# 14th NorthWest Algal Symposium

**NWAS 2000** 

Programme and Abstracts

University of British Columbia Vancouver, British Columbia Canada May 12-14, 2000

#### 14th NORTHWEST ALGAL SYMPOSIUM PROGRAM

#### Friday Afternoon, May 12

15:00 Registration: Begins in 2449 Biological Sciences Building

15:00 Poster Set Up: Room 2449, any time. Posters must be removed by 14:00 Sunday May 12.

#### Friday Evening, May 12

12:30-14:00

Lunch

19:00 Plenary Speaker: Room 2000. Dr. Paul J. Harrison, Depts. of Botany and Earth & Ocean Sciences, University of British Columbia. "IS THERE A CONNECTION BETWEEN DUST AND PHYTOPLANKTON? HOW IRON LIMITATION INFLUENCES PRIMARY PRODUCTIVITY IN THE NE PACIFIC"

20:00 - 22:00 Social. Room 2449

#### Saturday Morning, May 13 08:00 Registration resumes in Room 2449

08:30	Welcome
	Contributed Papers - Session Chair: Tim Nelson
08:45	Terrie Klinger. Friday Harbor Laboratories, Univ. of Washington, Friday Harbor, WA INCORPORATING SEAWEEDS AND SEAGRASSES IN THE DESIGN OF MARINE PROTECTED AREA NETWORKS
09:00	<u>Frank Shaughnessy</u> . Dept. of Biological Sciences, Humboldt State University, Arcata, CA A PRELIMINARY ANAYSIS OF ALGAL AND INVERTEBRATE RECOVERY FOLLOWING AN OIL SPILL IN HUMBOLDT BAY, CALIFORNIA
09:15	Gisela Fritz <sup>1</sup> , Timothy Mulligan <sup>2</sup> , & Frank Shaughnessy <sup>1</sup> Dept. of Biological Sciences & <sup>2</sup> Fisheries, Humboldt State University, Arcata, CA THE FLORAL RECOVERY OF A RESTORED COASTAL WETLAND: KUNZ MARSH, SOUTH SLOUGH, COOS BAY, OR
09:30	Paula C. Caron. Dept. of Biology, University of Victoria, Victoria, BC RESERVOIR DRAWDOWN: IMPLICATIONS FOR LITTORAL BENTHIC COMMUNITIES AND DRINKING WATER QUALITY
09:45	Grant G. Mitman. Dept. of Biological Sciences-Montana Tech of The University of Montana, Butte, Montana ALGAL BIOREMEDIATION OF THE BERKELEY PIT LAKE
10:00-11:00	C/T Break and Poster Viewing I
	Contributed Papers - Session Chair: Tom Mumford
11:00	Robert E. DeWreede. Dept. of Botany, Univ. of British Columbia, Vancouver, B.C. MAINTAINING SELF: AN OVERVIEW OF PHYSICAL AND BIOLOOGICAL INTERACTIONS IN THE HEDOPHYLLUM ZONE
11:15	Louis Druehl. Simon Fraser University at the Bamfield Marine Station, Bamfield, B.C. HYBRIDIZATIONS TEST KELP EVOLUTIONARY RELATIONSHIPS
11:30	Allison E. Henderson & Louis Druehl. Simon Fraser Univ., Bamfield Marine Station, Bamfield, B.C. KELP HYBRIDIZATION STUDIES
11:45	Hillary S. Hayden & J. Robert Waaland. Dept. of Botany, University of Washington, Seattle, WA WHO'S WHO IN THE ULVACEAE?
12:00	Louis Hanic. Dept. of Botany, University of British Columbia, Vancouver, B. C. UROSPORA (CHLOROPHYTA) ON THE PACIFIC COAST OF NORTH AMERICA
12:15	Andrea Sussmann & Robert DeWreede. Dept. of Botany, Univ. of British Columbia, Vancouver, B.C. FIELD STUDIES OF THE LIFE HISTORY OF ACROSIPHONIA (CODIOLALES, CHLOROPHYTA) IN SOUTHERN BRITISH COLUMBIA
12 20 11 00	

#### Saturday Afternoon, May 13

#### Contributed Papers - Session Chair: Rob Fitch

14:00 Craig D. Sandgren. Dept. of Biological Sciences, University of Wisconsin-Milwaukee, Milwaukee, WI A HEDGE AGAINST HERBIVORES? THE EFFECTS OF ALGAL COLONY SIZE AND THE PRESENCE OF 'SPINY" SILICEOUS CELL COVERINGS ON GRAZING RATES BY ZOOPLANKTON ON PLANKTIC CHRYSOPHYTE FLAGELLATES

14:15 <u>Dean M. Jacobson</u>, Dept. of Biology, Whitworth College, Spokane, WA
PHAGOTROPHY IN GONYAULACOID DINOFLAGELLATES AND A HYPOTHESIS FOR THEIR
ADAPTIVE RADIATION

14:30 Robert Edward Lee<sup>1</sup> & Paul Kugrens<sup>2</sup>. <sup>1</sup>Dept. of Anatomy & Neurobiology, College of Veterinary Medicine, Colorado State Univ., Fort Collins, CO 80523 & <sup>2</sup>Dept. of Biology, Colorado State Univ., Fort Collins, CO ANCIENT ATMOSPHERIC CO<sub>2</sub> AND THE TIMING OF EVOLUTION OF SECONDARY ENDOSYMBIOSES

14:45 BUSINESS MEETING

15:00-18:00 Informal tours, UBC and Vancouver

#### Saturday Evening, May 13

18:00-20:00 Banquet and related activities (Ponderosa)

**Banquet Speaker**. Dr. Paul Kugrens, Colorado State University, Fort Collins, CO. "PHYCOS, PHISH AND PHYCOLOGISTS I HAVE KNOWN"

#### Sunday Morning, May 14

#### Contributed Papers - Session Chair: Brian Oates

09:00 Sean M. Brigden<sup>1</sup>, Martin Baur<sup>2</sup>, Curtis A. Suttle<sup>1,2,3</sup>. <sup>1</sup>Depts. Of Botany, <sup>2</sup>Microbiology & Immunology, & <sup>3</sup>Earth & Ocean Sciences, Univ. of British Columbia, Vancouver, B.C.

DISTRIBUTION AND ABUNDANCE OF CYANOBACTERIA AND LYTIC AGENTS INFECTIOUS TO SYNECHOCOCCUS SPP. IN THE STRAIT OF GEORGIA

09:15 Vera Tai<sup>1</sup>, Janice E. Lawrence<sup>2</sup>, Amy M. Chan<sup>2</sup> & Curtis A. Suttle<sup>1,2,3</sup>. Depts. of <sup>1</sup>Botany, <sup>2</sup>Earth & Ocean Sciences & <sup>3</sup>Microbiology & Immunology, University of British Columbia, Vancouver, B.C. ISOLATION AND CHARACTERIZATION OF VIRUSES INFECTING HETEROSIGMA AKASHIWO (RAPHIDOPHYCEAE)

09:30 <u>Ioanna Visviki</u>. Dept. of Biology, College of Mount Saint Vincent/Manhattan College, 6301 Riverdale Avenue, Riverdale, NY
THE UNUSUAL CHONDRIOME OF *CHLAMYDOMONAS ACIDOPHILA* 

O9:45

Gregory L. Rorrer<sup>1</sup>, Mary P, Tucker<sup>1</sup>, Jason Polzin<sup>1</sup>, Sanjiv Maliakal<sup>2</sup>, & Donald P. Cheney<sup>2</sup>. <sup>1</sup>Dept. of Chemical Engineering, Oregon State Univ., Corvallis, OR, & <sup>2</sup>The Marine Science Center, Northeastern Univ., Nahant, MA

HALOGENATED MONOTERPENE PRODUCTION IN MICROPLANTLET SUSPENSION CULTURES OF THE TROPICAL RED ALGA OCHTODES SECUNDIRAMEA

10:00-10:30 C/T Break

	Contributed Papers - Session Chair: Brian Oates
10:45	Elena Filek, Zhaoduo Zhang & B. R. Green. Dept. of Botany, Univ. of British Columbia, Vancouver, B.C. DINOFLAGELLATE CHLOROPLAST GENOME: HOW MANY MINICIRCLES ARE THERE?
11:00	Kimberly Burns, Brian R. Oates & Kathleen M. Cole. Dept. of Botany, Univ. of British Columbia, Van-
	couver, B. C. FURTHER EXAMINATIONS INTO THE ULTRASTRUCTURE OF GLAUCOCYSTIS NOSTO-
	CHINEARUM: A COMPARISON OF CHEMICAL AND CRYO-FIXATION
11:15	Poster Viewing II
11:30	
11:45	
12:00	Lunch
12:15	
12:30-14:00	Take down posters and have a safe trip home

#### **Contributed Posters**

Sophie Boizard. Department of Botany, University of British Columbia, Vancouver, Canada. KELP HOLDFAST ANCHORAGE MECHANICS: FORM AND FUNCTION

Brooke Budnick, Suzanne Strom, Kelley Bright and Erin Macri. Shannon Point Marine Center and Biology Dept., Western Washington University Bellingham, WA. EFFECTS OF AMMONIUM ADDITION AND ENHANCED PAR AND UV-B IRRADIANCE ON PIGMENTS OF ZOOXANTHELLAE FROM THE CORAL MONTASTRAEA FAVEOLATA AT ELEVATED TEMPERATURE

Heather I. Decious 1 & Kathy Van Alstyne 2. 1 Department of Biology, Western Washington University, Bellingham, WA, USA & <sup>2</sup> Shannon Point Marine Center, Western Washington University, Anacortes, WA, USA. FEEDING PREFERENCE OF LITTORINA SITKANA ON BENTHIC DIATOMS, FUCUS GARDNERI EMBRYOS AND ULVA FENESTRATA SPORELINGS

Gretchen Frankenstein. Puget Sound Water Quality Action Team, Olympia, WA, USA ULVOID BLOOMS IN PUGET SOUND--CAUSE FOR CONCERN?

Cindy M. Frederickson, Steven M. Short & Curtis A. Suttle. Departments of Earth and Ocean Sciences, Microbiology and Immunology, and Botany, University of British Columbia, Vancouver, Canada. THE GENETIC DIVERSITY AND ABUNDANCE OF VIRUSES THAT INFECT CYANOBACTERIA IN THE STRAIT OF GEORGIA, BRITISH COLUMBIA, CANADA

Yao-ming Huang & Gregory L. Rorrer. Department of Chemical Engineering, Oregon State University, Corvallis, Oregon, USA. PHOTOBIOREACTOR CULTIVATION OF AGARDHIELLA SUBULATA REGENERATED MICROPLANTLET

Yao-ming Huang & Gregory L. Rorrer. Department of Chemical Engineering, Oregon State University, Corvallis, Oregon, USA EFFECTS OF PH, PHOTOPERIOD AND TEMPERATURE ON THE CUL-TIVATION OF AGARDHIELLA SUBULATA REGENERATED MICROPLANTLET

Ken-ichiro Ishida, Thomas Cavalier-Smith, & Beverley R. Green. Department of Botany, University of British Columbia, Vancouver, B.C., CANADA. IN VITRO PROTEIN IMPORT INTO CHLORO-PLASTS ISOLATED FROM A HETEROKONT ALGA: HETEROSIGMA AKASHIWO

Dean M. Jacobson, Department of Biology, Whitworth College, Spokane Washington, USA.. ULTRA-STRUCTURAL DEMONSTRATION OF INGESTION OF PHAESOME SYMBIONTS BY ORNI-THOCERCUS (DINOPHYCEAE)

Kari Jones, Amorah Lee, and Timothy Nelson. Department of Biology, Seattle Pacific University, Seattle, Washington, USA. A CHARACTERIZATION OF ULVOID BLOOMS AND THEIR ABIOTIC ENVIRONMENT IN THE SAN JUAN ARCHIPELAGO.

Minako Kaneda1, Ken-ichiro Ishida2 & Yoshiaki Hara1. 1Department of Biology, Yamagata University, Yamagata, Japan, 2Department of Botany, University of British Columbia, Vancouver, B.C., Canada. A CHLORARACHNIOPHYTE ISOLATE WITH A MULTINUCLEAR CELL STAGE IN THE LIFE CYCLE

Amorah Lee, Kari Jones, Timothy Nelson. Department of Biology, Seattle Pacific University, Seattle, Washington, USA. ENVIRONMENTAL DETERMINANTS OF ULVOID ALGAL SPECIES COMPOSITION

Sandra Lindstrom<sup>1</sup> & Gayle. Hansen<sup>2</sup>. <sup>1</sup>Department of Botany, University of British Columbia, Vancouver, Canada & <sup>2</sup>Hatfield Marine Science Center, Oregon State University, Newport, Oregon, USA. A FLORA OF THE BENTHIC MARINE ALGAE OF ALASKA--STATUS AND FUTURE PROSPECTS

A. Mazumder, B. K. Basu, Y. Liang, P.C. Caron, <u>J-M. Davies</u>, and W. Nowlin. University of Victoria, Department of Biology, Victoria BC, Canada. THE ENVIRONMENTAL MANAGEMENT OF DRINKING WATER ECOSYSTEMS: AN ECOSYSTEM AND WATERSHED APPROACH

Grant G. Mitman. Department of Biological Sciences, Montana Tech of the University of Montana, Butte, MT 59701. ALGAL BIOREMEDIATION OF THE BERKELEY PIT LAKE

Maurizio Murru\_and <u>Craig D. Sandgren</u>. Department of Biological Sciences, University of Wisconsin - Milwaukee, Wisconsin, USA. PHOTOSYNTHETIC UTILIZATION OF HCO<sub>3</sub><sup>-</sup> IN 38 SPECIES OF PUGET SOUND RED ALGAE: AN ECOLOGICAL AND TAXONOMIC COMPARISON.

Margaret Nordquist, Gisèle Muller-Parker, and Erin Macri. Shannon Point Marine Center and Biology Dept., Western Washington University, Bellingham, WA 98225-9081. EFFECTS OF AMMONIUM ADDITION AND ENHANCED PAR AND UV-B IRRADIANCE ON THE PRODUCTIVITY OF INHOST AND EXPELLED ZOOXANTHELLAE FROM THE CORAL MONTASTRAEA FAVEOLATA.

<u>Janise Palmer</u>, Erin Macri, Gisèle Muller-Parker, Suzanne Strom. Shannon Point Marine Center and Biology Dept., Western Washington University, Bellingham, WA 98225-9081. ROLE OF NITROGEN AVAILABILITY IN THE RESPONSE OF THE CORAL *MONTASTRAEA FAVEOLATA* TO ENVIRONMENTAL STRESS

<u>Gregory.L. Rorrer</u> & Ronald K. Mullikin. Department of Chemical Engineering, Oregon State University, Corvallis, Oregon 97331,USA. MODELING AND SIMULATION OF A TUBULAR RECYCLE PHOTOBIOREACTOR FOR MACROALGAL CELL SUSPENSION CULTURES

Mike Thorpe, John Hardy, and Jonathan Frostad. Shannon Point Marine Center and Huxley College, Western Washington University, Bellingham, WA, 98225-9081. MEASURING CHANGES IN PIGMENTATION OF THE CORAL MONTASTRAEA FAVEOLATA VIA PHOTOGRAPHIC ANALYSIS.

Ioanna Visviki. Department of Biology, College of Mount Saint Vincent/Manhattan College, 6301 Riverdale Avenue, Riverdale, NY 10471. THE UNUSUAL CHONDRIOME OF *CHLAMYDOMONAS ACIDOPHILA* 

Thomas B. Widdowson. 4635 West Saanich Road, Victoria, B.C. V8Z 3G7 Canada. DATABASE: THE MARINE MACROPHYTES OF THE NORTHEAST PACIFIC (CONT'D.)

# 14<sup>th</sup> NORTHWEST ALGAL SYMPOSIUM ABSTRACTS MAY 12-14, 2000, UNIVERSITY OF BRITISH COLUMBIA (T=Talk, P=Poster)

#### KELP HOLDFAST ANCHORAGE MECHANICS: FORM AND FUNCTION

Sophie Boizard. Department of Botany, University of British Columbia, Vancouver, Canada.

P A biomechanical approach based on simple engineering beam theory combined with morphological measurements will be employed to investigate the functional reasons behind the wide variation in kelp holdfast forms observed in nature. The benthic marine environment is to a large extent one of moving water in which the survival of sessile marine organisms, such as kelps, depends on their ability to withstand hydrodynamic forces and on the ability of their holdfast to remain attached to the substratum. Without the strong attachment provided by the holdfast, kelps would quickly be dislodged by breaking waves and cast ashore before reaching maturity. Despite the fundamental role kelp holdfasts play in survivorship, they have been largely ignored in past studies. The objectives of the proposed study are to elucidate the mechanics of holdfast anchorage systems in kelps and to examine the influence of wave action on holdfast development.

# DISTRIBUTION AND ABUNDANCE OF CYANOBACTERIA AND LYTIC AGENTS INFECTIOUS TO SYNECHOCOCCUS SPP. IN THE STRAIT OF GEORGIA

Sean M. Brigden<sup>1</sup>, Martin Baur<sup>2</sup>, Curtis A. Suttle<sup>1,2,3</sup>. <sup>1</sup>Departments of Botany, <sup>2</sup>Microbiology & Immunology, and <sup>3</sup>Earth & Ocean Sciences, University of British Columbia, Vancouver, Canada

T Abundances of cyanobacteria and lytic agents specific for Synechococcus DC2 were estimated from water samples collected during a June 23rd-25th, 1997 research cruise of the Strait of Georgia. Cyanobacterial abundances were determined by enumeration of filtered water samples using epifluorescence microscopy and ranged from 2.6x103 to 9.4x107 L-1 with a mean of 5.7x106 L-1. Abundances were lowest at the bottom of the water column and increased with decreasing depth. Abundances usually peaked at the thermocline and halocline and decreased slightly at the surface. Titers of lytic agents were determined from concentrates obtained through ultrafiltration of seawater from integrated samples collected throughout the water column. Synechococcus strain DC2 was chosen as the test organism due to its low occurrence of resistance to viral infection and its susceptibility a wide array of viral isolates. Abundances of lytic agents for Synechococcus strain DC2 ranged from 90.2 to 8.86x103 lytic agents L-1 with a mean of 1.59x103 lytic agents L-1. The mean ratio of lytic agents to cyanobacteria was 3.7x10<sup>-4</sup>. Based upon counts of the number of virus particles in the water, these lytic agents accounted for  $1.1x10^{-4}\%$  of the total. These low titers of lytic agents affecting the test organism may be attributable to low host cell densities, as earlier research has shown that a host density >10<sup>6</sup> L<sup>-1</sup> is required for efficient viral propagation.

EFFECTS OF AMMONIUM ADDITION AND ENHANCED PAR AND UV-B IRRADIANCE ON PIGMENTS OF ZOOXANTHELLAE FROM THE CORAL *MONTASTRAEA FAVEOLATA* AT ELEVATED TEMPERATURE.

Brooke Budnick, Suzanne Strom, Kelley Bright and Erin Macri. Shannon Point Marine Center and Biology Dept., Western Washington University Bellingham, WA 98225-9081

P Coral bleaching results from pigment changes in zooxanthellae or the loss of these symbionts from their animal hosts. Experiments were conducted with the coral *Montastraea faveolata* collected from N. Perry Reef (16.5 m), Lee Stocking Isl., Bahamas in July of 1999 to determine how pigments are affected by environmental stresses. Corals were exposed to 30°C (2°C above ambient) and either deep (DR) or shallow (SR) reef light conditions and ammonium enrichments of 0, 2, or 10 μM for 96 hours. HPLC analysis was used to investigate treatment effects on pigments of in-host (AZ) and expelled (EZ) zooxanthellae. For most pigments, concentrations in AZ and EZ cells declined from initial levels, but ratios of chlorophyll c to total chlorophylls remained constant. AZ cells from the DR treatment had lower pigment content than SR samples. Chlorophyll c comprised about 25% of the total chlorophyll in both AZ and EZ cells. AZ cells from the 0 and 2 μM ammonium enrichments in both light treatments increased 5% in the photoprotective pigment diadinoxanthin while the photosynthetic pigment peridinin decreased 7%. There were no obvious trends in the distribution of pigments in EZ and AZ cells under the different treatments, suggesting that the elevated temperature was the main stressor.

### FURTHER EXAMINATIONS INTO THE ULTRASTRUCTURE OF *GLAUCOCYSTIS* NOSTOCHINEARUM: A COMPARISON OF CHEMICAL AND CRYO-FIXATION

<u>Kimberly Burns</u>, Brian R. Oates and Kathleen M. Cole. Department of Botany, University of British Columbia, Vancouver, B. C. V6T 1Z4

T We have used both chemical and cryo-fixation techniques to further our studies into the ultrastructure <u>Glaucocystis nostochinearum</u>. These techniques have produced striking differences in the ultrastructue of this alga. Most notably the chemical fixation technique results in the expected traditional cyanelle (chloroplast) envelope with two membranes separated by a cisterna. However, cryo-fixation provides different results. Presently, our results indicate that the cyanelle is surrounded either by two membranes without a cisterna or by a single membrane. The Golgi body also appears dramatically different in the two preparations. These results imply that chemical fixation introduces numerous artifacts and these artifacts may have confused our understanding of the ultrastructure of <u>G</u>. <u>nostochinearum</u>.

### RESERVOIR DRAWDOWN: IMPLICATIONS FOR LITTORAL BENTHIC COMMUNITIES AND DRINKING WATER QUALITY

Paula C. Caron. Department of Biology, University of Victoria, Victoria, BC, Canada

T Seasonal drawdown is a major physical feature distinguishing reservoirs from lakes and is thought to influence the biological, chemical, and physical processes and their interactions in

reservoirs. Traditionally, most research on freshwater ecosystems has focused on the pelagic environment. In comparison littoral zones have received less attention. Changes in thermal structure, light regimes, water column mixing, and sediment exposure caused by drawdown may affect the function or structure of littoral benthic communities, potentially resulting in reduced water quality. To determine the influence of reservoir drawdown on littoral benthic communities, I will examine temporal and spatial changes in periphyton and macroinvertebrates, and subsequent effects on water quality, in a drinking water reservoir which experiences large drawdown (>5m). These results will be compared to a lake that experiences little to no drawdown. Samples collected from littoral-pelagic transects in two coastal waterbodies (Sooke Reservoir and Shawnigan Lake, Victoria, British Columbia, Canada), will be analyzed for composition and biomass, and concentration of periphyton, macroinvertebrates, and taste and odour compounds.

### FEEDING PREFERENCE OF <u>LITTORINA SITKANA</u> ON BENTHIC DIATOMS, <u>FUCUS</u> <u>GARDNERI</u> EMBRYOS AND <u>ULVA FENESTRATA</u> SPORELINGS

<u>Heather I. Decious</u> <sup>1</sup> & Kathy Van Alstyne <sup>2</sup>. <sup>1</sup> Department of Biology, Western Washington University, Bellingham, WA, USA & <sup>2</sup> Shannon Point Marine Center, Western Washington University, Anacortes, WA, USA.

P Understanding herbivore food preference is important in understanding the role of herbivores in structuring rocky intertidal communities. Food preferences of herbivores among macroalgae have been well studied, but much less is known about preferences among microalgae. To examine preferences among microalgae, experiments were done to determine the food choice of Littorina sitkana among benthic diatoms (Navicula spp.), Fucus gardneri embryos, and Ulva fenestrata sporelings growing on polyethylene disks. Snails were given two foods and the amount of food removed and time spent feeding on each disk was recorded. Recently collected individuals were more likely to have a food preference than starved individuals. However, at the population level, neither starved nor recently collected snails had a significant preference for diatoms or Fucus. When offered Ulva sporelings, Littorina always preferred the alternate food choice, either Fucus embryos or benthic diatoms. These experiments demonstrate that snails make food choices at small scales and have the potential to influence community structure by selectively consuming or avoiding patches of microalgae or macroalgal sporelings.

# MAINTAINING SELF: AN OVERVIEW OF PHYSICAL AND BIOLOOGICAL INTERACTIONS IN THE *HEDOPHYLLUM* ZONE.

Robert E. DeWreede. Department of Botany, The University of British Columbia, Vancouver, B.C. V6T 1Z4, Canada

T Hedophyllum sessile (Phaeophyta) is a common and often predominant seaweed on the low intertidal shores of temperate Pacific North America. Hedophyllum is most common on shores with medium wave exposure, where it shares the habitat with various coralline and fleshy algal crusts, articulated coralline algae, and filamentous red algae. The predominant herbivore is Katharina tunicata, a chiton. This community of organisms has been the subject

of studies over the years, studies focussing largely on the impact of *Katharina* on adult *Hedophyllum*. Here, I summarize the results of these studies, and add new data relating to *Hedophyllum* juvenile recruitment and survival.

Specifically I examine mechanisms which enable this community to maintain its identity (or alter its constituent species and their relative abundance) in the face of common biological and physical variations such as:

- 1. A change in herbivore density.
- 2. A change in abundance of the dominant kelp, Hedophyllum.
- 3. A change in abundance of both the herbivore and Hedophyllum.
- 4. An increase (or change) in wave impact.
- 5. An increase in average surface sea water temperature (e.g. an El Niño year).

#### HYBRIDIZATIONS TEST KELP EVOLUTIONARY RELATIONSHIPS

Louis Druehl. Simon Fraser University at the Bamfield Marine Station, Bamfield, B.C., CANADA, V0R 1B0

The genera of the kelp families Alariaceae, Laminariaceae, and Lessoniaceae are morphological distinct. On the basis of molecular assessment, however, these genera are genetically very similar. Further, genetically derived phylogenies arrange the genera in non-traditional groups of taxa. To better understand the significance of these molecular phylogenies, we have undertaken hybridization studies. Kelp species, separated by varying degrees of molecular divergence, as determined from nrDNA Internal Transcribed Spacer 1 sequences, were crossed. Divergences ranged from 0% (several Japanese Laminaria species) to 28.13% (Egregia menziesii/Undaria pinnatifidia). Putative hybrids resulted from crosses involving divergences up to 17.89% (Laminaria saccharina/Lessoniopsis littoralis). A molecularly confirmed hybrid resulted from a cross of Lessoniopsis littoralis/Alaria marginata (molecular divergence 4.99%).

# DINOFLAGELLATE CHLOROPLAST GENOME: HOW MANY MINICIRCLES ARE THERE?

Elena Filek, Zhaoduo Zhang & B. R. Green. Department of Botany, University of British Columbia, Vancouver, Canada.

We recently reported the first dinoflagellate chloroplast gene sequences (Zhang et al., Nature 400, 155-159,1999). Zhaoduo Zhang sequenced nine chloroplast genes, coding for seven chloroplast proteins (psaA, psaB, psbA, psbB, psbC, atpA and petB) and two ribosomal RNA (16S and 23S). He also showed that in a number of dinoflagellate species, including H.triquetra, each chloroplast gene is located on a separate minicircular chromosome (one gene, one circle). This was shown for all nine genes reported. The question is how many chloroplast genes are organized in this way? We now report that petD and psbE genes are also located on minicircles in H.triquetra, and discuss methods employed in obtaining these sequences

#### ULVOID BLOOMS IN PUGET SOUND--CAUSE FOR CONCERN?

Gretchen Frankenstein. Puget Sound Water Quality Action Team, Olympia, WA, USA

P Although ulvoid blooms are a normal occurrence in many places in Puget Sound and have been recorded as early as the 1930s. In the past several years, however, shoreline residents, local government officials and scientists have suggested that bloom activity is increasing around Puget Sound. At present, what constitutes a bloom versus normal seasonal accumulation, causes of blooms and impacts of blooms in Puget Sound are not well known. The Puget Sound Water Quality Action Team and others have begun the process of examining this issue, but much work remains to be done to fully understand the occurrence, causes and impacts of ulvoid blooms in Puget Sound.

# THE GENETIC DIVERSITY AND ABUNDANCE OF VIRUSES THAT INFECT CYANOBACTERIA IN THE STRAIT OF GEORGIA, BRITISH COLUMBIA, CANADA

Cindy M. Frederickson, Steven M. Short & Curtis A. Suttle. Departments of Earth and Ocean Sciences, Microbiology and Immunology, and Botany, University of British Columbia, Vancouver, Canada.

P Water samples were collected from three locations (Salmon Inlet, Malaspina Inlet and Pendrell Sound) within the Strait of Georgia, British Columbia during an unusual cyanobacteria bloom between the dates of August 18-21, 1999. At each location, depth profiles from the surface to the bottom of the pycnocline were sampled. The abundance of viruses that lyse cyanobacteria belonging to the genus *Synechococcus* was estimated using most probable number assay (MPN) and the genetic diversity of cyanophages was estimated using denaturing gradient gel electrophoresis (DGGE) of PCR amplified cyanophage gene fragments. Highest cyanophage titers (3.4x10<sup>6</sup>ml<sup>-1</sup>) were found in surface samples (0.5-10m) while lowest cyanophage titers (8.5x10<sup>3</sup>ml<sup>-1</sup>) were found in the deepest samples (25m). Highest abundance of cyanophage corresponded to highest genetic diversity in both Malaspina Inlet and Pendrell Sound, but not in Salmon Inlet. These results indicate that the abundance of viruses that infect cyanobacteria can be very high, especially in surface waters and that highest cyanophage genetic diversity is not necessarily associated with highest cyanophage abundance.

# THE FLORAL RECOVERY OF A RESTORED COASTAL WETLAND: KUNZ MARSH, SOUTH SLOUGH, COOS BAY, OR.

Gisela Fritz<sup>1</sup>, Timothy Mulligan<sup>2</sup>, and Frank Shaughnessy<sup>1</sup> Department of Biological Sciences and <sup>2</sup> Fisheries, Humboldt State University, Arcata, CA

T Former coastal restoration projects have lacked subsequent monitoring studies, which are necessary to assess the performance of restored areas. The South Slough Estuarine Research Reserve (SSNERR), Coos Bay, Oregon, has aggressively restored diked areas. This presentation focuses on the floral recovery of restored sites in Kunz Marsh, SSNERR, at different elevations and compares these sites to natural habitats. Dependent variables include percent cover of vascular plants and the density of diatoms. Year, season, and treatment had a significant influence on vegetation cover with the overall cover being higher in 1999 than 1998;

there was also less cover in restored versus control sites. Summer was the season with the highest cover. In the restored treatment, cover increased more rapidly in higher sites, which also had a species composition similar to higher control sites. In contrast, recovery of lower restored sites is slower and dominated by the introduced species *Cotula coronopifolia*. Diatom densities during 1998 were maximal in control sites during the summer, and this pattern was even stronger within vegetated versus open patches. In restored sites, diatom densities generally decreased through the year and did not appear to differ between patch types. Restoration efforts in Kunz Marsh have therefore resulted in a rapid increase in vascular plant cover in higher sites, whereas the re-establishment of diatom communities appears

#### UROSPORA (CHLOROPHYTA) ON THE PACIFIC COAST OF NORTH AMERICA

Louis Hanic. Department of Botany, University of British Columbia, Vancouver, B. C., Canada

T Field and culture studies on *Urospora* from 50 sites (San Luis Obispo, California, to Prince Rupert, B. C.) have yielded 9 forms (species?) based mainly on sexual characters: 5 anisogamous (4 dioecious, 1 monoecious), two unisexual (1 male, 1 female), and two asexual. Variations in filament morphology, gamete morphology and life history will be presented. Using these same criteria, seven species are recognized for Atlantic Canada and four for Europe. *Urospora penicilliformis* is the most common species on the Pacific Coast. Hybrid crosses between clones of *U. penicilliformis* from B. C., Germany and Atlantic Canada produced viable zygotes and F-1 sexual filaments. Positive matings were also obtained with clones from B. C. and Japan. This indicates that *U. penicilliformis* is circumboreal. This study is ongoing, and living samples, raw or cultured, are welcome. Clonal subcultures of main types are available to interested researchers.

IS THERE A CONNECTION BETWEEN DUST AND PHYTOPLANKTON?: HOW IRON LIMITATION INFLUENCES PRIMARY PRODUCTIVITY IN THE NE SUBARCTIC PACIFIC

Paul J. Harrison. Department of Oceanography University of British Columbia Vancouver, British Columbia, Canada

The time series at Stn Papa (Stn P) at 50N and 145W in the subarctic Pacific, spans more than 40 years, making it one of the longest open ocean time series in the world. Our understanding of this ecosystem during the last four decades has evolved. It was originally thought that the phytoplankton in this region were under 'top down' grazing control. Unlike most temperate oceanic ecosystems, there is no spring bloom and the phytoplankton biomass is relatively constant year round. This lack of seasonal cycle in phytoplankton was thought to be due to grazing by large zooplankton that kept the phytoplankton cropped down. To support this 'major grazer' hypothesis, was the fact that nitrate never became limiting during the spring and summer months. Shipboard experiments conducted by the late John Martin revealed, to the disbelief/amazement of many of his colleagues, that when a small (2 nM) amount of iron was added to surface seawater during shipboard experiments, large pennate diatoms grew up. For two other large areas, the Equatorial Pacific and the Southern Ocean,

iron has also been shown to limit phytoplankton biomass and productivity. Martin termed these areas, High Nitrate, Low Chlorophyll (HNLC) regions. Recent experiments at Stn P have shown that the small phytoplankton are mainly controlled by microzooplankton grazing, while the large phytoplankton are mainly controlled by iron. Thus, bottom up (Fe limitation) and top down (grazing) control both operate and control different size fractions of phytoplankton. Future research will focus on the sources of iron which appear to be episodic in nature. The potential sources od iron include dust from the Gobi Desert, large eddies which form off the Queen Charlotte Islands and move offshore, ash from Alaskan volcanoes and forest fires and horizontal surface water transport from the Aleutians via the Alaska Gulf Stream Current. Future large scale open ocean fertilization experiments are planned for 2002 at Stn P.

#### WHO'S WHO IN THE ULVACEAE?

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Ulvaceae are morphologically simple, cosmopolitan green macroalgae. Family members, particularly species of *Ulva* and *Enteromorpha*, are prevalent in coastal ecosystems and responsible for "green tide" events around the world. Little is known about the evolutionary history of Ulvaceae. Traditional classification of the family is based on shared morphological, reproductive and developmental characters, including life cycle, motile reproductive cell morphology and germling development. Several conflicting treatments of the family have been proposed. Within these, the placement of *Chloropelta*, *Enteromorpha*, *Ulva*, *Ulvaria* in Ulvaceae is common; however, the placement of *Blidingia*, *Capsosiphon*, *Kornmannia*, and *Percursaria* in the family is controversial. We test systematic hypotheses using nuclear and chloroplast DNA sequences. Phylogenetic analyses support a monophyletic Ulvaceae consisting of *Chloropelta*, *Enteromorpha*, *Percursaria*, *Ulva* and *Ulvaria*. The systematic position of *Blidingia*, *Capsosiphon* and *Kornmannia* will be discussed.

#### KELP HYBRIDIZATION STUDIES

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To further study the inter-relatedness of kelp, we conducted reciprocal crosses of female and male gametophytes from species with different degrees of genetic relatedness. Gametophytes were cloned from single-cell isolates. These were then put into the following situations: reciprocal crosses, selfings, and male and female controls, and cultivated at 10 °C, 16: 8, 80 μ Mol m<sup>-2</sup> sec<sup>-1</sup> and in F/2 medium. Sporophytes were screened through successive culturing stages. We were able to successfully rear viable normal and abnormal juvenile sporophytes. Species of Laminaria, Eisenia x Kjellmaniella, Eisenia x Laminaria, Pterygophora x Macrocystis and Laminaria x Lessoniopsis gave rise to putative sporophytes.

PHOTOBIOREACTOR CULTIVATION OF AGARDHIELLA SUBULATA REGENERATED MICROPLANTLET

<u>Yao-ming Huang</u> & Gregory L. Rorrer. Department of Chemical Engineering, Oregon State University, Corvallis, Oregon, USA

P Suspension cultures derived from macrophytic marine algae are a potential source of pharmacologically active compounds. Controlled biological production of these compounds first requires the development of an engineered biomass production system to illustrate the application of bioprocess engineering principles to this new area of marine biotechnology. A novel microplantlet suspension culture of the macrophytic marine alga *Agardhiella subulata* was established, and the effects of process parameters on photosynthetic growth and primary metabolic processes were examined. Specific process parameters included medium perfusion, CO<sub>2</sub> supply and light delivery. Controlled cultivation of *Agardhiella subulata* microplantlets was successful in both externally-illuminated bubble-column and stirred-tank bioreactors. The growth was not CO<sub>2</sub>-limited in either bioreactor system when aerated at 0.3 vvm with ambient air of a k<sub>L</sub> a greater than 30 hr<sup>-1</sup>. Light-saturated growth was achieved at an incident light intensity of 43 mE/m2-s. Specific oxygen evolution rate vs cultivation time measurements were used to estimate maintenance requirements for photosynthetic growth. Medium perfusion during the bioreactor cultivation improved the specific growth rate and oxygen evolution rate.

### EFFECTS OF PH, PHOTOPERIOD AND TEMPERATURE ON THE CULTIVATION OF AGARDHIELLA SUBULATA REGENERATED MICROPLANTLET

Yao-ming Huang & Gregory L. Rorrer. Department of Chemical Engineering, Oregon State University, Corvallis, Oregon, USA

P A microplantlet suspension culture of the red macroalga, *Agardhiella subulata*, was established through controlled regeneration of filamentous clumps. The microplantlet cultures were maintained in an incubator at 24°C and 20-36 mE/m²-s irradiance with a photoperiod of 10:14 LD on ASP12 medium at pH 8.0. To optimize microplantlet growth, the effects of pH, temperature and photoperiod were investigated. The optimal range of pH and temperature for the microplantlet culture was determined using oxygen evolution rate (OER) measurements during illumination phase. At pH of 8.7 or higher, the culture showed a lower OER as well as the specific growth rate. The OER increased when the temperature was increased from 24 to 33°C within the first two hours after the temperature change, but then decreased dramatically after 24 hours. Microplantlets incubated at 24°C had the highest OER during the 30-day cultivation period. At saturation incident light intensity of 43 mE/m²-s, the specific growth rate increased linearly with increasing photoperiod from 5:19 to 10:14 LD, whereas photoperiods of 20:4 LD or higher inhibited culture growth. From this information, the optimal photoperiod for the *Agardhiella subulata* regenerated microplantlets was between 12:12 and 16:8 LD.

IN VITRO PROTEIN IMPORT INTO CHLOROPLASTS ISOLATED FROM A HETEROKONT ALGA: HETEROSIGMA AKASHIWO.

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P Heterokont algae have chloroplasts with four envelope membranes and their nuclear-encoded chloroplast proteins have to be transported from cytoplasm to chloroplasts across the four membranes. In order to clarify the mechanism of protein import into the heterokont chloroplast, we isolated intact chloroplasts with three envelope membranes from *Heterosigma akashiwo*, and established an in vitro protein import system using these chloroplasts. Precursors we used were full-length fucoxanthin-chl a/c binding proteins (FCPs) that transcribed/translated from a cDNA clone and its deletion mutants which lacked the putative ER signal sequences. The FCP precursors with and without the signal sequence were both imported into intact chloroplasts, while green fluorescent proteins (GFPs) with only the FCP's ER signal sequence or with the whole presequence were not imported. We also confirmed, using canine microsomes, that the ER signal sequence of the FCP is functional and cleaved off in the microsomes. Endo-H digestion showed the proteins were not glycosylated in the microsomes.

### PHAGOTROPHY IN GONYAULACOID DINOFLAGELLATES AND A HYPOTHESIS FOR THEIR ADAPTIVE RADIATION

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T Gonyaulax verior cells taken from a single Puget Sound sample were found to contain only ingested cryptophyte prey (5 out of 5 cells, up to 3 prey/cell), suggesting selective feeding. G. verior is a small, dorsally compressed species, unlike the larger spheroidal gonyalacoid species including G. kofoidii, G. alaskensis, G. polygamma and numerous Alexandrium species, (including A. ostenfeldii and A. psueduogonyaulax). The latter all prey (at least in part) on ciliates larger than 15 um, as well as occasional dinoflagellate prey. Other spheroidal species that contain large food vacuoles (possibly ingested ciliates) include G. grindleyi and G. spinifera. These data suggests as a working hypothesis that some of the diversity of dinoflagellate size and shape may result from adaptations of niche partitioning (small species adapted to small prey, etc.) This unanticipated diversity of prey requirements among mixotrophic dinoflagellates also sheds light on the plankton paradox.

# ULTRASTRUCTURAL DEMONSTRATION OF INGESTION OF PHAESOME SYMBIONTS BY ORNITHOCERCUS (DINOPHYCEAE)

Dean M. Jacobson, Department of Biology, Whitworth College, Spokane Washington, USA

P Ornithocercus magnificus cells collected from the Gulf Stream off of Ft. Pierce, Florida, were found to contain numerous food vacuoles. These vacuoles fall into two groups: amorphous, well-digested spheroidal bodies (similar to food vacuoles seen in Dinophysis spp.) and cylindrical bodies containing numerous membrane profiles. The latter resemble the living cyanobacterial phaeosomes found in the inflated cingular space in size, and, in internal

appearance, resemble a lysed phaeosome. Evidently these intricate heterotrophic dinoflagellates maintain a ikitchen gardenî of cyanobacteria (to use Fenchelis term) which they utilize as a food supplement (probably ingested via peduncle); other prey, not yet identified, is also taken. In addition, small heterotrophic bacteria were found near the cytostome, as has also been found in a mixotrophic *Dinophysis fortii* cell.

# A CHARACTERIZATION OF ULVOID BLOOMS AND THEIR ABIOTIC ENVIRONMENT IN THE SAN JUAN ARCHIPELAGO.

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P Marine blooms of ulvoid macroalgae (e.g., Ulva, Ulvaria, and Enteromorpha) negatively impact their environments by competing with seagrass meadows and by creating anoxic or hypoxic conditions at night. This project quantified the extent and the composition of ulvoid blooms and examined the physical and chemical factors important to ulvoid communities. Six sites around Blakely Island were sampled quarterly over a two year period. Ulvoid biomass, chlorophyll content, and photosynthetic rate, were determined for samples collected at 0, -1, and -2 m MLLW. Water temperature, clarity, salinity, oxygen concentration and nutrient concentrations were measured at each site. Blooms consisted mostly of Ulva and Ulvaria. Enteromorpha is less abundant, especially at greater depths. The ratio of Ulvaria to total ulvoid biomass increases with depth. The four sites on north end or the east side of the island (and exposed to the highest currents and wave action) had the highest ulvoid biomass. In contrast, the two sites on the west side of the island (in more protected waters) had the lowest biomass. Chlorophyll a content was consistently higher in winter that in other seasons. Light-saturated photosynthesis was lower in the winter compared to other seasons. Physical and chemical factors also varied seasonally between sites. Oceanographic differences were considered to be a major cause of the variation in physical, chemical and biological parameters.

# A CHLORARACHNIOPHYTE ISOLATE WITH A MULTINUCLEAR CELL STAGE IN THE LIFE CYCLE

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P Chlorarachniophytes are marine unicellular algae that have chloroplasts with chloroplhylls a/b and four surrounding membranes. These chloroplasts, which originated from a green algal endosymbiont, are still accompanied by the endosymbiont's reduced nuclei (nucleomorphs). We isolated a chlorarachniophyte alga from a sand sample collected at a coral lagoon in Guam. In culture, the cells were amoeboid, approx. 8 micrometer in length, and uninuclear, and proliferated by simple cell division. Under certain conditions, however, large spherical cells, up to 70 micrometer in diameter, were observed. DAPI or SYBR-Green staining of these large cells demonstrated that each cell contained 2 to 18 nuclei. When culture medium was replaced by fresh one, these large cells became amoeboid, then divided into

several uninuclear-amoeboid cells. DAPI or SYBR-Green staining of these large cells demonstrated that each cell contained 2 to 18 nuclei. When culture medium was replaced by fresh one, these large cells became amoeboid, then divided into several uninuclear-amoeboid cells. Ultrastructural feature of pyrenoid and position of the nucleomorph were the same as those of the genus *Gymnochlora*, confirming that the present alga belongs to this genus. Taxonomic relationship between the present alga and *Gymnochlora* stellata (the type species of *Gymnochlora*) will be discussed.

# INCORPORATING SEAWEEDS AND SEAGRASSES IN THE DESIGN OF MARINE PROTECTED AREA NETWORKS

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Marine protected areas (MPAs) and MPA networks increasingly are being established for the protection and recovery of marine biological resources, and considerable attention is being devoted to their design. In particular, linkages between individual sites within a larger network are thought to provide benefit to the system as a whole. I performed a drift card study to describe dispersal envelopes within the San Juan Archipelago and surrounding waters. The results show generally high degrees of linkage within the San Juans and throughout the eastern basin of the Strait of Juan de Fuca. However, these linkages are strongest for animal species with extended planktonic phases and for buoyant macrophytes (e.g., Nereocystis). The implications of these findings for the conservation of macrophyte populations will be discussed.

#### ENVIRONMENTAL DETERMINANTS OF ULVOID ALGAL SPECIES COMPOSITION

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P Blooms of ulvoid algae may cause environmental problems such as smothering eelgrass eadows. In the vicinity of Blakely Island, Washington, blooms are composed primarily of Ulva and Ulvaria. These two genera are typically found at different depths: Ulva was found in shallow waters (0 m and -1 m MLLW) while Ulvaria was found in deeper water (-1 m and -2 m MLLW). Several experiments were conducted to determine the environmental factors that cause this difference in distribution. Growth rates and interspecific competition were measured in Ulva and Ulvaria at different depths. At 0 m Ulva grew significantly faster than Ulvaria. At -2 m there was no significant difference in the growth rate between the two species. When subjected to different light regimes, Ulva appeared to respond to changes in light, while Ulvaria did not adapt. The total pigment content and the ratio of accessory pigments in chlorophyll a in Ulva increased with depth. Desiccation tolerance was measured using air drying and saline treatments. Ulva has a higher desiccation tolerance than Ulvaria. Ulvaria discs desiccated for longer than 60 minutes excreted a dark colored substance. This excretion suggested that Ulvaria might be grazer resistant. Grazing experiments showed that Ulva is preferred over Ulvaria by Littorina sitkana. The results of these experiments suggest that the difference in species composition of ulvoids at different depths may be due to the combination of desiccation and grazing.

ANCIENT ATMOSPHERIC  $\mathrm{CO}_2$  AND THE TIMING OF EVOLUTION OF SECONDARY ENDOSYMBIOSES

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Atmospheric CO<sub>2</sub> levels were reduced to historic lows, probably lower than today's atmosphere, during the Pennsylvanian and Permian periods. This most likely resulted in dissolved inorganic carbon (DIC) becoming the limiting factor in photosynthesis in these ancient oceans. A number of algal secondary endosymbioses evolved between 259 and 285 million years ago during this minimum in atmospheric CO<sub>2</sub>. These algae were able to more efficiently utilize DIC because their chloroplasts were contained within an acidic compartment where DIC was largely in the form of CO<sub>2</sub>. We postulate that secondary endosymbioses arose continuously from the time of evolution of the chloroplast (about 2,000 million years ago). These secondary endosymbioses were quickly eliminated, however, because they possessed no selective advantage over existing phytoplankton in waters high in DIC. It was not until the ancient atmospheric CO<sub>2</sub> minimum that secondary endosymbioses were selected for, because these algae were able to more efficiently utilize the low DIC and outcompete existing algae. Under these favorable conditions, a number of secondary endosymbioses evolved and survived, and it is their ancestors that constitute most of the eukaryotic phytoplankton in today's oceans.

# A FLORA OF THE BENTHIC MARINE ALGAE OF ALASKA--STATUS AND FUTURE PROSPECTS

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We have initiated Phase 1 of a Flora of the Benthic Marine Algae of Alaska: an inventory of existing collections. To date, we have compiled ca. 25,000 records of specimens in an ACCESS database. Our inventory has revealed more than 1100 names that have been used for Alaskan seaweeds, including about 100 with Alaskan types. Somewhat less than 50% of these names probably represent distinct species. However, little work has been done to test this estimate. In order to complete further phases of the Flora, we need colleagues to take an interest in the systematics of Alaskan species. Taxa most in need of revision include red algae in the Kallymeniaceae, Phyllophoraceae, Gigartinaceae, Ceramiaceae, Delesseriaceae, and Rhodomelaceae, green algae in the Acrosiphoniaceae, Cladophoraceae, and Ulvaceae, and brown algae in the Coilodesmaceae, Dictyosiphonaceae, Punctariaceae, and Alariaceae. Our inventory, documenting existing collections, provides a starting point for these muchneeded systematic revisions.

# THE ENVIRONMENTAL MANAGEMENT OF DRINKING WATER ECOSYSTEMS: AN ECOSYSTEM AND WATERSHED APPROACH

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- P The NSERC-IRC program examining the environmental management of drinking water is a unique opportunity to apply basic ecological and limnological concepts and theories to directly benefit Canadian water utilities and forest and environmental technology based industries, with whom we are in partnership. Drinking water reservoirs, like natural lakes, are regulated by complex processes and interactions. Our goal is to develop a watershed-level understanding of the factors and processes regulating water quality within drinking water reservoirs. It is important to understand factors and processes regulating algal composition, major water column processes, trophic relationships, and the effects of variable drawdown. Better understanding and knowledge regarding the functioning of these ecosystems will allow for more environmentally sound management decisions.

#### A BIOLOGICAL SURVEY OF THE BERKELEY PIT.

- Grant G. Mitman. Department of Biological Sciences, Montana Tech of the University of Montana, Butte, MT 59701
- P The Berkeley Pit Lake System is estimated to contain more than 30 billion gallons of acidic, metal-laden water. Current investigations examine how the naturally occurring organisms in the Berkeley Pit Lake System might be stimulated by nutrient additions to begin a self-purification process for aqueous environments impacted by acid mine drainage. The combined physiological processes have been observed to bioremediate aquatic mine waste environments. Consequently, if a mine waste site like the Berkeley Pit Lake System is properly nutrified with Nitrogen, Phosphorous, or Potassium (eg. Manure or sewage as inexpensive sources), this nutrification may cause a successional cascade of increased diversity and biomass that is coupled with an increase in pH. A pH increase, in turn, may lead to a natural restoration process. Thus, if systems are to function correctly and to recover from pollution-induced perturbations, fundamental information both on the autotrophic and on the heterotrophic components of the microbial community is essential. Defining the baseline community structure is the first step toward understanding the interactions of the different groups of extremeophiles and toward assessing any improvement in biodiversity

#### ALGAL BIOREMEDIATION OF THE BERKELEY PIT LAKE

- Mitman, Grant G. Department of Biological Sciences-Montana Tech of The University of Montana, Butte, Montana 59701
- T Ongoing research is unraveling the intricacies of the microbial ecology of the Berkeley Pit Lake System, with ever increasing information becoming available regarding the diversity of Algae, Protistans, Fungi and Bacteria that inhabit this mine waste site. Defining the baseline community structure has been the first step not only toward understanding the interactions of the different groups of organisms, and also toward assessing any improvement in biodiversity

within the biotic community. Now that this first step has begun, some of these extremophiles, specifically algae, that have been isolated from the Berkeley Pit Lake System are being used as a potential solution for bioremediation. The specific objectives of this research are fivefold: 1) To evaluate the bioremediative potential of our four most rapidly growing species: (Chromulina freiburgensis Dofl., Chlorella ellipsoidea Gerneck, Chlorella vulgaris Beyerinck and Chlamydomonas acidophilla Negoro) in Berkeley Pit Lake System Water with the additions of NaNO3 and NaPO4 by using an experimental matrix. This matrix will be used to estimate the minimum nutrient concentrations that would be necessary to achieve the maximum growth of algae and maximum bioremediation of the Berkeley Pit Lake Sys-2) To determine which combination of nutrients will stimulate growth of the best bioremediator of our four isolated species in natural Berkeley Pit Lake System waters. In other words, what nutrient combination will give the best bioremediator a competitive edge over the other species. If time permits-different species may be grown in combination to determine if there are synergistic effects (protocooperation) between species. 3) To determine a temperature profile for these four species in order to determine their optimal growth temperature in Berkeley Pit Lake System water. 4) To continue to isolate organisms from the Berkeley Pit Lake System and determine their bioremediative potential. 5) Monitor algal and bacterial counts from a profile of Pit Lake System waters. The results to date will be presented for this conference.

PHOTOSYNTHETIC UTILIZATION OF HCO<sub>3</sub><sup>-</sup> IN 38 SPECIES OF PUGET SOUND RED ALGAE: AN ECOLOGICAL AND TAXONOMIC COMPARISON.

Maurizio Murru\_and <u>Craig D. Sandgren</u>. Department of Biological Sciences, University of Wisconsin - Milwaukee, Wisconsin, USA.

P All photosynthetic eukaryotes can utilize dissolved CO<sub>2</sub> as a source of carbon for the dark reactions of photosynthesis. The ecologically important ability to utilize the HCO<sub>3</sub> fraction of dissolved inorganic carbon (DIC) from seawater during photosynthesis was investigated for 38 species of intertidal and subtidal Puget Sound red algae. The pH-drift technique was used. In a closed system held at constant temperature, freshly collected specimens were incubated under constant and continuous illumination until a stable pH value was reached in the seawater medium. Among the red algae tested, there was a wide diversity observed in the ability to raise the pH in the medium sufficiently to require that the algae were utilizing the HCO<sub>3</sub> fraction of the DIC. These data were analyzed with respect to the distribution of the species with regard to tidal height on rocky shores and with regard to the consistency of physiological responses within taxonomic families and orders.

EFFECTS OF AMMONIUM ADDITION AND ENHANCED PAR AND UV-B IRRADIANCE ON THE PRODUCTIVITY OF IN-HOST AND EXPELLED ZOOXANTHELLAE FROM THE CORAL *MONTASTRAEA FAVEOLATA*.

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P Coral bleaching (the loss of symbiotic zooxanthellae) is a widespread phenomenon in nutri-

ent poor reef-bearing waters. Samples from *Montastraea faveolata* colonies at 16.5 m depth, Lee Stocking Isl., Bahamas were exposed to deep reef (DR) or shallow reef (SR) light conditions at ambient temperature (28°C) for 96 hours. Corals received 0, 2, or 10 µM ammonium enrichments. Photosynthesis-irradiance (P-I) parameters of zooxanthellae were compared. Zooxanthellae isolated from the host (AZ) did not change from initial P-I parameters after exposure to increased nutrients and irradiance. However, zooxanthellae expelled by the coral (EZ) under DR conditions showed increased photosynthetic efficiency (alpha) and photosynthetic capacity (Pmax) with exposure to increased ammonium. Alpha and Pmax of EZ also increased with ammonium under SR light, but did not exceed AZ values. These results indicate that the host coral animal withholds nitrogen from the zooxanthellae and that the host provides adequate protection from increased PAR and UV irradiance. Greater Pmax and alpha values for EZ cells under DR light compared to SR suggest the lack of coral protection is more detrimental to zooxanthellae at higher irradiance and UV-B levels. The increase in alpha and Pmax of EZ at greater ammonium concentrations show that expelled zooxanthellae utilize the added ammonium available in the seawater to enhance their productivity.

#### ROLE OF NITROGEN AVAILABILITY IN THE RESPONSE OF THE CORAL MON-TASTRAEA FAVEOLATA TO ENVIRONMENTAL STRESS

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P Evaluating the rate at which stressed coral, along with its endosymbiotic zooxanthellae, consumes or releases ammonium is important to assess coral responses to stress. In July 1999, we studied the coral *Montastraea faveolata* at Lee Stocking Island, Bahamas, as part of the Science Education and Research for Undergraduates (SEARUN) program funded by NSF. Under normal conditions, zooxanthellate corals take up ammonium from seawater. We hypothesized that high temperature and elevated light would progressively stress the coral, rendering it unable to maintain high uptake rates and perhaps causing ammonium excretion. We enriched coral plugs daily with ammonium at 2 and 10 μM seawater concentrations and measured seawater ammonium concentrations over time. Enrichment with 10 μM ammonium led to higher uptake rates than enrichment with 2 μM ammonium. Exposure to elevated light did not affect ammonium uptake of corals enriched with nutrients. Addition of ammonium led to elevated chlorophyll levels and seemed to enable the corals to withstand environmental stress. Unenriched corals, which showed evidence of bleaching, sometimes released large amounts of ammonium.

### HALOGENATED MONOTERPENE PRODUCTION IN MICROPLANTLET SUSPENSION CULTURES OF THE TROPICAL RED ALGA OCHTODES SECUNDIRAMEA

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T Photosynthetic microplantlet suspension cultures derived from the tropical red alga Ochtodes secundiramea contain seven novel halogenated monoterpenes and bromoperoxidase, an en-

zyme that halogenates organic substrates. Microplantlet suspension cultures of *O. secundiramea* were established by callus induction and shoot tissue regeneration techniques, and then cultivated in a bubble-column photobioreactor. Specific bromoperoxidase activity decreased during the exponential phase of growth then increased six-fold during stationary phase, suggesting that halogenation reactions were part of secondary metabolite biosynthesis. Halogenated monoterpene production by *O. secundiramea* microplantlets was strongly favored by cultivation at high light intensity and low nutrient availability, again consistent with patterns of secondary metabolite biosynthesis by the carbon-nutrient balance hypothesis. One specific biosynthetic pathway identified in the culture system was the conversion of geranyl diphosphate, the common precursor for monoterpenes, to myrcene, followed by the bromination of this acyclic monoterpene to bromom yrcene.

### MODELING AND SIMULATION OF A TUBULAR RECYCLE PHOTOBIOREACTOR FOR MACROALGAL CELL SUSPENSION CULTURES

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P The tubular recycle photobioreactor is a novel system for the mass cultivation of photosynthetic cell suspension cultures. In the tubular recycle photobioreactor, the suspension culture is re-circulated between two sections: a coiled tubular section which illuminates the culture with a small light path, and a non-illuminated aeration tank which supplies the dissolved CO<sub>2</sub> needed for photosynthetic biomass production. A mathematical model was developed to predict the cell density vs. time profile for the cultivation of photosynthetic cell suspension cultures in the tubular recycle photobioreactor. This model uniquely couples culture illumination parameters in the tubular section to interphase CO<sub>2</sub> mass transfer parameters in the aeration tank. Model simulations show that the tubular recycle photobioreactor is operating at saturation growth kinetics with respect to light, and that CO<sub>2</sub> delivery to the tubular photobioreactor limits the biomass production rate at high cell density until nutrient depletion is achieved.

A HEDGE AGAINST HERBIVORES? THE EFFECTS OF ALGAL COLONY SIZE AND THE PRESENCE OF 'SPINY" SILICEOUS CELL COVERINGS ON GRAZING RATES BY ZOOPLANKTON ON PLANKTIC CHRYSOPHYTE FLAGELLATES.

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Most planktic zooplankton select algal prey based strongly upon the apparent size of the algal 'particles'. Grazing rates on are inversely related to algal size: larger prey experience reduced loses to planktic herbivores, and sufficiently large particle size can constitute a true refuge from herbivory. Exuberant algal cell coverings and the colonial growth habit both represent potential morphological adaptations by which microalgae can become larger in apparent particle size as a hedge against herbivores, and at the same time, avoid suffering many of the physiological constraints imposed by large individual cell size. Testing this possibility with the existing published literature is not possible, however, because of the many inconsisten-

cies in experimental techniques among the many previous grazing studies. We have developed techniques using nutrient stress and mechanical agitation to create gradients of colony size and cell covering morphology among populations of individual clones of planktic chrysophytes in culture; e.g. normally-scaled vs. scale-free populations of *Synura* and *Mallomonas*, unicellular vs. large-colony populations of *Synura* and *Dinobryon*. These morphologically distinct populations have been fed to freshwater herbivorous zooplankton to develop prey density-dependent feeding curves using a standard experimental protocol. Similar studies were performed with the same prey populations for a gradient of different-size herbivores (ciliate, rotifer, four cladocerans) in order to examine the interaction between animal size and the morphology of chrysophyte cells and colonies. All of the chrysopyte species and clones tested were grazed by all animals tested; no chrysophyte demonstrated a complete refuge from herbivory based on particle size or shape. But, both coloniality and siliceous scale coverings greatly reduced grazing rates by zooplankton, and both adaptations were more effective against smaller-bodied animals.

### A PRELIMINARY ANAYSIS OF ALGAL AND INVERTEBRATE RECOVERY FOLLOWING AN OIL SPILL IN HUMBOLDT BAY, CALIFORNIA

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T On November 6, 1997 the M/V Kure collided with the Louisiana-Pacific Company dock in Humboldt Bay and released about 5,000 gallons of bunker fuel oil into the bay. Several intertidal riprap sites were either lightly covered by oil and not cleaned, or more heavily impacted and cleaned with high pressure and high temperature water. Cleaned sites were also rinsed with ambient seawater. Several students and I designed a study to determine the effects of this oiling event on the mid-intertidal riprap community. Two oiled and cleaned sites, two oiled and uncleaned sites, and two unoiled sites were established. The percent cover of algal and invertebrate functional groups (e.g. cartilaginous algae: mostly *Mastocarpus papillatus* and *M. jardinii*) over the last two years was greater at nonoiled sites, but among oiled sites algal abundance was aseasonal, and abundance was consistently higher in cleaned versus uncleaned sites. In contrast, limpet abundance among oiled sites does not differ until summer recruitment events at cleaned sites. Both the algae and invertebrates are therefore still recovering, and cleaning may be slightly increasing the rate of recovery. The small size of the oil spill, and consequently the lower intensity cleaning effort, is probably why recovery is occurring relatively quickly in Humboldt Bay.

### FIELD STUDIES OF THE LIFE HISTORY OF ACROSIPHONIA (CODIOLALES, CHLOROPHYTA) IN SOUTHERN BRITISH COLUMBIA

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The relationship of the endophytic sporophytes and filamentous, free-living gametophytes of *Acrosiphonia* was established in nature. Filamentous plants were seasonal (spring / summer), becoming fertile almost immediately after their appearance. The sporophytes colo-

nised their hosts one month later, and showed higher tolerance to abiotic factors than the gametophyte: high summer temperatures (which correlated with death of the free-living plants) were survived and they overwintered in their hosts. Endophytes matured primarily in winter, with zoospore release occurring throughout winter / spring. Filamentous gametophytic plants were re-established in spring. The hosts, *Mazzaella splendens* and 'Petrocelis franciscana' (= crustose tetrasporophytic phase of *Mastocarpus papillatus*), were available for endophyte colonisation in spring / summer. Several factors, *e.g.* herbivory, winter storms and senescence, were identified in significant host tissue loss, thus potentially affecting endophyte survival. I suggest the endophytes have evolved a strategy whereby duration in the host is synchronised with host seasonality. An investigation of other hosts for *Acrosiphonia*'s sporophyte established a wide range of hosts. A bet-hedging strategy is proposed for *Acrosiphonia*'s life history: not only have two morphologically different phases adapted to a seasonally variable environment, but the sporophytic phase colonises both crustose and foliose red algae, and low host specificity is evident for both endophytes.

### ISOLATION AND CHARACTERIZATION OF VIRUSES INFECTING HETEROSIGMA AKASHIWO (RAPHIDOPHYCEAE)

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T Blooms of Heterosigma akashiwo are implicated in kills of wild and farmed fish around the world. The factors regulating H. akashiwo bloom dynamics are not well understood, but viral infections have been implicated in bloom termination. We have detected viruses from the Strait of Georgia, British Columbia that lyse H. akashiwo. The detection, isolation, and characterization of H. akashiwo viruses involved ultrafiltration, infection assays, large-scale cultures, and purification techniques. Using these procedures, we have isolated viruses that are very different from H. akashiwo viruses detected in Japanese waters (HaV) and other known algal viruses. One of these isolates causes ultrastructural changes in the cytoplasm of infected cells. The virus is small, ~25 nm in diameter, spherical, and may contain RNA. We have also isolated another H. akashiwo virus of similar size, but assembles as crystalline arrays inside the nucleus (HaNIV). These results indicate that the diversity of viruses infecting H. akashiwo is higher than anticipated. The viruses we have isolated do not appear to belong to the Phycodnaviridae, the algal virus family that includes HaV and other algal viruses. Further molecular characterization and phylogenetic studies are required to determine the relationships between H. akashiwo viruses and the Phycodnaviridae.

### MEASURING CHANGES IN PIGMENTATION OF THE CORAL MONTASTRAEA FAVEOLATA VIA PHOTOGRAPHIC ANALYSIS.

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P Algal symbionts (zooxanthellae) are critical to the growth and survival of reef-building corals. Elevated temperature and UVB radiation can cause coral to lose their algal symbionts

and pigmentation in a process known as coral bleaching. Photoanalysis may provide a convenient and rapid method for quantifying coral bleaching without extensive chemical or biological analysis. Experiments were performed at the Caribbean Marine Research Center, Lee Stocking Island, Bahamas in July of 1999. Two-cm diameter plugs of the coral *Montastaea faveolata* were collected from a reef at 16 meters depth, placed in an incubator and exposed to combinations of enhanced PAR (photosynthetically active radiation), UVB (ultra-violet b), and elevated temperature for 96 hours. After treatment the coral plugs were individually photographed against a white scale, and then processed for pigments, nitrogen, carbon, and photosynthetic rates of zooxanthellae. We quantified the mean and variance of the luminosity for each coral sample using Adobe PhotoShop, and examined the changes in these values as the level of bleaching increased. Initial data shows that coral bleaching is easily detected and differentiated by comparing the mean and variance of luminosity. Further analysis will examine the relationships between luminosity and changes in pigmentation, and photosynthetic rates.

#### THE UNUSUAL CHONDRIOME OF CHLAMYDOMONAS ACIDOPHILA

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P Chlamydomonas acidophila is a unicellular, flagellated green alga of the order Volvocales, family Chlamydomonadaceae. It has been isolated from lakes and acidic bogs with pH as low as 2. The ultrastructure of this chlorophyte is unique, due to the large size and position of its mitochondria. The mitochondrial volumes of other Chlamydomonas species vary between 1-3% and the mitochondria are located directly below the cell membrane. The mitochondrial volume of C. acidophila varies between 4-6% and the mitochondria are located below the chloroplast. Examination of the chondriome, quantified by morphometric analysis, indicates that it is a dynamic cellular component, changing continuously during the light cycle via fragmentation, fusion and autolysis. At the onset of the light cycle (L<sub>0</sub>-L<sub>4</sub>) small mitochondria predominate. At L<sub>6</sub>-L<sub>8</sub> the average number of mitochondrial segments decreases significantly and giant mitochondria appear via fusion, bearing clear foci and disorganized cristae. During L<sub>9</sub>-L<sub>12</sub> the average number of mitochondrial segments increases significantly and small mitochondria are prevalent. The significance of these results is discussed and comparisons are made with other Chlamydomonas congeners.

### DATABASE: THE MARINE MACROPHYTES OF THE NORTHEAST PACIFIC (CONT'D.)

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P This past year, I have concentrated on systematizing the retrieval of data by locality and ecology. The main difficulty with these is that, while there are a finite number of species, there are an infinite number of possible localities and ecological parameters. In the latter case, there are also numerous missing data. Of the 67,227 records in the database; 63,348 contain information about the season of the year; 24,820 information about tidal zone; 16,735 information about wave exposure; and only 10,314, information about substrate.

