, pp. 113–115, fig. 48, *a–c*) included the following remarks in his discussion of *C. peltata:* "The shape of the thallus is highly variable. . . . In other specimens the shape of the branchlets was not so markedly peltate (cf. fig. 48, *a*, *b*). The upper surface of the branchlets in these plants was not flat but often somewhat convex and the stipe was broadly conical making a rather even transition to the disc. . . . All the tufts I have seen had branchlets of more or less varying shape making even transitions from those of *Caulerpa racemosa* to var. *Chemnitzia* and further to *Caulerpa peltata.* Pure tufts were found only of the latter form, for which reason I refer the different forms found to this 'species' [*C. peltata*]."

Gilbert (1942, p. 22), basing his observations on material from the Philippine Islands and Java, remarked: "In this species [*C. peltata*] have been included all those specimens with few to all peltate ramuli. Transition forms are found in the specimens listed under var. *typica;* they vary from plants in which all ramuli are peltate to ones in which only a few of the ramuli are typically peltate and the others range in shape to the clavate form of *Caulerpa racemosa* var. *clavifera,* the trumpet-like and imbricate ramuli of *C. racemosa* var. *Chemnitzia,* or the suddenly expanded spherical-tipped ramuli of *C. racemosa* var. *occidentalis.* All these transition forms may at times be found on the same plant and largely even on the same frond. This serves to illustrate once again that the line distinguishing *C. peltata* from *C. racemosa* is apparently quite arbitrary."

No previous record has been made of this variety from the Hawaiian Group.

Collections: Papenfuss 10532, Sept. 3, 1940; Papenfuss 10533, Mar. 1, 1942, and Eubank 540 (leg. Conger), Sept. 20, 1942—all from Hanauma Bay. Eubank 519, Nov. 23, 1941, Nanakuli; Eubank 523, Nov. 30, 1941, Black Point; Eubank 527, May 16, 1942, reef at foot of Puu O Hulu; Eubank 518, Mar. 27, 1942, between Blow Hole and Makapuu Point, Oahu.

Those collections which are not classifiable owing to the diversity of ramular types are: Papenfuss 10534 and Eubank 524, Nov. 30, 1941, Black Point; Eubank 521, Mar. 1, 1942, Hanauma Bay; Papenfuss 10535 and Eubank 525, April 19, 1942, Halona Reef, Oahu.

Type locality: Antilles.

Geographic distribution: All tropical seas.

7e. *C. racemosa* var. **exigua** (Weber-van Bosse) comb. nov.

C. peltata var. *exigua* Weber-van Bosse 1898, p. 377.

Plant small, 2 to 4 mm. to occasionally 1 cm. in height; rhizome usually producing single ramuli, occasionally fronds; flattened peltate ramular discs 1 to 3 mm. in diameter with irregular serrations and proliferations at the margins.

This is the first record of *C. racemosa* var. *exigua* (fig. 2, *u*, *v*) from Hawaii. The plants of this variety appear to be more constant in their form than *C.*

racemosa var. *peltata*. *C. racemosa* var. *exigua* is considered to be of equal rank with the preceding members of the *C. racemosa* complex because the serrations of the apical margins of the ramuli delimit these plants as sharply as the various shapes and sizes of the ramuli of other varieties of *C. racemosa*.

Collections: Papenfuss 10536, Mar. 29, 1942; Papenfuss 10537, April 19, 1942, Halona Reef; Papenfuss 10538 and Eubank 533, May 16, 1942, reef at foot of Puu O Hulu near Nanakuli, Oahu.

Type locality: Not indicated by Weber-van Bosse (1898, p. 377).

Geographic distribution: Samoa; Mangaia; Oahu, Hawaiian Islands.

7*f*. *C. racemosa* var. **imbricata** (Kjellm.) comb. nov.

Chauvinia imbricata Kjellman, in Wittrock and Nordstedt, Exsiccatae, No. 436.

Caulerpa peltata var. *typica* f. *imbricata* Weber-van Bosse 1898, p. 375.

Plant small, .5 to 1 cm. tall, densely imbricate; ramuli rarely over 1 to 1.5 mm. in diameter, with inflated apices which vary from plano-convex to obovate.

Weber-van Bosse (1898, p. 375) placed *C. racemosa* var. *imbricata* as a form of *C. peltata*, while Svedelius (1906, p. 134) retained its specific rank. Svedelius's figures 37, 38, and 40 to 42, which are based on Ceylon plants, resemble the habit of the Hawaiian plants. Because the ramular types of the plants in question are strictly within the limits of the *C. racemosa* group, a separate species designation is superfluous. *C. racemosa* var. *imbricata* is considered here to be based on the small and densely imbricate nature of the ramuli (fig. 2, *w*), which may range in shape from obovoid to peltate on a given plant. However, in assigning varietal rank, I admit doubt about its correct status. Its rather constant growth form seems to justify consideration of it as an autonomous entity. On the other hand, it may be but an ecological variant of any one of several varieties of *C. racemosa*. No previous record of this variety has been made from Hawaii.

Collections: Eubank 532, Jan. 12, 1942; Eubank 559, Mar. 1, 1942, Hanauma Bay; Eubank 531, Mar. 27, 1942, between Blow Hole and Makapuu Point; Eubank 561, April 19, 1942, Halona Reef; Papenfuss 10542, April 1, 1942, Waikiki Reef at Elks Club, Honolulu; Papenfuss 10539, May 2, 1942, Waikiki reef in front of Marine Biological Laboratory; Eubank 520, May 16, 1942, reef at foot of Puu O Hulu, Oahu; Herb. Univ. Calif. 622066 (leg. Reed) foot of Diamond Head, Oahu, Hawaiian Islands.

Type locality: Ceylon.

Geographic distribution: All tropical seas.

THERE ARE ALIENS AMONG THE ALGAE, TOO--OR LIMU MALIHINI

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This past summer, many residents of Kūau, a coastal section just southeast of Pā'ia, Maui Island, were up in arms over the appearance of masses of a "new" seaweed on their beaches. Indeed, the culprit alga was new to Maui, having come over from O'ahu Island where it had been spreading since its introduction to Kāne'ohe Bay about 1974. The red alga alien is *Hypnea musciformis* which is common in the Caribbean and in the warm Atlantic (carried by the Gulf Stream). Stories about its introduction are not agreed to by all parties, one saying that it was deliberately introduced, another saying that it was accidental, having come with another species of algae as an epiphyte from the Caribbean. I began to record its spread in the Fall of 1978 when I found it at Ka'a'awa (not far from the mouth of Kāne'ohe Bay), and in the Fall of 1979 it was at Lā'ie Point; in 1980 at Waikīkī; 1981 at 'Ewa Beach and by 1982 when I moved here and was able to get out more often it seemed to have reached every nook and cranny of intertidal O'ahu. Although principally an epiphyte, and ideally suited for that mode of existence due to large hooks that are modified tips of branches, *H. musciformis* also occurs on rock. It makes up between 1/3 to 1/2 of the wet biomass of most drift piles on the beaches, and it is my (unscientific) opinion that it accounts for larger masses than previously of *Sargassum* on the beaches, since it adds drag and weight to the *Sargassum* plants. *Sargassum* was frequently in the drift in late Summer and Fall in pre-Hypnea times.

When *Hypnea musciformis* was introduced to Hawai'i, it was thought to be a good source of kappa carrageenan, a useful colloid usually obtained from species of *Eucheuma*, red algae that grow naturally in the Philippines where the bulk of kappa carrageenan "weed" comes from. However, because of its delicate thallus, barely 0.5 to 1.0 mm diam., it is difficult to process ("clean") for colloid extraction (commercial *Eucheuma* species, by contrast are frequently 4.0 mm or more in diameter).

I seriously suggested to the Maui folks that they cook with *Hypnea*, extracting the gelatin-like colloid and using it in molded salads, or desserts like *yokan*, or use it as a fertilizer in their gardens. Do anything to get it off the beaches so as to short-circuit the life history. Haven't heard what happened since. *Hypnea musciformis* has turned out to be a worse pest than the earlier introduced *Acanthophora spicifera* which was noticed some time after World War II years (Doty 1961).

There was a short period during the 1970s when it was thought desirable to investigate potential domestic sources of

The most recent seaweed introduction under permit is *Gracilaria tikvahiae* from Florida, imported to grow under aquacultural conditions because the "natural" species in Hawaii (*G. parvispora*, also erroneously known as *G. bursa-pastoris*, a species that does not occur in Hawaii) has a normal low growth period in the winter. Under some protest from the advisory committee, this species was introduced, and subsequently has escaped into the intertidal. Whether or not it will spread is too soon to say. It is enjoying a commercial success as "ogo" (locally the favorite seaweed to be eaten with raw fish). If it becomes widely distributed, people will harvest it for their own food supply, and so partly control its spread, but as a phycologist, I have no idea (as no one does) how it might live with the seaweeds that are already here. The best thing that can be said for it at this time is that it cannot vegetatively propagate itself by using hooks that *Hypnea musciformis* has.

Recently, an application to import several species of agar-producing seaweeds was made with the Department of Agriculture. At their urging, I was encouraged to file a negative evaluation of the request, and so strongly suggested that a few experiments that might include sea water temperature tolerance and other physical tests be made before applications were approved. At the same time, I encouraged the applicant to experiment with local species in the same genera before seeking alien species. I was pleased that my recommendations were sent to the applicant, who agreed to hold off until more investigations on local species had been made.

There may be other species of seaweeds introduced to Hawaiian waters without anyone's knowledge, just as there appear to be flowering plants that are introduced as seeds. Many culinary herbs, for example, are found in some of the ethnic stores in Honolulu for which there are not specimens in the Bishop Museum herbarium; some of them are frequently used for their leaves alone so no fertile portions are obtained and it becomes difficult to identify them. We know nothing, or very little about their habits, their growth patterns, their competitive abilities and, like the dozens of ornamental plants that are introduced each year to Hawaii, we have no idea what they will do to the "permanent party". How many will become the pestiferous "java plum", *Lantana camara*, "banana poka", "Koster's curse", or even *Wedelia* which is now at the top of Mount Ka'ala?

Some day, perhaps sooner than we know, it may be impossible to do a baseline study of a shallow subtidal bottom with a "normal" seaweed flora. Both *Hypnea musciformis* and *Acanthophora* are now so widespread and abundant that one cannot help but wonder "what species were there before these came?" I would liken the occurrence of *Convolvulus arvensis* (field bindweed, introduced before 1927, according to St. John, 1973) and *Pluchea indica*, introduced before 1922 (St. John, 1973) to these two seaweed aliens, in habit as well as abundance and distribution.

colloid-producing algae (Abbott 1980) since overseas operations frequently had a boom-and-bust nature, or governments were unstable, and because the demand for colloids frequently exceeded the supply by 10-20 times (Doty 1973). It was in this period that *Hypnea musciformis* was brought to Hawaii without a permit.

In this period, also, Dr. M. S. Doty of the Botany department, University of Hawaii at Manoa, brought a species of *Eucheuma* to Hawai'i *under permit* for experimental work in Kāne'ohe Bay. As is common with many newly introduced plants, there was a very large "bloom" of this very conspicuous species, some plants being more than a meter high, with thick branches up to 5 or more mm diameter. It was quickly named "The Green Blob" and a lot of energy was spent both in trying to get rid of it near Hawaii Institute of Marine Biology, and talking about it. Dr. Doty said at the time that as soon as the plants went over the reef edge into deeper water, they would be killed since they could not photosynthesize at depth. This had turned out to be the case. Nowadays it has seemed to establish itself in small pockets in Kāneohe Bay, and has not in my experience at least, left the Bay. This species is called "giant ogo" and is available for sale at Tamashiro Market in Kalihi most days. (It makes a fairly good pickle in the Japanese style "namasu", and is outstanding in a spice cake).

Several other seaweeds have been brought to Hawai'i in the past few years under permit. Included is *Macrocystis pyrifera*, the so-called "Giant Kelp" which is the food of choice for the abalone that are being grown under aquaculture conditions near Keāhole Point. The reason this species can be grown at Keāhole Point is because of the cold water which is brought up from depth, and is thought to be controlled from spreading because surface sea water temperatures are too warm for the kelp to complete its life history. However, occasionally small sporophytes (the spore-bearing kelp plant) may be seen and have been a source of concern to Dr. E. Alison Kay, who is a member of an advisory committee for the State Department of Land and Natural Resources. The advice of this committee is sought when applications are made for importation of plants or animals. Without living near Keāhole Point and being able to monitor the status or progress of such alien plants, I could only predict, out of experience in California, that the plants would die before they reached spore production. (Such plants have been physically removed before such stages were reached, so the value of the prediction will never be known).

At about the same time, a species of *Porphyra* (nori) was brought in under permit from Japan, again with the understanding that its life history could not be easily completed in surface sea water temperature, and this has proven to be the case.

LITERATURE CITED

Abbott, I. A. 1980. Some field and laboratory studies on colloid-producing red algae in central California. Aquatic Bot. 8: 255-266.

Doty, M. S. 1961. Acanthophora, a possible invader of the marine flora of Hawaii. Pac. Sci. 15: 547-552.

-----. 1973. Farming the red seaweed, Eucheuma for carrageenans. Micronesica 9: 59-73.

St. John, H. 1973. List and summary of the flowering plants in the Hawaiian Islands. Pac. Trop. Bot. Gard. Mem. 1: 519 pp. Lawai, Kaua'i, Hawai'i.

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CONFERENCE ANNOUNCEMENT

RESTORING THE EARTH 1988 - The first national gathering to consider the restoration of all natural resource types and the redesign of urban areas will be held on January 13-16 at the University of California, Berkeley. The conference is organized by the Restoring the Earth project of The Tides Foundation, San Francisco, and cosponsored both by the College of Natural Resources and the Center for Environmental Design Research of the University of California, Berkeley. It will bring experts in natural resource restoration and management together with a broad selection of academic, government, industry, foundation, labor, public health, and environmental representatives. Participants will help create new solutions to the nation's environmental problems, through restoration of damaged resources.

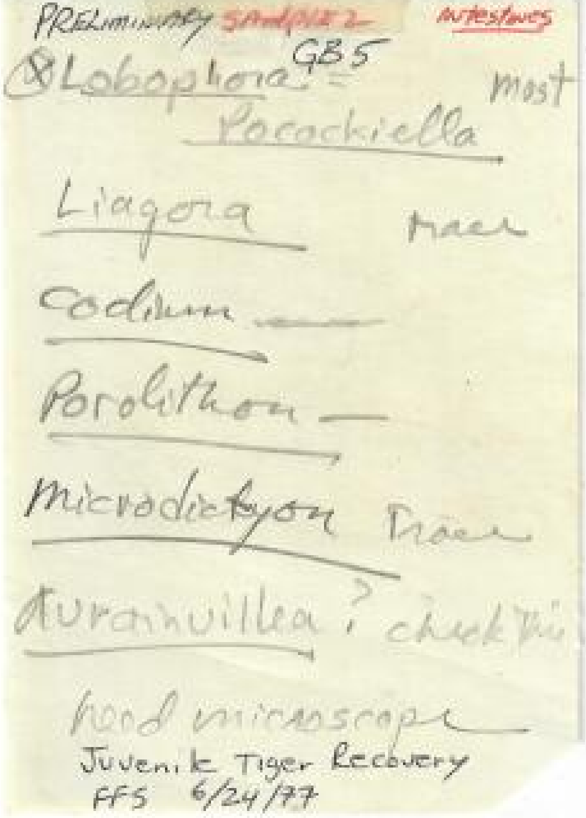
Topics to be covered include restoration of coastal ecosystems and estuaries; rivers and lakes; streams and fisheries; rangelands, prairies, mined lands, forests and wildlife; atmosphere and climate; dry lands and agricultural lands; urban environmental planning; and control of toxic wastes. Formal refereed papers will be presented at scientific and technical sessions. Non-technical sessions will include accounts of restoration successes and discussions of policy issues, legislation, litigation, trends, and resource conflict resolution. The program also includes keynote panels, plenary sessions, workshops, films, and exhibits.

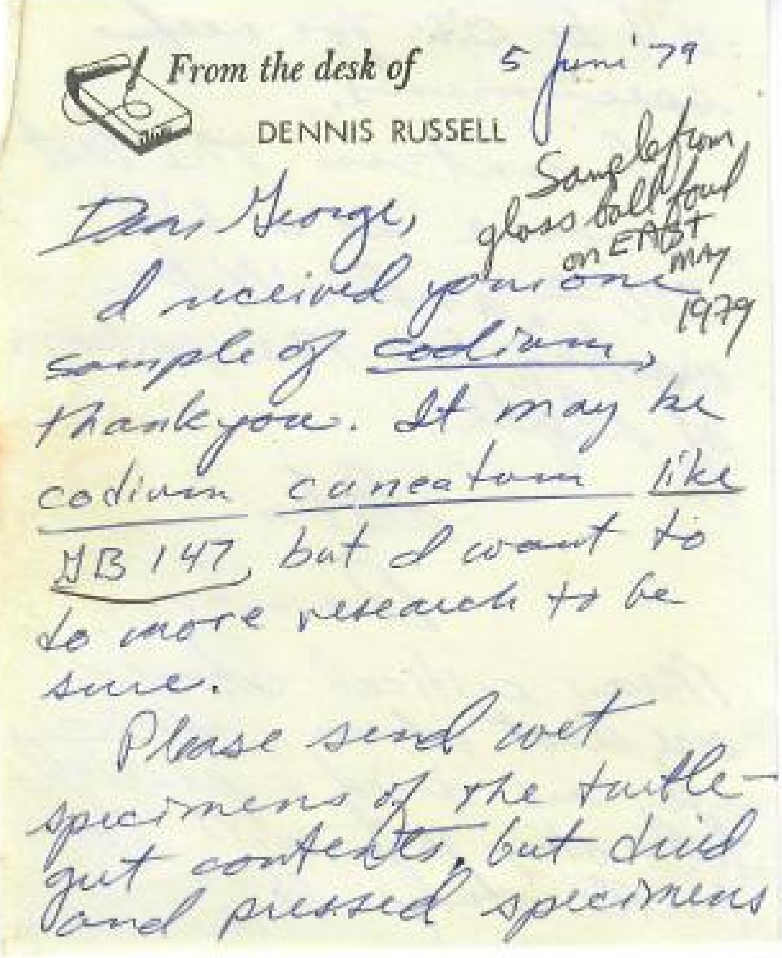
Information is available from: Restoring the Earth Conference, 1713 C Martin Luther King Jr. Way, Berkeley, CA 94709, or telephone (415) 843-2645.

We would also
like you to share
in our wedding
ceremony at 12 noon on
April 20th at Heeia
State Park in Kaneohe

<u>Melosira</u>
is a
diatom
a per Russell – Aug 1979
telephone call

<u>Rosenvingea</u> - <u>orientalis</u>,
"saimin noodles" alga -
3/11/76 Julian -
from Turtle raft


PRELIMINARY SAMPLE 2 INTESTINES
GB5
⊗Lobophora = most
Pocockiella
Liagora trace
codium —
Porolithon —
microdictyon trace
Avrainvillea ? check this
need microscope
Juvenile Tiger Recovery
FFS 6/24/77


From the desk of 5 June '79
DENNIS RUSSELL

Sample from glass ball found on EAST MAY 1979

Dear George,

I received your one sample of codium, thank you. It may be codium cuneatum like GB 147, but I want to do more research to be sure.

Please send wet specimens of the turtle-gut contents, but dried and pressed specimens

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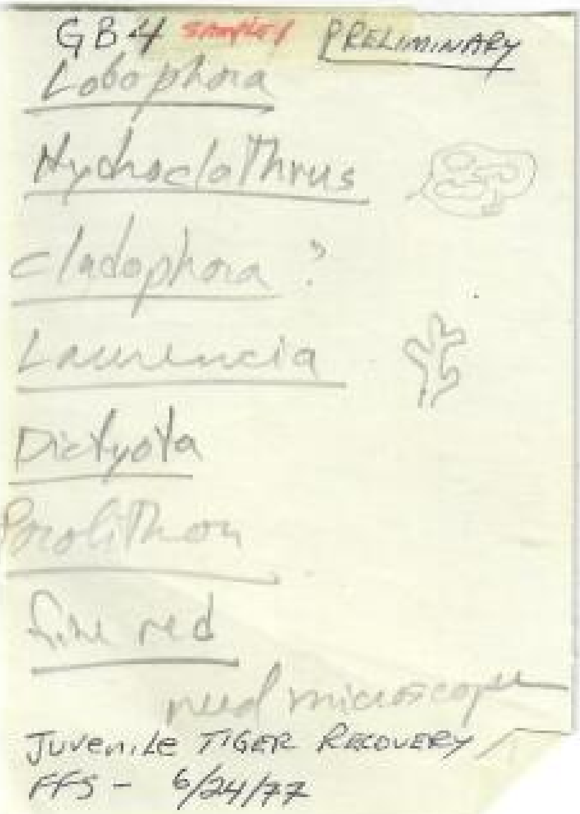
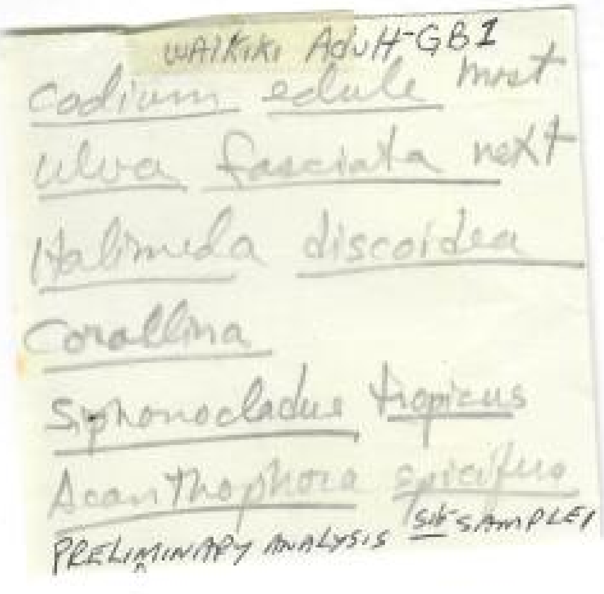

GB4 SAMPLE 1 PRELIMINARY
Lobophora
Hydroclathrus
cladophora ?
Laurencia
Dictyota
Porolithon
fine red
need microscope
JUVENILE TIGER RECOVERY
FFS - 6/24/77


WAIKIKI Adult-GB1
Codium edule most
ulva fasciata next
Halimeda discoidea
Corallina
Siphonocladus tropicus
Acanthophora spicifera
PRELIMINARY ANALYSIS SH SAMPLE 1

will be O.k. for reef specimens.

I sent in the last invoice as a bill $\$110^{\underline{00}}$, and will be waiting for the packages of algae.

aloha,

Dennis

Many critical details are lost when _codium_ etc. are dried + pressed. Let's stay wet with the turtle study. Ok.?

SAMPLE 1 GB 6 PRELIMINARY

~~Dictyosphaeria versluysii~~

Halophila

~~mottly~~

Caulerpa serrulata

S. sertularioides

Sphacelaria

Midway Mortality
7/12/77

upper stomach

JUNE 75

1 Turbinaria ornata

MAY 75
2. Caulerpa ~~sertularioides~~ taxifolia - B. Allender (green)

Laurencia sp. (pinkish)

MAY 75
3. Codium (edule?) sp. (green)

det: Linda Smith

MAY 75
3. Spyridia filamentosa (brownish)
best guess (Wulfen) Harvey
det: B. Allender

Handled by
Janet Blair
July 14, 1975

through Botan

Introduced

Acanthophora spicifera

Gelidium

Bryopsis — possible