

**21ST NORTH WEST ALGAL SYMPOSIUM**  
**FRIDAY HARBOR LABS FEB 23-25, 2007**

**ABSTRACTS**

**POSTERS**

**PHYSIOLOGICAL EFFECTS OF 2,4,6-TRINITROTOLUENE ON  
MICROPLANTLETS OF THE TROPICAL RED MARINE MACROALGA  
*PORTIERIA HORNEMANNII***

Octavio Cruz-Uribe & Gregory L. Rorrer. Department of Chemical Engineering, Oregon State University, Corvallis, Oregon 97331.

The physiological response of microplantlet suspension cultures of the marine red macroalga *Portieria hornemannii* to dissolved 2,4,6-trinitrotoluene (TNT) was determined by photosynthetic oxygen evolution (OER) and biomass growth rates. The response of microplantlets to both a single TNT pulse and from continuous long-term TNT addition was determined. Microplantlets were challenged with single pulse additions at the three concentrations of 1, 9, and 21 mg L<sup>-1</sup> and monitored for seven days. For the long-term study, microplantlets were exposed to TNT through a continuous perfusion addition of 0.35 mg days<sup>-1</sup> and 2.5 mg days<sup>-1</sup> for a period of 21 days. An immediate inhibition in photosynthetic activity was observed at all concentrations of TNT addition. For single pulse additions of TNT a more significant reduction in OER and growth rates were observed with an increase in TNT concentrations. Also the recovery period from TNT exposure was longer as the concentration increased, with microplantlets no longer viable at 21 mg L<sup>-1</sup>. In long-term studies with continuous TNT addition, microplantlets exposed to the lower concentration recovered within 24 hrs and proceeded to evolve oxygen at a steady rate for the next 20 days. At the higher TNT concentration the OER did not recovery and proceeded to steadily decline for the next 20 days. In summary the results from the current investigation have shown the physiological effects and the robust ability of the red seaweed *P. hornemannii* to continuously take up TNT during continuous exposure at both low and high concentrations.

**IMPLICATIONS OF CALCIFICATION AND SURFACE-ADHERED PHOSPHORUS  
ON THE C:N:P STOICHIOMETRY OF *CLADOPHORA***

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The filamentous green alga *Cladophora* has surpassed nuisance levels in local urban streams and in Lake Michigan, resulting in a flurry of new research activity. Concrete lined storm channels and restored streams lined with limestone cobble provide excellent substrates for *Cladophora*. Such habitats, characterized by elevated nutrients and extreme light intensity, become completely dominated by *Cladophora* in early summer, with

nearly 100% spatial cover and biomass exceeding 160 mg per cm<sup>2</sup> dry weight. Problems develop when ratios of potentially growth-limiting nutrients, carbon, nitrogen and phosphorus, are applied to identify nutrient stress in this macroalga. Despite field data indicating a constant excess of dissolved phosphorus and nitrogen available to the algae, elemental analysis of the filaments indicate severe nutrient deficiencies. High photosynthetic rates of *Cladophora* can generate diurnal fluctuations in stream water pH between 7.5-9.5, and localized microzones of increased pH on the outside of filaments are sufficient to precipitate CaCO<sub>3</sub> in these streams where the alkalinity can exceed 200 mg CaCO<sub>3</sub> per liter. Our data indicates that deposition of even a thin layer of CaCO<sub>3</sub> can greatly skew *Cladophora* C:N:P ratios. Additionally, surface-adsorbed phosphorus deposition, especially associated with calcareous deposits, can skew elemental ratios by over-estimating the quantity of phosphorus actually inside the cells. This study compares the resulting C:N:P of *Cladophora* filaments treated with and without an acid rinse to remove calcification and with and without an oxalate rinse to remove adsorbed phosphorus. The effect of calcification and surface-adsorbed phosphorus on *Cladophora* photosynthetic and growth rates requires additional study.

#### CHARACTERIZATION OF MYCOSPORINE-LIKE AMINO ACID CONTENT OF RED ALGAL SPECIES ON THE CENTRAL COAST OF CALIFORNIA

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Ultraviolet radiation (UVR) reaching the earth's surface and penetrating ocean waters can have various harmful effects on intertidal organisms. Red algae of the Phylum Rhodophyta protect themselves from UVR by producing mycosporine-like amino acids (MAAs), which absorb UVR. MAA content of red algae on the Central Coast of California has not been previously investigated. Various species of red algae in San Luis Obispo County were tested for mycosporine-like amino acid content. Specimens were collected in March and April 2003 and October 2006 and tested for MAAs using high-performance liquid chromatography (HPLC). Twelve species were tested in 2003; all were found to contain at least one MAA and total MAA content ranged from 0.51±0.06 - 2.42±0.88 nmol/mg dry wt. In 2006, we characterized MAAs in additional species of red algae in greater depth. Nine species were tested, four of which were previously tested in 2003. Preliminary data indicate that at least three species contain MAAs. When available, algal specimens were collected from north-facing, horizontal, and south-facing rock surfaces. Temperature and light measurements were taken at the three exposure levels every 13 minutes for over 3 months. Quantifying the MAA content of red algae on the central coast is valuable because potential increases in UVR due to stratospheric ozone depletion make these chemical defenses increasingly important. These defenses are not only significant for Rhodophytes but also for many organisms in higher trophic levels that acquire incorporate dietary MAAs from their algal diets.

THE STABLE INCORPORATION OF GROUP IV METALS INTO THE SILICON OXIDE MATRIX OF THE FRUSTULE OF THE MARINE DIATOM *PINNULARIA SP.*

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The marine diatom *Pinnularia sp.* has a silicon oxide frustule that is structured at both the micro and nano scales. Silicon oxide materials doped with Group IV metals, such as germanium and titanium, which are structured at both the micro and nano scales, are both difficult to fabricate and hold great promise in optoelectronic and semiconductor technologies. The properties of the metal oxide composite material may be dependent on both the element being doped and the abundance of the dopant within the silicon oxide matrix. The dopant level is therefore a tunable parameter. For the diatom frustule to have any value as a novel material the organic constituents must first be removed from the organism leaving behind only the inorganic frustule. However, most wet chemical methods used to remove this organic material also solubilize the dopant, making it difficult to tune the dopant level. The stable incorporation of two dopants, germanium and titanium, into the diatom frustule are presented, as well as the current wet chemical and alternative methods used to prepare diatom frustules and to preserve the dopant within the matrix.

NEW AND RARE MARINE ALGAE OF PUGET SOUND, WASHINGTON

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Intensive floristic studies, starting in the early 1900's and emanating primarily from the University of Washington's Friday Harbor Laboratories, have resulted in approximately 600 species in the rich marine algal flora of Puget Sound. However, this flora has been largely based on collections in the rocky intertidal areas in the San Juan Archipelago and some traditional repeatedly-dredged subtidal areas. Several recent developments continue to expand this known flora: SCUBA diving, underwater video, more studies in areas such as South Puget Sound and in areas of soft substrates in salt marshes, mud flats and eelgrass beds, and the explosion of the use of molecular systematics. The flora is also changing due to the introduction of new species from ballast water, aquaculture practices, and other means of transport, and shifts in distribution due to global warming. We report several species poorly or not previously known from Puget Sound, or from only one specimen. These include *Chorda filum*, *Laminaria longipes*, *Desmarestia tortuosa*, *Chondria dasyphylla*, *Caulacanthus ustulatus*, salt marsh ecads of *Fucus sp.*, *Chondracanthus canaliculatus*, *Geldium vagum*, and *Faucheia sp.* Several species may

be cryptogenic. These studies show the importance of continuing work in alpha taxonomy to determine and protect biodiversity

#### A BLUEPRINT FOR INTERPRETING THE IMPACT OF EXOTIC INVADING INVERTEBRATES ON THE LAKE MICHIGAN PLANKTON FOODWEB

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In the past 20 years, Lake Michigan has experienced four invasions by Ponto-Caspian invertebrates that directly impact the middle links in the planktonic foodweb. *Bythotrephes cederstroemii*, a predatory cladoceran that preys mostly on daphnids appeared in 1985, followed by the zebra mussel (*Dreissena polymorpha*) with its microplankton-consuming veliger larvae in 1990. Then *Cercopagis pengoi*, another predatory spiny water flea appeared in 2001, and now a new deep water mysid shrimp (*Hemimysis anomala*) has just been discovered this past summer. Each invader has somewhat different but overlapping habitat and food requirements; all compete directly with native planktonic herbivores and invertebrate predators. Together, they may precipitate an “ecological meltdown” of the Great Lakes foodwebs. Results will be presented from an integrated network of experimental procedures developed in the early 1990’s for interpreting the effects of the first two invaders as the perturbations cascaded both up and down the planktonic food web. Data include: 1) population structure and spatial distribution of the exotic species, 2) changes in the species composition and reproductive rates of native herbivorous zooplankton, and 3) the impact of these changing predatory and herbivorous zooplankton dynamics as they influence the standing crop, nutritional status, productivity and growth rates of the phytoplankton assemblages that support the food web. These approaches represent a blueprint will be repeated over the next three years in a newly-funded Sea Grant project with the addition of immunological techniques for identifying prey species in consumer guts.

#### ANABAENA AND APHANIZOMENON: ENIGMATIC FILAMENTS

Charles R. Williams & J.Robert. Waaland. Department of Botany, University of Washington, Seattle, Washington.

Representatives of Pacific Northwest planktonic *Anabaena* and *Aphanizomenon* morphotypes were compared via direct filament PCR with representative strains from several culture collections. Phylogenetic relationships between *Anabaena* and *Aphanizomenon* were investigated using DNA sequence from *cpc* BA-IGS (cyanophycocyanin alpha and beta subunits with intergenic spacer) and *rpo* C1 (RNA Polymerase subunit C1) gene sequence. The results seriously question the current taxonomy of the Nostocaceae, and in combination with other molecular phylogenetic studies strongly suggest that *Aphanizomenon* be subsumed within the genus *Anabaena*. The diversity of *Anabaena* genotypes in Puget Sound lowland lakes will also be discussed briefly.

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## TALKS

### A COMPARISON OF TAXONOMIC AND FUNCTIONAL-GROUP DISTANCES AS PREDICTORS OF SEAWEED-INVERTEBRATE EPIFAUNA ASSOCIATIONS.

Colin Bates, Department of Botany, University of British Columbia, & the Bamfield Marine Sciences Centre.

Seaweeds provide habitat for myriad mobile invertebrate epifauna. Previous investigations of these host-epifauna associations have shown that seaweed morphology can be used as a predictor of epifauna diversity, but several, more general, frameworks remain untested. As phylogenetic information becomes more readily available, community ecologists are increasingly using evolutionary history to generate hypotheses about species performance in an ecological context. Yet, the effectiveness of taxonomic relatedness as a predictor of seaweed-host associations is uncertain because of widespread convergent evolution in seaweed form. However, this potentially confounding convergence may prove useful in the application of functional-form groupings, which are designated independently of taxonomic affinity; functional form groups may provide a more general framework for using seaweed morphology as a predictor of seaweed-invertebrate habitat interactions. To test the relative efficacy of seaweed taxonomic relatedness and functional-group membership as predictors of host-use by invertebrate epifauna, I sampled invertebrate assemblages from over 1600 individual thalli across 30 species of seaweed host. I will present results of these comparisons, and discuss the merits of each approach in the context of understanding the role of seaweeds as facilitators of intertidal biodiversity.

### SPATIAL SUBSIDY OF DRIFT ALGAE SUSTAINS DEEP SUBTIDAL RED URCHINS

Kevin Britton-Simmons, Gerard Foley, Daniel Okamoto and Elizabeth Bloczynski\*  
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The red sea urchin *Strongylocentrotus franciscanus* plays a key role in structuring subtidal marine communities and has substantial economic importance as a commercially harvested species. Although the vertical distribution of red urchins extends to depths of more than 100 m most of what we know about the ecology of this species comes from studies in the shallow subtidal. We carried out a comparative study to address two primary questions: 1) What is the diet of deep subtidal red urchins in the San Juan Islands?, 2) Is there a reproductive cost to using deep habitats (due to food limitation) in this region? We studied red urchins in shallow, algal dominated habitats (-5 m) and deep, invertebrate dominated habitats (-23 m) at 5 sites in the San Juan Islands for one year. Our results indicate that deep subtidal red urchins feed almost exclusively on drift algae, a resource that is spatially and temporally variable. Our analysis of gonad mass, and numerous morphometric characters indicate that deep subtidal red urchins are not food-limited and do not have reduced fecundity, contrary to our expectations. Our results

suggest that the deep subtidal is a high quality habitat for red urchins and that deep populations may contribute substantially to the pool of larvae that replenishes deep and shallow habitats alike. Deep subtidal habitats may be an important refugia from harvesting and could play a key role in sustaining the red urchin fishery in the long-term.

#### THE MECHANICS OF DELAYED DEVELOPMENT IN THE MICROSCOPIC STAGES OF KELP

Laura T. Carney and Matthew S. Edwards. Department of Biology, San Diego State University, San Diego, California.

For some kelp species, population persistence may depend on the presence of a bank of microscopic propagules that delay development during unfavorable conditions and then reproduce once conditions improve. Little is known, however, about the processes that regulate delayed development of these stages. We conducted laboratory experiments to determine the importance of nitrate concentration ( $1-100 \mu\text{mol L}^{-1}$ ), irradiance ( $<10-60 \mu\text{mol photons m}^{-2}\text{s}^{-1}$ ) and seeding density ( $10^2-10^5 \text{ spores ml}^{-1}$ ) in regulating development of *Macrocystis pyrifera* and *Pterygophora californica* microscopic stages. Our results suggest that nitrate concentration is the most important cue in delaying development and inducing reproduction of the microscopic gametophytes. When nitrate was increased for gametophytes cultures  $<1$ , 14 and 37 days after being grown under limiting conditions, the time to reproduction decreased with length of delay for *Macrocystis* but not for *Pterygophora*. Further, while the minimum number of days to first egg and blade production increases with spore density, delaying development may allow gametophytes to escape the effects of this density dependence. Our repeated culture work with the delayed stages of *Macrocystis* shows that initially delaying the gametophytes increases reproductive success once development is resumed, suggesting that spores attained from the field may not always be prepared for reproduction and that a delay allows this to develop.

#### NEW TECHNOLOGY STREAMLINES CHLOROPLAST GENOME ANALYSIS.

Rose Ann Cattolico<sup>1,2</sup>, Michael Jacobs<sup>3</sup>, Yang Zhou<sup>3</sup>, Jean Chang<sup>3</sup>, Han Ong<sup>1,2</sup>, John McKay<sup>2</sup> and Gabrielle Rocap<sup>2</sup>. 1. Biology Department, University of Washington; 2. Department of Ocean Sciences, University of Washington; 3. Department of Genome Sciences, University of Washington

Stramenopiles represent a major branch in the Tree of Life, whose diverse members contribute extensively to the maintenance of global ecosystem function. Unfortunately, little data is available concerning the genetic complement of this important taxon. Our study targets sequencing thirty chloroplast genomes of these organisms, including picoplanktonic as well as macroalgae representatives. Fourteen different stramenopile classes are being examined. We have streamlined the chloroplast sequencing process by developing a fosmid-based cloning approach. This technique eliminates the need to

recover highly purified chloroplast DNA from total cellular DNA, which often represents a limiting first step in organelle DNA analysis. Aspects of the completed *Heterosigma akashiwo* (strain CCMP 452) chloroplast genome sequence will be discussed, and this data will be compared to those obtained from *Aureoumbra lagunensis*, *Heterosigma akashiwo* (strain NIES 293), *Nannochloropsis oculata*, and *Pinguicoccus pyrenoidosus* (e.g., repeat elements, indels, gene co-linearity, unique genes). Data will also be discussed with respect to the three diatom chloroplast genome sequences that are presently available.

#### A NEW SPECIES OF HALOSPHERA FROM NORTHWEST WASHINGTON

Robin B. Kodner. Department of Organismic and Evolutionary Biology, Harvard University, Cambridge, MA.

I am reporting a new species of phycomite prasinophyte from the genus *Halosphaera*. Collected in the phycocyst stage, these green algae are the dominant phytoplankton in the winter in the waters around the San Juan islands. The new species has been assigned based on rosette patterning and flagellate scale morphology. Molecular analysis of 18S ribosomal DNA agrees that this *Halosphaera* species is different than the known *Halosphaera* species. Little is known about the ecology of these phytoplankton, however, they may be represented in the fossil record as far back as 1.6 billion years ago. This new *Halosphaera* species has been used for chemical and morphological comparisons of some ancient putative phycocyst fossils.

#### *ACROSIPHONIA* (CHLOROPHYTA) – COPING WITH EXTREME HETEROMORPHY.

Andrea Sussmann and Robert DeWreede. Department of Botany, University of British Columbia, Vancouver, Canada.

*Acrosiphonia* (Chlorophyta) is found on the rocky intertidal shores of British Columbia and alternates between a haploid filamentous gametophyte growing on rocks, and a diploid unicellular sporophyte ('*Chlorochytrium*' and '*Codiolum*') endophytic in bladed or crustose red algae. Life history theory proposes that heteromorphic life histories such as shown by *Acrosiphonia* decrease competition between phases, and are adaptive to predictable biological and seasonal changes. We predict that once heteromorphy occurs, reversion to isomorphy is not favored. However, the requirement for a host by the unicellular phase also entails risk, and we investigated whether *Acrosiphonia* requires a host for completion of its life history, and the exclusivity of the two phases. By carefully mapping the *Acrosiphonia* filamentous phase we determined whether a filament arose from a spore or from vegetative regeneration from last year's filament. In turn, these data enabled us to evaluate the relative success of these two types of regeneration, the potential advantage of early vs. later recruitment, and the influence of substratum on survival of the filamentous gametophytic phase of *Acrosiphonia*. Our results show that

approximately 50% of the current year's filaments arise from vegetative remnants of last year's filaments, thus bypassing the diploid sporophyte phase and its requirement for a host species. Filaments arising from such remnants survive significantly longer than those filaments arising from spores. Earlier filamentous recruits survive significantly longer than later ones and, frequently, filaments growing among articulated coralline algae survive longer than those growing on bare rock.

#### *CERAMIUM* IN THE PACIFIC NORTHWEST: HOW MANY SPECIES?

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Currently nine *Ceramium* species are recognized as occurring in the Northeast Pacific north of California. Two of these species, *Ceramium pacificum* (Collins) Kylin and *C. washintoniense* Kylin, are conspicuous and readily recognized by prolific spine-like side branches throughout the thallus. *Ceramium pacificum* (Collins) Kylin is widely distributed on the western coastlines of both North and South America while *C. washintoniense* Kylin has a narrower distribution and is reported only from British Columbia to Oregon. Kylin differentiated these two species based on differences in thallus color, size, and degree of cortication. Due to the seemingly plastic nature of these particular characters, these two species have been hypothesized to be conspecific. Phylogenetic analysis of nuclear (*RPB1*), chloroplast (*rbcL*), and mitochondrial (*cox 2-3* spacer) genes from material collected from the type localities of both species supports the hypothesis that they are one species. In addition, specimens from north of Oregon that were originally identified as *C. codicola* J. Agardh also were found to be *C. pacificum*, calling into question the presence of this species north of Oregon. These data along with data showing the phylogenetic placement of these species with respect to other Pacific Northwest *Ceramium* species will be presented.

#### UPTAKE AND METABOLISM OF POLYCYCLIC AROMATIC HYDROCARBONS (PAH) BY THE MARINE GREEN ALGA *ACROSIPHONIA COALITA*

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Polycyclic aromatic hydrocarbons (PAH) are pollutants commonly found in marine environments. PAH compounds are known to have toxic, mutagenic, and carcinogenic effects on both marine organisms and humans. These compounds do not readily degrade in the environment, and they often bioaccumulate in marine organisms. The goal of this research is to determine the ability of tissue cultures of the marine green seaweed *Acrosiphonia coalita* to take up and metabolize PAH from seawater. In general, seaweeds are known to tolerate and take up a wide range of organic compounds. However, little work has been done specifically with PAH compounds. Ongoing research indicates that *Acrosiphonia coalita* has the ability to remove PAH compounds

naphthalene and phenanthrene from seawater, but does not appear to metabolize the intracellular PAH compounds.

#### DIEL VARIATION IN PHOTOSYNTHETIC PERFORMANCE OF GIANT KELP AT DIFFERENT DEPTHS

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The giant kelp, *Macrocystis pyrifera*, is the dominant subtidal algal along much of the west coast of California, USA and Baja California, Mexico. Unlike many other species, giant kelp's morphology results in parts of the thallus occurring in low light conditions near the benthos while other portions of the thallus occur in high light conditions at the surface. As a result, giant kelp's thallus is exposed to extreme spatial and temporal variation in light quantity that may result in differential photoadaptation in various parts of the thallus. We used Diving PAM to examine diel changes in giant kelp's photosynthetic performance at three depths over an 18 hr period. Sampling just before dawn, mid-morning, noon, early afternoon, and just before sunset, we examined changes in photosynthetic performance (i.e. photoacclimation) in giant kelp blades near the benthos, in the mid-water, and near the surface. Rapid light curves ( $p_{max}$ ,  $I_k$ ,  $\alpha$ ) and measures of photoacclimation (photochemical quenching) show that giant kelp thalli in the mid-water and near the benthos exhibit stable photosynthetic characteristics similar to those of shade-adapted species, while thalli near the surface begin the day exhibiting characteristics of a shade-adapted species but photoacclimate and exhibit characteristics of a light-adapted species as the day progresses.

#### PHOTOACCLIMATION ALONG A VERTICAL GRADIENT IN DIFFERENT GROWTH STAGES IN THE ELK KELP, *PELAGOPHYCUS PORRA*

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The Elk Kelp, *Pelagophycus porra*, is commonly observed in deep (20-30m) water on the outer edge of Giant Kelp, *Macrocystis pyrifera*, forests in southern California and northern Baja California, but rarely occurs in shallower water or within the giant kelp beds. We used a series of transplant experiments, demographic monitoring, and physiological measurements to investigate *P. porra*'s apparent inability to encroach into the more abundant giant kelp beds along the southern California coast. Our results indicate that transplanted *P. porra*'s juveniles exhibit similar growth and survival across a vertical gradient. Physiological measurements using PAM fluorometry indicate that while this species exhibits characteristics of a species adapted to low light environments, such as deep-water habitats, individuals are able to photoacclimate to increasing light

levels as they grow towards the surface. This ability to photoacclimate is variable within the large (up to 20m long) blades, with portions near the surface behaving like light-adapted species and parts hanging in deeper water behaving like shade-adapted species. These results show that this deep-water species has the capability to function as both a low light and a high light adapted species as it grows through the water column as well as simultaneously within a single blade.

#### WHY SEQUENCE TYPE AND HISTORICALLY IMPORTANT SPECIMENS? LESSONS FROM *PRIONITIS* (HALYMENIACEAE, RHODOPHYTA)

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Four species of *Prionitis* were reported from the NE Pacific at the beginning of the 20th Century, three with type localities within that area, *P. angusta* (Harvey) Setchell [currently known as *P. filiformis* Kylin], *P. lanceolata* (Harvey) Harvey and *P. lyallii* Harvey, and one from Peru, *P. decipiens* (Montagne) J. Agardh. One hundred years later, eight are reported, six with type localities within that area, the three listed above plus *P. australis* (J. Agardh) J. Agardh, *P. linearis* Kylin and *P. simplex* Hollenberg *et al.* A. Abbott, and two from Japan, *P. angusta* (Okamura) Okamura and *P. cornea* (Okamura) E. Y. Dawson. Based on molecular "fingerprints", none of the species with type localities outside the NE Pacific are present, and two earlier names exist for *P. lyallii*, *P. sternbergii* (C. Agardh) J. Agardh and *P. jubata* J. Agardh. The distributions of nearly all the species are off by hundreds of miles, obscuring not only our understanding of their biogeography but also their ecology. Our ability to obtain DNA sequences from herbarium specimens of seaweeds adds immeasurably to their value, not only for systematic studies, but also for biogeography, ecology, population biology and invasive species studies.

#### BIOFUNCTIONALIZATION OF *CYCLOTELLA SP.* FRUSTULES

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The conjugation of proteins, peptides and other biomolecules to silica substrates is an emerging technology that has an array of applications ranging from drug delivery systems to biosensors. Biogenic silica derived from diatoms is an attractive substrate for these applications because its surface is populated with SiOH groups which are amenable to amine functionalization. Furthermore, diatom frustule may serve as a template to pattern biomolecules onto a silica surface at the nanoscale and microscale. In this study, silica frustules of the centric marine diatom *Cyclotella sp.* were biologically functionalized by a two-step process. In the first step, *Cyclotella sp.* biosilica frustules were covalently functionalized with 3-aminopropyl trimethoxysilane. The presence of amine groups on the frustule surface were validated with the fluorescent probe fluorescamine. In the second step, the amine-functionalized diatom biosilica was biotinylated. Biotinylation was confirmed by binding of the amine-biotin complex with Avidin that was

fluorescently labeled with FITC. This allowed for direct observation of the microscale patterning of biomolecules on the diatom frustule by epi-fluorescence microscopy. This two-step process for functionalization of diatom biosilica affords a new way to ornately pattern biological molecules onto a template with high reproducibility at the nano and microscales.

#### WILLAPA BAY AND ITS SEAWEEDS

Gayle I. Hansen<sup>1</sup> and Brett R. Dumbauld<sup>2</sup>. <sup>1</sup>Oregon State University and <sup>2</sup>the U. S. Dept. of Agriculture, Newport, Oregon

Located in the SW corner of Washington, Willapa Bay is a large muddy embayment with little hard substratum other than a few man-made structures and bivalve shells. It would seem to be fairly inhospitable to seaweeds except for a few estuarine species. However, it is the nation's largest commercial oyster producer, and juvenile oysters have been imported into the bay for cultivation since the late 1800's, providing ample opportunity for introductions. During this initial survey of the seaweeds in Willapa Bay, we have found a variety of native species that also occur in estuaries in Oregon, but we have also found a suite of introductions. Some of these are well known in the area, and others are not so well known. The introductions include *Sargassum muticum*, *Ceramium kondoi*, *Codium fragile* subspecies *tomentosoides*, *Callithamnion corymbosum*, and possibly two varieties of *Dasya sinicola*. These and other seaweeds found in the bay will be discussed.

#### INTERTIDAL ALGAE AS SENTINELS OF ENVIRONMENTAL CHANGE.

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Because intertidal organisms live close to their environmental tolerance limits, intertidal species are considered early harbingers of the effects of global change. Seaweeds will be influenced by the direct effects of a variety of climatic factors, including temperature, carbon dioxide, sea-level change, UV radiation. Although we are beginning to understand the effects of these factors individually, many important ecological responses to climate change will depend on interactions among variables. Furthermore, because climate change may differentially affect competitors and consumers, changes at the community level will dictate the ultimate outcomes of climate change. In addition to summarizing the available literature, I will present a case study of how climate change may alter the distribution and abundance of *Mazzaella parksii* (formerly *Mazzaella cornucopiae*), a turf-forming red alga. Long-term data from Tatoosh Island, WA, suggest that the upper limit of *M. parksii* can remain relatively constant for over a decade. However, during the 28 year time series, the upper limit of *M. parksii* on Tatoosh Island has shifted downshore by approximately 25 cm, virtually all of which occurred in two sudden jumps. The precise step-like pattern of change is related to stochastic environmental conditions, namely the coincidence of hot weather and calm seas on consecutive days. Although the

upper limit of *M. parksii* is sensitive to increasing abiotic stress, the lower limit, which is set by herbivores, may not be. Thus, species such as *M. parksii* may become locally extinct as the combination of abiotic and biotic factors reduce their livable space in the intertidal zone.

#### DO WATERBORNE CUES FROM A LITTORINE SNAIL INDUCE MORPHOLOGICAL OR CHEMICAL RESPONSES IN *FUCUS DISTICHUS*?

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The intertidal rockweed *Fucus distichus* responds chemically and morphologically to mechanical damage from herbivores. Recent evidence suggests that a related species, *Ascophyllum nodosum*, responds chemically to waterborne cues that indicate the presence of herbivores. To determine whether *F. distichus* responds chemically or morphologically to chemical cues from herbivores we grew juvenile *F. distichus* in outdoor mesocosms for 70 days from August to October in the presence of waterborne cues from: 1) the snail *Littorina sitkana* feeding on adult *F. distichus* (experimental treatment), or 2) adult *F. distichus* (control). We measured a variety of morphometric characters, growth, tissue C and N, and phlorotannin levels, of all *F. distichus* thalli. There was no significant difference between treatments in morphology, growth, tissue C or phlorotannin levels, suggesting that juvenile *F. distichus* either cannot detect or do not respond to chemical cues indicating the presence of *L. sitkana*.

#### USING HERNARIA TO DO ECOLOGY: SEAWEED DYNAMICS IN WESTERN NORTH AMERICA

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Biological collections have played an important role in the development of biology, but their underappreciated value today is reflected, at least in part, by momentum away from natural history and organismal biology in academics. Despite this trend, ecologists can profitably use collections to advance population and community ecology, phenology, and systematics. I discuss both the potential utility and the limitations of using historical specimens to test ecological hypotheses. I also outline ongoing research using herbarium data to understand the temporal dynamics of kelps and rockweeds off the western coast of North America. Can records from the last 50 to 100 years be used to elucidate longer-term variation in the distribution of individual species, and thus potential changes in composition or diversity at sites across the region?

RECIPROCAL INTERACTIONS BETWEEN ALGAE, BACTERIA AND THE ENVIRONMENT MEDIATE THE TOXICITY OF A HARMFUL PHYTOPLANKTER

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Many species of phytoplankton have ecosystem-wide harmful impacts due to their ability to produce toxic levels of extracellular reactive oxygen species (ROS) such as hydrogen peroxide or superoxide. Despite the ecological and economic impacts of this toxicity, little is known about the mechanism of its production. Using the HAB species *Heterosigma akashiwo*, we investigated the interactions of the algae with their biotic and abiotic environment, and how these factors contribute to ROS production. Surprisingly, we found that the abiotic environment had perhaps a greater influence on ambient ROS levels than the state or nature of the algal cells tested. Light flux was observed to be a key determinant of ROS concentration, even in the absence of algae, prompting us to investigate acellular mechanisms of ROS generation. Metal ions are known to be photogenic of ROS and we observed different background levels of superoxide and hydrogen peroxide in different media types. Yet the correlation of harmful blooms and ROS-induced toxicity cannot be explained by completely abiotic mechanisms. We therefore sought to determine if some form of excreted factor may be responsible for photogenic ROS production. We found that cell-free conditioned media (CFCM) from cultures at bloom-like densities had significantly higher ROS levels than the cultures themselves. Further, the addition of bacteria was found to reduce the ROS production in the CFCM, potentially indicating microbial degradation of the responsible factor. CFCM were analysed for DOC content and bacteria were enumerated. The relationships between DOC, bacteria abundance, and ROS levels will be presented.

*MAZZAELLA JAPONICA*, A NEW RECORD FOR THE NORTHEAST PACIFIC

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Recently, a previously unrecognized species has appeared in abundance in beach-cast material along Vancouver Island in the northern Strait of Georgia following strong southeasterly storms. The species resembles East Coast *Chondrus crispus* in being abundantly repeatedly dichotomously branched, and local entrepreneurs have applied for and received a permit to harvest “BC moss,” as the species has been called. Molecular analysis of the *rbcL* gene and the ITS region of the nuclear ribosomal cistron in specimens from Bowser indicate that the material corresponds to *Mazzaella japonica*, a species native to the northwest Pacific (Japan, Korea and China). Because of its abundance near a major Japanese oyster growing area, we suggest that the species was introduced with that species, as were Manila clam and wireweed, among other species. We present a revised key to the species of *Mazzaella* of Southeast Alaska, British Columbia, Washington and Oregon, and we briefly discuss the placement of *M. japonica* in *Mazzaella* rather than *Chondrus*, which the species subtends in phylogenetic analyses.

LIGNIN AND SECONDARY CELL WALLS IN RED ALGAE LEND BIOMECHANICAL SUPPORT

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Lignified secondary cell walls are widely considered to be key innovations in the evolution of terrestrial plants from aquatic ancestors, preventing collapse of water-conducting vessel elements and lending biomechanical support to upright stems. Here we report our discovery of lignified secondary cell walls in the red alga *Calliarthron cheilosporioides* (Corallinales, Rhodophyta). Unlike fleshy seaweeds, *Calliarthron* fronds are calcified with flexible joints, called genicula, that allow fronds to bend and go with the flow. Genicula play a critical role in the mechanical survival of calcified fronds in wave-swept intertidal habitats. TEM micrographs have revealed that, as genicula mature, genicular cells develop secondary cell walls, ultimately resembling tracheids in terrestrial xylem. Using the DFRC method, we have detected three distinct monolignols in *Calliarthron* tissue – subunits of terrestrial lignins never described in marine algae. Histological stains suggest that lignins found in gymnosperms and angiosperms are also found in genicular tissue, but are absent from calcified intergenicular tissue. Lignified cell walls may underlie the unique material properties of genicular tissue and may have evolved convergently in *Calliarthron* and terrestrial plants as common adaptations to mechanical stress. Contrary to the current paradigm describing a terrestrial origin of lignin, our data suggest that the developmental pathways for lignin may have evolved in a common ancestor of red and green algae more than 1 billion years ago.

THE EFFECT OF SPATIAL AND TEMPORAL SAMPLING FREQUENCY ON QUANTIFYING EUTROPHICATION PROCESSES IN A HEAVILY IMPACTED ESTUARINE ECOSYSTEM

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A four year (2000-2004) campaign of monthly sampling for nutrients, chlorophyll and physical parameters in the Dublin Bay (Ireland) estuarine system revealed that the freshwater end of the estuary was hypernutrified and the saline end was seasonally nitrogen limited. With the exception of a single chlorophyll sample from the upper estuary (>100 mg Chl *a.* m<sup>-3</sup>) associated with extremely high concentrations of *Cryptomonas* sp., there was little evidence of excessive phytoplankton blooms from the monthly data. This single sample prompted a 2.5 month deployment of a moored YSI 6600 sonde, measuring physical parameters and phytoplankton fluorescence on a 30 minute basis. Deployment of the sonde elucidated intense phytoplankton blooms occurring in a highly stratified part of the estuary, which had been aliased by the larger

sampling interval. The timing of the blooms was controlled by flushing due to both river flow and the spring neap tidal cycles. At the marine end of the continuum monthly sampling revealed annual minor spring bloom events but little in the way of eutrophication,. After a shift in nutrient inputs to the system, caused by an upgrade of Dublin's major sewage treatment plant a pronounced brown discoloration of waters was observed in the intertidal waters of the north shore of Dublin Bay. This phenomenon prompted weekly sampling of this previously unsampled area revealing an intense and persistent bloom of the diatom *Odontella aurita*. The results show that the quantification of eutrophication effects on phytoplankton are strongly impacted both by spatial and temporal sampling frequency.

#### ABUNDANCE, BIOMASS AND BIODIVERSITY OF PHYTOPLANKTON AT STATION NH05 OFF THE CENTRAL OREGON COAST: 2001 TO 2005.

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Temporal variations in phytoplankton population structure were examined at station NH05 along the Newport Hydrographic (NH) Line between 2001 and 2005. This is a well studied coastal ecosystem with clearly described higher trophic levels however study of lower trophic levels and, in particular, phytoplankton population dynamics have been limited to measurements of chlorophyll *a*. Here we present results of a floristic survey of phytoplankton population structure and examine the effects of various physiochemical parameters on species composition, dominance and diversity. The central Oregon coast is known to experience regular periods of intense seasonal upwelling however periodic regime shifts due to the effects of El Nino and the Pacific Decadal Oscillation (PDO) can result in increased surface temperatures, decreased upwelling and reduced productivity. During such events the zooplankton community has been shown to shift from a pattern of low species diversity and high biomass to one of high biodiversity with accompanying low biomass (Hoof and Peterson, 2006).

#### TWO-STAGE BIOREACTOR CULTIVATION OF THE DIATOM *NITZSCHIA FRUSTULUM* PRODUCES UNIQUE FRUSTULE MORPHOLOGIES

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Diatoms are single-celled algae capable of soluble silicon uptake and biomineralization. One unusual property of diatoms is that they produce amorphous silica shells called "frustules" that possess highly-ordered nanoscale and microscale features. Ongoing work in our laboratory has shown that diatom frustules possess unique optoelectronic properties for electronic device applications, including blue photoluminescence. A two-stage photobioreactor process was used to cultivate the marine diatom *Nitzschia*

*frustulum* under controlled conditions. The morphology and optoelectronic properties of the frustules were determined at various points in the cell cycle during the two-stage cultivation process. In the first stage of cultivation, HR-TEM images showed that frustules stationary phase appeared to have a coarse surface composed of nanoparticles, whereas the frustules in the growth phase of the cell cycle appeared smooth. In the second stage of cultivation, after surge-uptake of soluble Si or Si+Ge, the frustules had two novel morphologies at stationary phase. The first morphology was “screen-like” structure where nanoparticles were uniformly deposited as a monolayer within the frustule pores. The second morphology was a slit-like structure where three or more pores in a lateral array fused together. This morphology was only induced by metabolic insertion of germanium into the diatom frustule following surge uptake of soluble Si+Ge during the second stage of the cultivation. These cell-cycle dependent morphologies also affected the intensity of blue photoluminescence from the diatom biosilica. Hence, harvesting of diatoms at specific points in a bioreactor-controlled cell cycle offers a means to control the biological fabrication of these optoelectronic materials.

#### BIOLOGICAL RESPONSE OF GIANT KELP (*MACROCYSTIS PYRIFERA*) TO DIFFERENT PHYSICAL CONDITIONS IN AND AROUND A KELP BED

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Stands of the giant kelp (*Macrocystis pyrifera*) function as ecosystem engineers, altering physical factors within and around them. Kelp in the interior of beds can experience different flow and light conditions than kelp at the edge of beds, affecting flux of nutrients and rates of photosynthesis, respectively. Measurements of flow velocities, light, temperature, and seawater nitrate concentrations tracked changes to these factors inside and outside of a kelp stand off the coast of Santa Barbara, California. Semi-monthly analyses of kelp tissue nitrate and the carbohydrate storage compound mannitol indicated physiological differences between kelp on the inside and edge of the bed, and the strength and direction of these differences was correlated to temporal variation in the physical variables. Data on frond morphometrics and elongation rates indicated that kelp grew faster and into bushier shapes at the edge of the stand relative to the interior. Differences in growth between fronds at the edge and interior of the bed were more pronounced at higher frond densities. Thus the growth and physiology of kelp reflected the extent to which its presence altered the physical environment in which it lives.

EFFECT OF KELP COMMUNITY TYPE ON FISH DENSITY OFF THE WEST COAST OF VANCOUVER ISLAND, BRITISH COLUMBIA.

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Kelps provide a major source of habitat structure for temperate-reef fishes; in fact the presence of kelp has been shown to increase fish abundance significantly. From 1994 - 1996 I tested the hypothesis that community type affects the density of two functional groups of fish species. Using SCUBA visual transect surveys I monitored the density of mid-water and benthic fish at 19 sites representing four rocky-bottom community types; those dominated by 1) giant kelp 2) bull kelp 3) stalked kelp and 4) red urchins. Mid-water fish species were significantly more abundant in kelp-dominated rather than urchin-dominated communities, but the type of kelp forest did not have a significant effect on fish density. Benthic fish density was not significantly affected by community type although fish were generally (but not significantly) more abundant in communities dominated by kelp. Fish abundance, although highly variable, did not differ significantly between years. These results support those of previous studies which demonstrate the importance of kelp as a habitat structure to fish, but differ in that kelp forest type did not affect fish density significantly.

A MACROALGAL INVADER AS A SENTINEL OF ECOSYSTEM CHANGE

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The question of whether invasions cause net increases or decreases in local or regional species richness is hotly debated. We investigated how *S. muticum* affects native diversity in macroalgal communities surrounding Bamfield Marine Sciences Centre, on the west coast of Canada. We established 48 permanent plots in the low intertidal and manipulated density of the exotic in one of two ways. To test whether effects of *S. muticum* arise through competition for space we manually removed the whole alga, or through competition for light, we removed only the annual reproductive frond. The response of native macroalgal taxa was monitored over five months. Because experimental scales can be misleading, we compared experimental results with field observations. Results suggest invasion by exotic *S. muticum* negatively impacts native diversity. Plots with high exotic cover had low native diversity, and plots with low exotic cover had the highest increases in native diversity over time. A previous study we conducted found invasibility in this system was ultimately highest in low diversity plots. Results from the current study indicate that invasion by *S. muticum* may lead to a loss of native diversity, suggesting a potential negative feedback which may facilitate further invasion.