

S C H E D U L E

4th Northwest Algal Symposium

Walla Walla College Marine Station

Rosario Beach

Anacortes, Washington

Friday,	10:00am	_	1:00pm	Arrival and check-in
March 24			1:00pm	LUNCH
Halch 24	1:30			Welcome and Orientation Susan Dixon, Convenor
	2:00	-	3:20pm	Mini-Symposium: Molecular Biology and Physiology of Algae Rose Ann Cattolico, Chair
	3:20	_	3:50pm	Break
	3:50		5:30pm	Mini-symposium, cont.
	5:30		6:00pm	Business Meeting
	6:00		7:30pm	DINNER
	8:00	-	_	Evening lecture, Russell Meints
	9:00	-		Social/Campfire
Saturday,	7:00	_	8:00am	BREAKFAST
March 25	8:20	_	10:00am	Session 1
	10:00	_	10:30am	Break
	10:30	_	12:30pm	Session 2
	12:30	_	1:50pm	LUNCH
	1:50	_	2:00pm	In Memoriam: Maurice Dube
	2:00	_	3:00pm	Posters/Demonstration
	3:00		6:00pm	Local sightseeing/Tours
	6:00	-		No host reception, Leal Dickson and Bob Waaland, Organizers
	6:30	-	8:00pm	BANQUET Skyline Beach Club
	8:00	-	9:00pm	Evening Lecture, Richard Castenholz
	9:00	-	11:30pm	Social Skyline Beach Club
Sunday,	8:00	_	9:00am	BREAKFAST
March 26	9:00	_	10:30am	Church/Free-time
narch 20	10:30	_	12:10pm	Session 3
	12:10	_	1:30pm	LUNCH
	1:30	_	3:10pm	Session 4
	3:10	_	0.105"	Check-out and Departure
	3.10			

PROGRAM

9:00 -

			Friday, March 24
10:00am	_	1:00pm	Arrival and Check-in
11:30am	-	1:00pm	LUNCH
1:30	-	2:00pm	Welcome and Orientation, Susan Dixon, Convenor.
2:00	-	5:30pm	Mini-Symposium: Molecular Biology and Physiology of Algae Rose Ann Cattolico, Moderator.
2:00	-	2:40pm	MOLECULAR DISSECTION OF THE THYLAKOID MEMBRANE. Beverly Green, University of British Columbia.
2:40	-	3:20pm	SEAWEED BIOGEOGRAPHY: SINGLE COPY DNA-DNA HYBRIDIZATION. Wytze T. Stam, Univeristy of Groningen, The Netherlands.
3:20	-	3:50pm	Break
3:50	-	4:30pm	THE OXYGENIC PHOTOTROPHS: MOLECULAR PHYLOGENETIC ANALYSIS OF A MAJOR RADIATION. Steven Giovannoni, Oregon State University.
4:30	-	5:10pm	RUBISCO EVOLUTION. Rose Ann Cattolico, University of Washington.
5:10	-	5:30pm	GENERAL DISCUSSION. Rose Ann Cattolico, Moderator.
5:30	-	6:00pm	Business Meeting
6:00	-	7:30pm	DINNER
8:00	-	9:00pm	ALGAL-VIRAL INTERACTION IN A PLANT-ANIMAL SYMBIOSIS. Russell Meints, Oregon State University.

Social/Campfire

Saturday,	March	<u>25</u>
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			Saturday, maich 25
7:00	-	8:00am	BREAKFAST
8:20	-	10:00am	Session 1, Tom Mumford, Moderator.
8:20	-	8:40am	PROTOCOL DEVELOPMENT FOR INVENTORY AND MONITORING OF PUGET SOUND NEARSHORE HABITATS: KELP AND EELGRASS. J. Anne Shaffer, Thomas F. Mumford, Jr., and Daniel J. Saul, Washington State Department of Natural Resources.
8:40	-	9:00am	PLANT STANDING STOCK AND PRODUCTIVITY ON TIDAL FLATS IN PADILLA BAY, WASHINGTON: A TEMPERATE NORTH PACIFIC ESTUARINE EMBAYMENT. Ronald M. Thom, University of Washington.
9:00	-	9:20am	TRANSIENT PHOTOSYNTHETIC RESPONSES OF NITROGEN-LIMITED PETALONIA FASCIA AND LAMINARIA SACCHARINA. Susan L. Williams and Stephen K. Herbert, University of Washington and Carnegie Institute of Washington, Stanford, CA.
9:20	-	9:40am	PROBABILITY OF HERBIVORES ENCOUNTERING AND CONSUMING THE CRUSTOSE GREEN ALGA CODIUM SETCHELLII. Cynthia D. Trowbridge, Oregon State University.
9:40	-	10:00am	STIPE DAMAGE - A CAUSE OF MORTALITY IN PTERYGOPHORA CALIFORNICA AND OTHER KELPS? Robert E. De Wreede, University of British Columbia.
10:00	-	10:30am	Break
10:30am	-	12:10pm	Session 2, Bill Rayburn, Moderator.
10:30	-	10:50am	TRANSOCEANIC DISPERSAL MECHANSIMS OF NERITIC PHYTOPLANKTON: THE ROLE OF JAPANESE BALLAST WATER IN THE DISTRIBUTION OF COASTAL AND ESTUARINE DIATOMS. Tora Johnson, Oregon Institute of Marine Biology.
10:50	-	11:10am	GRAZER-PERIPHYTON INTERACTIONS IN DEVASTED STREAMS OF MOUNT ST. HELENS. Dean M. Denicola, Oregon State University.
11:10	-	11:30am	SOIL ALGAL POPULATION AND COMMUNITY DYNAMICS FOR BUMILLERIA BORZI, CHLOROZEBRA REISIGL AND MESOTAENIUM KRAMSTAI NAGELI. John Ashley and William R. Rayburn, Washington State University.

11:30 - 11:50am

11:50am - 12:10pm

12:10 - 12:30pm

12:30 - 1:50pm

1:50 - 2:00pm

2:00 - 3:00pm

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saturday, March 25 (continued)

COMPARISONS OF NOSTOC FROM GUNNERA SPECIES OF DIFFERENT GEOGRAPHIC REGIONS. William J. Zimmerman and Thomas A. Lumpkin, Washington State University.

PRELIMINARY DATA AND PROPOSED RESEARCH CONCERNING THE QUESTION OF ECOLOGICAL DIFFERENCES BETWEEN PHASES OF ISOMORPHIC ALGAE. Frank J. Shaughnessy, University of British Columbia.

CYTOKINESIS IN <u>BANGIA</u> <u>ATROPURPUREA</u>
REPRODUCTIVE CELLS. Brian R. Oates,
University of British Columbia.

LUNCH

In Memoriam: Maurice Dube, Presented by Louis Druehl.

Posters/Demonstration

DEVELOPMENT OF PRACTICAL METHODS FOR THE COMMERCIAL CULTIVATION OF NEREOCYSTIS LUETKEANA. David Gillingham and John E. Merrill, American Sea Vegetable Co., Seattle, WA.

NORI FARMING IN ANACORTES, WASHINGTON: A POLITICAL DILEMMA. Gayle I. Hansen, Shannon Point Marine Center.

A RAPID METHOD FOR ISOLATION OF CHLOROPLAST DNA FROM THE RED ALGA, POLYSIPHONIA BOLDYII, AND HYBRIDIZATION WITH A CYANOBACTERIAL PHYCOERYTHRIN GENE. Marina K. Roell and Daniel E. Morse, Univeristy of California, Santa Barbara.

PORPHYRA AQUACULTURE IN WASHINGTON STATE. J. Robert Waaland and John E. Merrill, University of Washington and American Sea Vegetable Co., Vashon Island, WA.

"EXPERT SYSTEM": A COMPUTER GUIDE TO IDENTIFYING THE COMMON LOCAL SEAWEEDS. Tom Widdowson, University of Victoria.

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0				Saturday, March 25 (continued)
0	3:00	-	6:00pm	Local sightseeing/Tours
0	6:00	-	6:30pm	No host reception, Skyline Beach Club, Organized by Leal Dickson and Bob Waaland.
0	6:30	_	8:00pm	BANQUET, Skyline Beach Club
0	8:00	-	9:00pm	STROMATOLITES, FROM PAST TO PRESENT. Richard Castenholz, University of Oregon.
0	9:00	-	11:30pm	Social, Skyline Beach Club
0				Sunday, March 26
0	8:00	-	9:00am	BREAKFAST
0	9:00	-	10:30am	Church/Free-time
0	10:30am	_	12:10pm	Session 3, Susan Waaland, Moderator.
0	10:30	-	10:50am	ASEXUAL PROPAGATION OF <u>PORPHYRA</u> <u>THURETII</u> FROM PROTOPLASTS AND MONOSPORES. Leal G. Dickson and J. R. Waaland, University of Washington.
0	10:50	ons	11:10am	TECHNICAL AND ECONOMIC FEASIBILITY OF COMMERCIAL CULTIVATION OF NEREOCYSTIS LUETKEANA. John E. Merrill and David Gillingham, American Sea Vegetable Co., Seattle, WA.
0	11:10	-	11:30am	A STABLE VIRUS INFECTION IN A BROWN ALGA. Eric C. Henry, Stefan Lanka and Russell H. Meints, Oregon State University and Universitat Konstanz, West Germany.
0	11:30	-	11:50am	POPULATION VARIATION IN AN ANDRODIOECIOUS SPECIES OF PORPHYRA, P. PSEUDOLANCEOLATA. Sandra C. Lindstrom, University of British Columbia.
0	11:50am	. -	12:10pm	PHOTOSYNTHETIC, OXYGENIC PROKARYOTES. Ralph A. Lewin, University of California, San Diego.
0	12:10	_	1:30pm	LUNCH
0	1:30	-	3:10pm	Session 4, Louis Druehl, Moderator.
0	1:30	-	1:50pm	MOLECULAR APPROACHES TO THE TAXONOMY OF
0				THE KELP FAMILY LESSONIACEAE (PHAEOPHYTA). Gary W. Saunders and Louis D. Druehl,
0				Simon Fraser University.

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			Sunday, March 26 (continued)
1:50	-	2:10pm	RIBOSOMAL DNA (rDNA) VARIANTS IN <u>ALARIA</u> STANDS FROM VANCOUVER ISLAND, CANADA. Lawrence W. Mroz and Louis D. Druehl, Simon Frazer University.
2:10	-	2:30pm	MOLECULAR INDICATORS OF CHLOROPLAST EVOLUTION. Terrence P. Delaney and Rose Ann Cattolico, University of Washington.
2:30	-	2:50pm	CHARACTERIZATION AND REGULATION OF RUBISCO GENES IN <u>OLISTHODISCUS</u> <u>LUTEUS</u> . Linda K. Hardison and Rose Ann Cottolico, University of Washington.
2:50		3:10pm	THE RUBISCO SMALL SUBUNIT GENE IS CHLOROPLAST ENCODED IN TWO BINUCLEATE DINOFLAGELLATE SPECIES. Joby M. Chesnick and Rose Ann Cattolico, University of Washington.
3:10	-		Check-out and departure

THE ABSTRACTS WHICH FOLLOW ARE ARRANGED IN THE ORDER IN WHICH THEY APPEAR IN THE PROGRAM.

SEAWEED BIOGEOGRAPHY: SINGLE COPY NUCLEAR DNA-DNA HYBRIDIZATION STUDIES

Wytze T. Stam, P.V.M. Bot, J.L. Olsen and C. van den Hoek (Dept. of Marine Biology, University of Groningen, P.O. Box 14, NL-9750 AA Haren, THE NETHERLANDS)

The present distribution patterns of benthic marine algae can be explained (1) by ecologically mediated temperature and photoperiod responses of the species involved and (2) by historical conditions and configurations of the oceans in the geological past. Due to the almost complete lack of fossil "archives", reconstruction of distribution patterns in the past were traced by sampling the genomic "archives" of seaweeds with molecular techniques. Divergence times among disjunct populations, species and genera of Cladophora, Dictyosphaeria, Laminaria and Chorda species were estimated by single copy DNA-DNA hybridization. Dictyosphaeria and Cladophora are of probable Tethyean origin, whereas Laminaria and Chorda species are of modern origin.

PROTOCOL DEVELOPMENT FOR INVENTORY AND MONITORING OF PUGET SOUND NEARSHORE HABITATS: KELP AND EELGRASS.

J. Anne Shaffer, Thomas F. Mumford, Jr., Daniel J. Saul. Division of Aquatic Lands, Washington State Department of Natural Resources, EX-12, Olympia, Washington 98504 USA.

A standard protocol is needed to determine location and quantities of intertidal and subtidal habitats in Puget Sound and to monitor these habitats. To address the concern about current status of nearshore estuarine and marine habitats in Puget Sound, The Washington State Department of Natural Resources, working in conjunction with the Puget Sound Water Quality Authority and the Environmental Protection Agency, is developing these protocols, Habitats included in this project are intertidal salt marshes, mud flats, eelgrass beds, and kelp beds, and subtidal eelgrass and kelp beds. Methods tested include remote sensing data (Color Infrared (CIR) photography and digitally derived Multi-Spectral Scanning (MSS)). CIR and MSS information was gathered from six different test sites in Puget Sound during the summer of 1988. Photography was taken at three different scales (1"=500'; 1"=1000'; 1"=2000'), and the resulting diapositives interpreted on a zoom transfer scope. In addition, 1"=1000' diapositives from one site were also interpreted with a stereo plotter. Interpretation consisted of mapping identifiable habitats at each photo scale for each site. The final maps were digitized and the information compared to MSS data. Preliminary results indicate that intertidal Salicornia, Elvmus, Scirpus, Ulva/Enteromorpha, Zostera, and giant kelp, and subtidal Zostera and giant kelp habitats can be identified in 1"=500' and 1"=1000' diapositives; little identification is possible on 1"=2000' diapositives with the zoom transfer scope. Similar habitats were identifiable with the stereo plotter, but location of habitats was much more accurate than with the zoom transfer scope. Preliminary analysis of MSS (5 meter pixel size) data indicates that due to uncorrectable plane movement, and inability of the computer system to generalize the signatures of identified habitats to other test areas (unsupervised classification), Multi-Spectral Scanning may not be useful for nearshore habitat monitoring at this time.

PLANT STANDING STOCK AND PRODUCTIVITY ON TIDAL FLATS IN PADILLA BAY, WASHINGTON: A TEMPERATE NORTH PACIFIC ESTUARINE EMBAYMENT

Ronald M. Thom. Fisheries Research Institute, University of Washington 98195 USA.

The standing stock, production and respiration rates of sediment associated microalgae, seaweeds, eelgrass and eelgrass epiphytes were studied on a seasonal basis in Padilla Bay during 1986-1988. Biomass, net primary productivity (NPP) and respiration (R) were greatest in spring, intermediate in summer, and lowest in winter. seasonality was related to variations in irradiance, temperature and nutrient (particularly nitrate) concentrations. Annual gross primary production (GPP) for the sand-mud flats, marsh habitat and gravel patch were 149, 277 and 355 gC m⁻². The greatest proportion of total Bay annual GPP was attributed to the Zostera marina subsystem (59%), followed by the Zostera japonica subsystem (23%), the sand-mud subsystem (8%), gravel patch subsystem (0.2%) and the marsh subsystem (0.1%). Total annual NPP, R and GPP for the Bay were $7414\ \mathrm{x}10^3$, $9462\ \mathrm{x}10^3$ and $16876\ \mathrm{x}10^3\ \mathrm{kgC}$, respectively. The GPP:R ratio (1.8) indicated that the Bay system was autotrophic and exported 44% of the annual production. Along with providing rearing habitat, the Bay may provide a substantial proportion of energy to the vast fisheries resources inhabiting the waters adjacent to Padilla Bay.

TRANSIENT PHOTOSYNTHETIC RESPONSES OF NITROGEN-LIMITED PETALONIA FASCIA AND LAMINARIA SACCHARINA.

Susan L. Williams and Stephen K. Herbert. Friday Harbor Labs, Univ. of WA, Friday Harbor, WA 98250 USA and Carnegie Institution of Washington, Dept of Plant Biology, 290 Panama St, Stanford, CA 94305 USA.

Stimulation of growth of nutrient-limited marine macroalgae following increased nutrient supply presumably results from elevated rates of photosynthesis in species lacking significant carbohydrate reserves. We investigated the short-term (minutes-weeks) photosynthetic response of nutrient-depleted Petalonia fascia to an ammonium pulse to elucidate the physiological basis for the longer term growth response. Gross light-limited O2 evolution was suppressed 40% within 20 min post-pulse. Recovery of the pre-pulse rate occurred within 1-3 d. The decline (20%) of gross lightsaturated O₂ evolution was more transient. suppression was similar to that shown for certain microalgae but recovery took longer. A similar suppression occurred in Laminaria saccharina differed. The apparent slowness of short-term recovery from nitrogen-elicited photosynthetic suppression may have ecological consequences for macroalgae under conditions of patchy nutrient supply.

PROBABILITY OF HERBIVORES ENCOUNTERING AND CONSUMING THE CRUSTOSE GREEN ALGA CODIUM SETCHELLII.

Cynthia D. Trowbridge. Department of Zoology, Hatfield Marine Science Center, Oregon State University, Newport, OR 97365 USA.

Marine algae are attacked by a diverse array of invertebrate herbivores. Past studies on herbivore - algal interactions have focused on generalist consumers. conditions under which grazing by marine specialists is important are not well known. I examined the herbivores associated with the low intertidal green alga Codium The oligophagous sea slug Placida dendritica dominated the assemblage on Codium under conditions of low to moderate sand and wave disturbance. The density of Codium and the complexity and diversity of the surrounding algal community did not influence herbivore attack. Stressed Codium thalli were attacked by Placida more frequently than unstressed thalli. Furthermore, the specialist Placida created visible grazing damage significantly more frequently than did generalists. high probability of Placida encountering and consuming Codium, coupled with the consequences of Placida's gregarious feeding, indicate that the sea slug may reduce survival and growth of Codium.

STIPE DAMAGE - A CAUSE OF MORTALITY IN <u>PTERYGOPHORA</u> <u>CALIFORNICA</u> AND OTHER KELPS?

Robert E. De Wreede. Department of Botany, the University of British Columbia, Vancouver, B.C. V6T 2B1 CANADA.

The age structure of three populations of <u>Pterygophora californica</u> was analyzed using stipe rings. Data from three years were then used to construct a matrix model, which was tested for its predictive ability. In testing the sensitivity of the model it was found that, in common with previous findings, the model was most sensitive to fluctuations in mortality. Since a commonly observed source of damage (and thus presumably of mortality) to <u>Pterygophora</u> stipes is due to grazing by sea urchins the stipes of <u>Pterygophora</u> were biomechanically tested to find a critical cut length; this was calculated to be about 0.2 mm. Any cut longer than this would tend to propagate with little extra input of energy. Mortality from such damage might result if a sea urchin were to graze the stipe, followed by a storm which would impose bending strain on this stipe.

To test the ecological relevance of the calculated critical cut length, 7 mixed populations of Pterygophora californica, Eisenia arborea, and Laminaria setchellii were subjected to stipe damage imposed by a razor blade and by a triangular file. The razor blade cuts were less than critical length and greater than critical length; the triangular file was used to file a notch greater than the critical length. The razor blades mimic the biomechanical test situation, the file the type of damage done by sea urchins.

The experiment was begun in July, 1988, and is continuing; to date, the only significant mortality (comparing controls with treatment) is for the filed <u>Laminaria</u>.

TRANSOCEANIC DISPERSAL MECHANISMS OF NERITIC PHYTOPLANKTON: THE ROLE OF JAPANESE BALLAST WATER IN THE DISTRIBUTION OF COASTAL AND ESTUARINE DIATOMS.

Johnson, T. Oregon Institute of Marine Biology, University of Oregon, Charleston, Oregon 97420 USA

Cargo ships arrive almost daily in the Pacific Northwest from Japan carrying up to ten million gallons of sea water as ballast. These ocean-going vessels take on ballast in pays, harbors, coastal waters, or in the open ocean. Although the ballast water is without light for the duration of the voyage (fifteen days or more). I have documented over 100 species of living diatoms in the ballast water of Japanese, cargo vessels that arrive at the port of Coos Bay, Oregon. Experimental cultures show that many of these diatoms collected upon arrival are also able to successfully reproduce. My results therefore indicate that ballast water can be an important dispersal mechanism for coastal diatoms, as well as other phytoplankton, and is probably responsible for the invasion of exotic species in areas of intense shipping activity.

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GRAZER-PERIPHYTON INTERACTIONS IN DEVASTATED STREAMS OF MOUNT ST HELENS

Dean M. DeNicola. Department of Botany, Oregon State University, Corvallis, Oregon 97331-2902.

The effects of herbivory on periphyton assemblages were examined experimentally in two second order streams whose forests and riparian vegetation were obliterated by the May 1980 eruption of Mt. St. Helens. In a stream with forested headwaters, raised substrates which excluded grazing by tadpoles, Ascaphus truei, had periphyton assemblages composed primarily of filamentous chlorophytes, <u>Ulothrix</u> spp., and the large stalked diatom Gomphonema rhombicum. Periphyton assemblages on control substrates in this stream contained mostly nonadnate diatoms and had biomass values ten times less then assemblages on raised substrates. In a completely deforested stream lacking Ascaphus, both raised and control substrates were similar in having periphyton assemblages low in biomass which were dominated by the adnate diatom Achnanthes minutissima. Apparently, herbivorous mayflies and chironomid midges were able to graze raised as well as control substrates in this stream. In the stream lacking Ascaphus the density of these other herbivores was greater, and effects of grazing on periphyton were more severe, than in the stream dominated by Ascaphus. Primary production and herbivory are key processes in streams surrounding Mt. St Hellens because allochthonous organic inputs are minimal.

SOIL ALGAL POPULATION AND COMMUNITY DYNAMICS FOR BUMILLERIA BORZI, CHLOROZEBRA REISIGL AND MESOTAENIUM KRAMSTAI NAGELI.

<u>John Ashley</u> and William R. Rayburn. Department of Botany and Department of Microbiology, Washington State University, Pullman, Washington. 99164-4230.

Field experiments were conducted to determine the interaction between specific species and other algal groups. A split-plot designed field study was conducted using three species of soil algae, <u>Bumilleria</u>, <u>Chlorozebra</u> and <u>Mesotaenium</u>. They were inoculated singly and in combinations onto sterile and unsterile soil. Irrigated subplots were sampled monthly over one year to study the interactions between the inoculated species and the naturally occurring algae which recolonized the soil surface. Analysis of variance indicated that little interaction could be detected between treatments. Low cell densities in the inocula and the rapid change of the environment (from liquid to soil) probably accounted for the inability of two species to establish. Naturally occurring green/yellow-green algal, diatom and blue-green algal densities were measured. They increased rapidly within two months. The pattern of recolonization appeared to be the result of the preemption of space by those algae in the vicinity of the subplots. Correlation analyses indicated that cell densities of inoculated species and the naturally occurring flora were not strongly correlated to abiotic or biotic factors.

COMPARISONS OF <u>NOSTOC</u> FROM <u>GUNNERA</u> SPECIES OF DIFFERENT GEOGRAPHIC REGIONS.

<u>William J. Zimmerman</u> and Thomas A. Lumpkin. Department of Agronomy & Soils, Washington State University, Pullman, Washington 99164-6420 USA

Four cyanobionts were isolated from plants of three Gunnera species (Haloragaceae) grown in two different Swedish soils. These isolates were identified as Nostoc and compared with other strains of Nostoc isolated from Gunnera in New Zealand and the United States. Three of the four Swedish isolates, taken from plants grown in the same soil, were very similar morphologically and in their protein The five New Zealand isolates were also similar among themselves. The size of glutamine synthetase and Fenitrogenase proteins were identical in all isolates. Swedish isolates from the same soil also correlated in their DNA hybridization patterns with glnA and nifH probes, and differed from the other cyanobacteria. Cyanobacteria living in symbiosis with Gunnera may be a reflection of indigenous soil microflora. Minor cyanobionts might also exist.

PRELIMINARY DATA AND PROPOSED RESEARCH CONCERNING THE QUESTION OF ECOLOGICAL DIFFERENCES BETWEEN PHASES OF ISOMORPHIC ALGAE. Frank J. Shaughnessy, Department of Botany, University of British Columbia, Vancouver, B.C. V6T 2B1 Canada.

DeWreede and Green (in prep) used the resorcinol method to identify haploid and diploid phases of an intertidal Iridaea cordata population at Stanley Park, Yancouver and demonstrated that approximately 80% of blades are haploid during symmer months and 80% are diploid during winter months. The present study is using similar methods to sample subtidal and intertidal populations of <u>L. cordata, L. lineare, L. heterocarpa, L. cornucopiae</u> and <u>Gigartina exasperata</u> in l Vancouver and Barkley Sound. Data collected since July, 1988 indicate these L. cordata populations follow a similar temporal pattern as described by DeWreede and Green (in prep) while the relative abundances of the 1. lineare haploid phase have remained at approximately 30%. In the process of sampling the Barkley Sound populations it was noticed that a morphological continuum exists between narrow blades of wave pounded I, lineare and wide blades of more sheltered I. condata. A common ganden experiment that includes I. condata. I. lineare and I. heterocarpa will be set up this spring. If the experiment indicates that they are the same species then demographic data suggest that the diploid phase may be selected for at wave pounded sites. An experiment using outdoor tanks was initiated September, 1988 in order to compare phase survivorship and growth of I, cordata individuals. Hupotheses generated from this experiment and the above demographic data will then be experimentally examined to determine if ecological differences exist between phases of these isomorphic algae.

CYTOKINESIS IN BANGIA ATROPURPUREA REPRODUCTIVE CELLS

Brian R. Oates. Department of Botany, University of British Columbia, Vancouver, B. C. V6T 2B1 CANADA

As part of a detailed investigation into ultrastructural differentiation along the <u>Bangia</u> thallus an unusual pattern of cytokinesis was observed in the reproductive cells. Cell division was accomplished by the formation of a complete plasmalemma between the daughter cells with no cell wall furrowing, the manner that virtually all plant cytokinesis is thought to be completed. Preparation for cytokinesis is initiated in cells just distall to the meristem and is manifested by an association between the plasmalemma, chloroplast envelope and mitochondria. Additionally, chloroplasts, which occupy the majority of the cell volume, appear to be 'sliced', as opposed to pinching into two chloroplasts. The plasmalemma may originate from the endoplasmic reticulum. Cell wall material is added evenly along the entire plasmalemma. This cell division phenomenon was observed in both female and male individuals.

-P O S T E R-

DEVELOPMENT OF PRACTICAL METHODS FOR THE COMMERCIAL CULTIVATION OF NEREOCYSTIS LUETKEANA.

<u>David Gillingham</u> and John E. Merrill. American Sea Vegetable Co., Box 31104, Seattle, WA 98103.

We are developing practical methods for the commercial cultivation of "Bull-Whip Kelp", Nereocystis luetkeana. report summarizes our results for gametophyte culture and sporophyte ("seed") production. We have strongly emphasized identification of methods that can be achieved outside of a "laboratory" setting and that are relatively inexpensive. Sori are selected from mature plants based on their apparent readiness to release zoospores. These are prepared for release by careful cleaning and surface disinfection with iodine solution. Release is induced by overnight storage between moist towels at Spore concentration is adjusted to 5 % 10³ per ml before inoculation. Substrate consists of #18 white nylon "seine twine" spooled on sections of 2 inch PVC pipe. Reduction in contamination of cultures is greatly enhanced by filtration of the culture seawater through inexpensive cartridge filters. Seawater is enriched with a commercially produced Guilliard's "f" enrichment. Young sporophytes develop in 2.5-3 weeks under light levels of ca. 100 uE m $^{-2}$ s $^{-1}$ and water temperature of 10C. Resulting twines are used to "seed" long-line ropes for cultivation of adult sporophytes.

NORI FARMING IN ANACORTES, WASHINGTON: A POLITICAL DILEMMA

Gayle I. Hansen. The Shannon Point Marine Center, 1900 Shannon Point Road, Anacortes, Washington 98221 USA.

The area around Anacortes, Washington, with its islands, semi-protected bays and inlets and its nutrient-rich, well-mixed waters seems an ideal place for sea vegetable farming. The off-shore cultivation of the most nutritious sea vegetable, nori or Porphyra, has been studied intensively by State researchers and by the phycologists at the University of Washington. With an approval by the govenor, the future of nori farming in Washington should be bright. Anacortes could be looking forward to a non-polluting, CO2-absorbing industry that would employ its unskilled and provide revenue through lease fees for its parks and schools.

However, when proposals to begin nori farming were submitted, there was a tremendous public outcry, particularly from the local shore-line owners and fishermen. They were afraid that nori farms would glut their waterways, destroy their views, lower their property values, and take away their crabbing, fishing, and boating rights. Worse, they felt the plants might rot on the beaches and the nets trap and kill seals and birds -- and even erode away their beaches. The opposition was strong, belligerant, well-organized -- and vocal. They formed "environmental groups", held demonstrations, sold bumper stickers, and wrote in numbers against the farms to the newspapers, local legislators, and to the county planning committee.

Even now, the battle rages on -- and the permitting decisions have yet to be made. If you were an elected official, or appointed by one, in Anacortes, -- what would be your decision?

This poster will consist of a collage of newspaper articles and letters on the proposal for nori farming in Anacortes, Washington.

-P O S T E R-

A RAPID METHOD FOR ISOLATION OF CHLOROPLAST DNA FROM THE RED ALGA, <u>POLYSIPHONIA</u> BOLDYII, AND HYBRIDIZATION WITH A CYANOBACTERIAL PHYCOERYTHRIN GENE

Marina K. Roell and Daniel E. Morse. Marine Science Institute and Department of Biological Sciences, University of California, Santa Barbara, CA 93106

Current methods for the isolation of chloroplast DNA from macrophytic red algae make use of the DNA-binding dye, bisbenzimide H (Hoecsht 33,258), which binds differentially to DNA's with different A+T:G+C ratios, altering their densities to allow separation by isopycnic ultracentrifugation. When it is not feasible to isolate chloroplasts, time-consuming multi-step procedures are used to carefully isolate total nucleic acids with minimum shearing of nuclear DNA. A more rapid and simple procedure, based on the method of Chirgwin et al (Biochemistry, 18:5294, 1979), has been shown to be effective for isolating nucleic acids from cultured Polysiphonia boldyii. In this method, nucleases are inactivated immediately and manipulations of the DNA are minimized. The potent protein denaturant guanidine isothiocyanate, with 2-mercaptoethanol and sarcosyl is used to quickly and gently lyse the cells and organelles of algae that have been ground in liquid nitrogen. The cleared lysate is layered on top of a dense CsCl solution and centrifuged at 174,000 x g for 21 hours. The DNA bands in the CsCl solution, while the RNA pellets, and the proteins remain in the lysate layer. To separate the chloroplast, nuclear and mitochondrial DNAs, the total DNA is centrifuged in a CsCl solution with bisbenzimide H for 40 hours at 100,000 x g. After this spin, three fluorescent bands are visible, and can be collected separately. The band containing chloroplast DNA has been identified in previous preparations, as follows. Aliquots of the DNA fractions were digested with a restriction enzyme, and the fragments were separated by agarose gel electrophoresis, transferred to a nylon membrane, and incubated with a phycoerythrin gene probe from Pseudoanabaena (a gift of Donald Bryant). Hybridization with this gene, believed to be chloroplast-encoded in red algae, was detected only in the lane containing DNA from the intermediate density fraction. To ensure that the signal was specific, aliquots from this DNA fraction were digested with several restriction enzymes, blotted, and probed under conditions of low and moderate stringency (55C and 65C). No hybridization was detected at moderate stringency, while at low stringency, different bands were seen in each digest, as would be expected for specific hybridization with a heterologous probe.

-P O S T E R-PORPHYRA AQUACULTURE IN WASHINGTON STATE

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Commercial aquaculture of Porphyra for the edible product nori and for phycobiliproteins was initiated in Washington State in 1987 after nearly a decade of research, planning and cooperation by government (Washington Department of Natural Resources, Washington Sea Grant), university (University of Washington) and private industry (American Sea Vegetable Co. and New Channel Nori Farms). Two active cultivation sites are in operation, a 10 hectare, 300 net-unit site in Tramp Harbor, Vashon Island in Puget Sound near Seattle and several smaller sites totaling 250 net-units adjacent to Johns Island in the San Juan Archipelago. Conchocelis culture, net seeding, nursery culture, production culture, harvesting and nori processing have all been implemented. In addition research on reproduction and strain selection and improvement has been conducted at these aquaculture sites.

ASEXUAL PROPAGATION OF *PORPHYRA THURETII* FROM PROTOPLASTS AND MONOSPORES

Leal G. Dickson and J.R. Waaland

Control of reproduction and the selection/development of new strains of *Porphyra* is important in a Washington State nori industry. Production of monospores provides for a rapid reseeding of nets; however only one Pacific Northwest species has been shown to produce these asexual spores. We have shown for the first time that *P. thuretii* produces copious quantities of monospores which we have used to seed nori netting. Additionally, we have successfully produced protoplasts from *P. thuretii*; these rapidly regenerate into new blades. These protoplasts will be useful in parasexual fusion of potentially important nori species. Completion of the life cycle in the laboratory has been accomplished for monospores and protoplasts.

TECHNICAL AND ECONOMIC FEASIBILITY OF COMMERCIAL CULTIVATION OF NEREOCYSTIS LUETKEANA.

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The American Sea Vegetable Co. has initiated a research and development project with the goal of determining the technical and economic feasibility of cultivating the native Pacific Northwest "bull-whip" kelp, Nereocystis luetkeana. Impetus for this study derives from growing interest in this species for a number of commercial purposes, increasing harvest pressures on natural beds, and concern among knowledgeable professionals for both the ecological importance of these beds and the potential for devastation through over-use. We are emphasizing a modified "long-line" style of cultivation as practiced for such Asian crops as "Kombu" and "Wakame". We have gathered data on such factors as optimum depth of cultivation, optimum inoculation densities, suitable substrate materials, handling methods, growth rates, harvest yields, etc. We have been able to deploy new plantings throughout the Summer, Fall, and Winter seasons with excellent development, although growth rates may vary. A more pronounced effect of depth on growth rates during winter months suggests a strong influence of total irradiance on growth. Preliminary harvest estimates are 8-16 kg per meter of long line with 3 or more crops possible per year, depending on size or age of desired product. We are currently implementing a 1-acre expanded test planting that will hopefully provide additional detailed information on yields per m² and costs of production.

A STABLE VIRUS INFECTION IN A BROWN ALGA.

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We have established laboratory cultures of a brown alga (a uniseriate, branched, filamentous form, probably an undescribed species of *Feldmannia* that maintains a stable virus infection. Vegetative cells appear normal but formation of spores in the unilocular (meiotic) sporangiais prevented. Transmission electron microscopy reveals that at "maturity" the sporangia contain no intact nuclei, only a few remnants of mitochondria and chloroplasts, and numerous polyhedral virus-like particles ca. 150 nm in diameter with a dark, spherical core. No plurilocular zoidangia are formed. Very rarely, morphologically normal unilocular sporangia are formed that release apparently normal zoospores that developinto "gametophyte" filaments bearing plurilocular (mitotic) zoidangia only. The zoids they release do not act as gametes, but instead germinate to repeat the same "gametophyte" stage. No trace of virus particles has been found in vegetative cells of the sporophyte or any cells of the gametophyte. Apparently the viral genome is integrated into the genome of the alga, and the virus only replicates in the unilocular sporangia. Viral DNA can be readily prepared without extensive viral purification. Agarose gel electrophoresis of restriction endonuclease digests of the viral genome reveals double-stranded DNA of about 150 kilobase pairs. Methylation of some of the bases is demonstrated. A viral genomic library has been prepared. This is the first study of the molecular biology of a virus of a eukaryotic marine alga.

POPULATION VARIATION IN AN ANDRODIOECIOUS SPECIES OF PORPHYRA, P. PSEUDOLANCEOLATA.

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Porphyra pseudolanceolata is a linear-lanceolate species of Porphyra that occurs on mid to high intertidal rocky shores through the winter months and into the spring. Thalli are usually unisexual but can be split vertically, horizontally, or in a wedge-shape into female and male sectors. Five populations (two on San Juan Island, one along southern Vancouver Island, one in the Strait of Georgia, and one in northern B. C.) were sampled, and extracts from more than 200 thalli were run on horizontal starch gels. Gels were stained for 13 enzymes, two of which had two isozymes. Banding patterns were also recorded for the phycobiliproteins, phycocyanin and phycoerythrin. The banding patterns revealed a major discontinuity in the genetic structure of P. pseudolanceolata between the Strait of Georgia and the remaining populations.

Photosynthetic, Oxygenic Prokaryotes. Ralph A. Lewin, Scripps Institution of Oceanography, University of California, San Diego, La Jolla, CA 92093 U.S.A.

Prochloron didemni was originally described as a zoochlorella, later as a cyanophyte (Synechococcus didemni). When it was found to contain chlorophyll b as well as a, and to lack bilin pigments, it was assigned to a new subclass, the Prochlorophyta. Nevertheless, in most features it closely resembles a blue-green alga, specifically Synechococcus raspaigallae. Both species have round cells about 10-12 um in diameter, with several peripheral (not central) DNA bodies, and both occur as extracellular symbionts of didemnid ascidians or sponges. Did Prochloron arise from such a cyanophyte ancestor, perhaps at the time when didemnids were evolving, or could it have a much older history, and perhaps be related to ancestral chloroplasts? Now another prokaryote has been discovered, wholly dissimilar in form but having somewhat similar pigments (two chlorophylls and no phycobilins). It has been called *Prochlorothrix* and has been assigned, like *Prochloron*, to the prochlorophytes. Nevertheless, in form it is almost indistinguishable from a thin Oscillatoria like some with which it occurs in certain freshwater lakes. This brings into question the whole classification of algae, where subclasses have been distinguished primarily on the basis of pigmentation. For eukaryotic algal classes we have many other correlated characters, but among the prokaryotes few features are of fundamental taxonomic value. Should we associate all the latter under the name "Prokaryophyta", or even, for biopolitical expediency, "Phycobacteria"? New data from molecular biology - amino-acid and nucleotide sequences - lead to equivocal conclusions about their respective phylogenies. Obviously we need more studies of such microbes.

MOLECULAR APPROACHES TO THE TAXONOMY OF THE KELP FAMILY LESSONIACEAE (PHAEOPHYTA).

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To better understand taxonomic and phylogenetic affinities within the Laminariales we have initiated molecular investigations on a variety of kelp species. The techniques required for restriction enzyme mapping of the ribosomal cistron are described. Preliminary data for the Lessoniaceae are presented with particular emphasis placed on Lessoniopsis littoralis (Farlow & Setchell) Reinke. This alga is of interest in that it questions the traditional taxonomic system of the kelp.

RIBOSOMAL DNA (rDNA) VARIANTS IN <u>ALARIA</u> STANDS FROM VANCOUVER ISLAND, CANADA.

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Alaria taxonomy is confusing due to the combination of environmentally induced morphological variation, and the genetic variability of the genus. A. marginata, A. tenuifolia and A. nana, are usually morphologically distinct around Vancouver Island. Some stands, however, exhibit intermediate A. marginata x A. tenuifolia, and A. marginata x A. nana hybrid-like morphologies, which suggests interbreeding. To separate genetic and environmental influences on morphology, recombinant DNA methods were used to investigate genetic relationships between Alaria stands. Individuals from various stands were examined for genetic differences in their ribosomal RNA genes (rDNA) using the small subunit rDNA as a probe. Restriction fragment length difference (RFLD) analysis has revealed that Alaria rDNA has differentiated into four major variants with length and restriction site polymorphisms. Most individuals contain copies of a single species-specific variant. Others, containing pairs of these variants, suggest that hybridization has occurred in some stands.

MOLECULAR INDICATORS OF CHLOROPLAST EVOLUTION.

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Molecular sequence data have provided the strongest evidence for an endosymbiotic origin of chloroplasts from prokaryotes. However, these data are derived almost entirely from chlorophyll b containing taxa. Comparable data are required from chromophytic (chl c) and rhodophytic algal taxa before questions of plastid origin and evolution can be addressed. We have determined the nucleotide sequence of 16S rRNA genes from plastids of Olisthodiscus luteus (chl c), and the rhodophytes Griffithsia pacifica and Porphyra yezoensis. These data are discussed in the context of a mono- vs polyphyletic plastid origin. We have also determined the sequence of the Rubisco small subunit gene in Olisthodiscus (B. Boczar, TPD and RAC, submitted). This gene is more similar to that found in a purple bacterium than to the cyanobacterial counterpart. The implications of this finding are presented. An integrated phylogeny of 16S rRNA and Rubisco genes is discussed.

CHARACTERIZATION AND REGULATION OF RUBISCO GENES

IN OLISTHODISCUS LUTEUS

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In higher plants and green algae, the genes for the large (rbcL) and small (rbcS) subunit polypeptides of the photosynthetic enzyme Rubisco are encoded in the chloroplast and nuclear genomes, respectively. The golden-brown alga <u>Olisthodiscus luteus</u>, however, is unique in that both genes are found in the chloroplast. This chloroplast encoding of both genes, as well as the nucleotide sequence similarity of rbcS to that of a chemoautotrophic bacterium, have implications regarding the evolutionary origin of the algal chloroplast.

The large subunit polypeptide of Rubisco contains the binding site, and the small subunit is thought to be involved in holoenzyme regulation. Thus the consequences of the chloroplast encoding of rbcS are profound when one considers the coordination and regulation of polypeptide synthesis and assembly. The two genes are separated by 54 nucleotides, with rbcL 5' to rbcS. RNA studies are underway to determine if the two genes are co-transcribed. Progress is being made in the sequencing of the 5' promoter region. Data of the structure of these photosynthetic genes will be coupled to their expression in various light regimes.

THE RUBISCO SMALL SUBUNIT GENE IS CHLOROPLAST ENCODED IN TWO BINUCLEATE DINOFLAGELLATE SPECIES.

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Peridinium balticum and Peridinium foliaceum are binucleate dinoflagellate species hypothesized to be evolutionary intermediates in chloroplast acquisition events in the Pyrrhophyta. During the past year research concerning chloroplast DNA analyses of these two organisms (each containing a chlorophyll c photosynthetic endosymbiont) has demonstrated two cpDNA genomes (110 kb and 180 kb) for P. balticum and a single (110 kb) cp genome for P. foliaceum. Although restriction fragment length polymorphisms were evident among these three genomes, a 32-P labeled probe made to ribulose 1,6 bisphosphate carboxylase small subunit gene (rbcS) from the goldenbrown alga Olisthodiscus luteus hybridized to a 2.8 kb fragment of Hind III restricted cpDNA in each case. This probe (rbcS from O. luteus) was recently shown to have high sequence homology to the rbcS gene found in the bacterium Alcaligenes eutrophus, a member of the purple photosynthetic bacterial group, implicating an alternative origin for the evolution of this gene in chromophytic plants. This research supports the theory that a common prokaryotic ancestor occurred for chromophytic chloroplast DNA and suggests that possible common domains exist between the cp genomes of P. balticum and P. foliaceum.

IN MEMORIAM: MAURICE A. DUBE (1927-1989)



On the evening of February 2, 1989, Maurice Dube was killed by a bicyclist while walking near his home in Bellingham, Washington. Knocked to the ground, he hit his head and died instantly.

As phycologists, most of us knew Maurice for his discovery of an isomorphic life history in Monostroma fuscum (now Ulvaria obscura) and for his ultrastructure studies of the Prasinophyte Halophaera. As a friend and research associate of Maurice, I knew him for his gentle manner and his subtle humor. He was clever at catching the interest of students, and his courses were

always popular. He was a wonderful natural historian and loved both taking his students into the field to collect algae and recounting the stories about early northwest phycologists.

Maurice had a varied background. He earned his B.A. degree at Washington State University in 1950 majoring in Mechanical Engineering. After teaching high school for a year in Oregon, he joined the Army and worked in guided-missile research in Alabama. Then, with a change of interests, he began graduate school at Oregon State University and finished his M.S. degree in General Science in 1958 and his Ph.D. in Botany in 1963 under the direction of Harry K. Phinney. He then received an offer from Robert F. Scagel to do a postdoc at the University of British Columbia. From there, he was recruited by Western Washington University to begin his 25-year term as their phycologist.

Maurice was a multi-talented man. From his engineering background, he was extremely adept at building gadgets for use in lab. He designed anything from camera lucidas, to densitometers, to giant plant presses. Hardly a machine came into his hands that he didn't modify so that it exactly fit his needs. He was a talented pen-and-ink and watercolor artist and an equally good photographer. He developed the Biological Illustration course at Western. He loved music and was an accomplished accordian player. He and his wife, Judy, who played the bass fiddle, frequently performed at local coffee houses, dances, and weddings.

Maurice's interests were many: engineering, biology, art, and music. In addition, he was deeply interested in theology. Shortly before he died, he had prepared and given an invited general lecture for the University entitled "Science and the Christian Faith".

He is survived by his wife, Judy Dube, and stepson, Kevin Shelley, of 304 Morey Avenue, Bellingham, Wa. 98225

Maurice was a close friend to many of us, and we will miss him greatly.

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