1

SEAWEED ABUNDANCE AND DIVERSITY IN HIGH ENERGY AND LOW ENERGY AREAS AT PORT ARANSAS, TEXAS JETTIES

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Benthic algal sampling from high and low energy zones at a selected site on the south jetty at Port Aransas, Texas was completed between April 1999 and February 2000. Species composition and seasonal periodicity in relation to temperature and salinity fluctuations were determined. Dominant plants throughout the year included *Bryocladia cuspidata, Bryocladia thyrsigera, Gelidium pusillum, Centroceras clavulatum, Ulva fasciata,* and *Padina gymnospora*. The Rhodophyta dominated species coverage, along with biomass accumulation, although Chlorophyta species accrued the greatest biomass on the high energy side in April and June sampling. Chlorophyta are important to benthic coverage and biomass in the shallowest of waters, despite being fewer in species richness. Phaeophyta species including *Petalonia fascia, Hincksia mitchelliae,* and *Ectocarpus siliculosus* were found only during the cooler months. *Padina gymnospora* was the one exception as it was collected year-round. Results indicate that a greater Rhodophyta abundance was found on the channel side (low energy), where as, the surf side (high energy) contained a greater Chlorophyta abundance. Phaeophyta abundance for both high and low energy sites were comparable possibly due to the greater depth of water in which they are located. Little variance occurred in average biomass accumulation throughout the year. Highest biomass was in August (216.613g dry weight) with lowest occurring in April (107.4205g dry weight). This presentation is to be a poster contribution and Ecology will be the subject area assigned.

2

MARINE ALGAE ASSOCIATED WITH CARIBBEAN ROCKY SHORES, QUINTANA ROO, MEXICO

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Rocky shores located on the east coast of the Yucatan Peninsula are typically high-energy habitats associated with fringing or barrier reef systems. They are phytokarstic (iron-shore) consisting of fossiliferous limestone that is composed of stony coral skeletons. This study compares the macroalgae from three rocky shores located on the east coast of the Yucatan Peninsula, Quintana Roo, Mexico. Fifteen 0.25m^2 quadrates were randomly placed in the intertidal and subtidal zones and evaluated for dominant algal cover. In addition, fifteen 0.25 m^2 quadrates of standing stock material was randomly collected and used to characterize algal composition, species richness, abundance, and biomass. A voucher collection including herbarium mounts and preserved specimens were prepared for all species encountered. Thirty-five species were identified representing three divisions: Rhodophyta, Chlorophyta, and Phaeophyta. Rhodophyta had the highest species richness with a total of fifteen taxa identified. Phaeophyta had the lowest species richness with seven taxa but dominated algal coverage at two of the three sites. Chlorophyta was represented by thirteen species. Species common to all points were represented by *Digenia simplex*, *Laurencia poiteaui*, *Laurencia* sp., *Polysiphonia* sp., *Microdictyon marinum*, *Halimeda opuntia*, *Dictyosphaeria cavernosa*, *Padina sanctae-crucis*, and *Turbinaria tricostata*.

3

PHOTOSYNTHETIC EFFICIENCY AS A DIAGNOSTIC INDICATOR OF PHYTOPLANKTON PHYSIOLOGICAL STATUS IN THE PERUVIAN UPWELLING ZONE

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Upwelling off the coast of Peru produces some of the most productive oceanic waters in the world. High macronutrient (nitrate, phosphate, and silicate) concentrations from upwelled Peruvian water fuel high primary production rates. This increased primary production leads to high fisheries yields. Recent evidence from our research suggests, however, that iron limitation can limit photosynthetic production even along the Peruvian coastline. To assess the relative importance of Fe-limitation on algal growth rates along the Peru coastline, we monitored the photosynthetic efficiency of photosystem II (F_v/F_m) using a fast repetition rate fluorometer as an indicator of algal physiological state. Vertical profiles at selected stations and underway surface F_v/F_m measurements were performed on transects perpendicular to the coastline. We also monitored hydrography, phytoplankton biomass (chl *a*), species composition (HPLC pigments), and concentrations of both macronutrients and Fe during August/September 2000. In high macronutrient regions, a positive correlation was observed between iron concentrations and F_v/F_m. The usefulness for utilizing F_v/F_m as a diagnostic indicator for Fe-limitation in the Peruvian upwelling zone will be discussed.

4

THE BEGINNINGS OF ANTARCTIC MACROALGAL CHEMICAL ECOLOGY: DEFENSES

AGAINST HERBIVORES IN A NITROGEN REPLETE, CARBON LIMITED OCEAN

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We examined palatability of Antarctic Peninsula macroalgae in feeding bioassays with three common, sympatric macroalgal-consuming omnivores (amphipod, fish, sea star). Antarctic macroalgae have low C:N ratios, high nitrogen contents, and are usually growth limited by carbon (light). The Carbon Nutrient Balance Hypothesis (CNBH) predicts that macroalgae would produce nitrogenous secondary metabolites for defense rather than high levels of non-nitrogenous defenses under these conditions. To date, feeding bioassays have been performed on fragments of thallus from 26 macroalgal species and 21 (81%) were rejected by at least one omnivore. Organic extracts from 13 macroalgal species rejected as thallus were used in feeding bioassays. At least one extract from 12 species (92%) was rejected by at least one omnivore, suggesting that chemical defenses against herbivores probably are present in at least some of the macroalgal species. We have identified a number of specific, non-nitrogenous secondary metabolites in these extracts and previous workers have also reported non-nitrogenous secondary metabolites from antarctic macroalgae. Additional extracts targeting nitrogenous metabolites from 25 species were subjected to thin layer chromatography and visualized by stains specific for nitrogenous compounds. No nitrogenous secondary metabolites were identified by this or other methods. Nitrogenous secondary metabolites are also extremely rare in macroalgae from other areas of the world. Consequently, although our bioassays suggest that chemical defenses probably do occur, our data cast doubt on the applicability of the CNBH for predicting the chemical composition of macroalgal defenses under carbon limited conditions. (NSF OPP9814538, OPP9901076)

5

DINOFLAGELLATE GENOMICS

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Dinoflagellates are a distinct phylum of unicellular, primarily photosynthetic microaglae, characterized by unique cellular and nuclear morphologies. Ecologically they comprise an extremely important group of primary producers, not only as free living organisms, but also as symbionts within the cells of many invertebrates. Dinoflagellates accumulate significant amounts of lipid in the form of triglyceride for carbon storage. These lipids are predominately omega-3 polyunsaturated fatty acids, particularly EPA and DHA. Applications for these fatty acids in human nutrition, particularly for infants has developed into a significant consumer market. Elucidation of the biosynthetic pathway for these fatty acids is a major motivating force driving industry efforts for genomic research. Genome and gene structure in dinoflagellates is poorly understood. The DNA content of these organisms is extremely large for unicellular organisms, reaching an estimated 1011 Mb, which is larger than the human genome. The large genome size makes complete genome sequencing impractical, however sequencing of expressed cDNA's can be done efficiently. The limited number of nuclear encoded genes that have been characterized are present in multicopy repeats. For example, the pcp genes appear to be present in an estimated 5,000 copies. This presentation will provide an overview of both nuclear and plastid genome structure. Along with recent advancements from sequencing projects currently underway by both academic and industry efforts.

6

DIATOM GENOMICS: IMPORTANT CONSIDERATIONS FOR GENOME SEQUENCING OF ECOLOGICALLY RELEVANT ORGANISMS

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Diatoms are one of the most abundant groups of aquatic eukaryotic microorganisms found in nature, with a wide distribution in marine and freshwater habitats. These unicellular organisms are responsible for a huge proportion of global primary productivity – in fact, Mann (Phycological Rev. 38:437) recently suggested that diatoms are responsible for more carbon fixation than all the world’s rain forests. Furthermore, because of their dominance under high nitrate conditions and their enhanced sinking rates due either to aggregation and/or packaging into fecal pellets, diatoms play a critical role in the biological pump that mediates carbon and nitrogen fluxes out of euphotic zones and into deeper waters. Due to their important role in carbon sequestration, the Department of Energy’s Microbial Genome Program is initiating a diatom whole genome-sequencing project. An update on the status of this project will be provided and some of the stumbling blocks encountered in the early stages of the project will be discussed. In addition, important considerations for choosing ecologically relevant organisms for genome studies, such as the ability to perform genetic manipulations, will be highlighted. An emerging theme in the genome information accumulated thus far from a variety of organisms is that large numbers of putative genes have unknown functions. Sequence data alone is not enough to discern all the attributes of an organism – mutational analyses of genes are also critical. Since diatoms are diploid, sexual reproduction is an obligate step in mutant generation. Examples of what is known about diatom sexual cycles will be provided.

7

BROWN ALGAL POLYPHENOLS: PRIMARY METABOLITES WITH MULTIPLE, TRANSITIONAL ROLES

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Brown algal polyphenolics serve multiple functions. As defensive metabolites, they may serve to deter grazers, inhibit microbes, or absorb harmful UV radiation. However, these compounds are not classic ‘ secondary metabolites’; they also have primary functions as cell wall strengtheners, spermatozoid inhibitors, and putative elements of algal holdfasts. These civilian roles of algal polyphenolics, which have been largely ignored in studies of plant defense, have implications for estimates of polyphenol cost. For example, in studies where growth-defense tradeoffs are used to indicate a cost for a multi-functional metabolite, the true cost is likely to have been significantly overestimated. To illustrate this, I propose a mechanism by which apparent growth vs. defense trade-offs can be predicted in the absence of a true cost of algal polyphenolics or in the absence of any defensive role for these compounds. This transitional-role model is based on the progression, or “aging” of phenolics and diff-erentiates between physode-bound (reactive) polyphenols and the oxidized (unreactive) wall components. It predicts that variations in the levels of brown algal phenolics measured by colorimetry result from changes in the synthesis – secretion balance. Potential tests of this model, including pathway- and gene-level assessments of polyphenolic metabolism in brown algae, will be presented.

8

RESTORATION OF *SARCOTHALIA* AND *GIGARTINA* BEDS IN SOUTHERN CHILE

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Sarcothalia crispata and *Gigartina skottsbergii* are the most important species in volume and value for the domestic production of carrageenan in Chile. Harvesting pressure has been intensified during the last years decreasing the biomass of the natural beds. Because of the importance of this seaweed resources three restoration techniques were develop and assayed in two localities in southern Chile. Densitiy and growth of the fronds was measured in time. Results show that higher densities for *Sarcothalia crispata* were obtained in autumn through direct seeding. For *Gigartina skottsbergii* higher densities were in winter also through direct seeding. Methods develop to enhance *Sarcothalia* and *Gigartina* beds are discussed.

9

TESTING THE REVERSE-SPLICING MODEL OF RDNA INTRON ORIGIN

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Group I introns are widely distributed in the nuclear ribosomal RNA genes of algae and fungi. Two models are proposed for intron mobility. The first relies on an endonuclease open reading frame that allows intron "homing" at the DNA-level into homologous allelic sites. The second model proposes mobility at the RNA-level through reverse-splicing. Whereas endonuclease-mediated homing is depen-dent on a significant sequence requirement (14-35 nt) at the insertion site, reverse-splicing requires as little as 4-6 nt at the 5' flanking sequence to initiate intron integration. For this reason, reverse-splicing may play the more important role in intron lateral transfer in the nuclear genome. In this presentation, we test two major predictions of the reverse-splicing model with introns in the small (SSU) and large subunit (LSU) nuclear rRNA: 1) Group I introns reverse-splice into rRNA regions which contain a 4-6 nt 5'-exon flanking sequence that builds a helix with the intron internal guide sequence required for forward- and reverse-splicing. 2) Introns are non-randomly distributed, with most of them clustering in regions that are not "hidden" by rRNA tertiary structure. Phylogenetic analyses show that multiple group I introns in the SSU rDNA of lichen fungi have likely originated through reverse-splicing of one intron into a heterologous site. The reverse-splicing model is also supported by the observation that both group I and spliceosomal introns are primarily found in highly conserved rRNA regions that map on the surface of the SSU ribosome. These introns show a positive correlation with rRNA sites that form cross-links with tRNA and mRNA.

10

CHEMISTRY OF ATTRACTION AND DEFENSE IN MACRO- AND MICROALGAE

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A diverse group of brown seaweeds produce bouquets of C_{11} and C_8 metabolites, some of which act as pheromones that trigger gamete release or attract sperm to eggs following release [1]. The same compounds and, especially, their oxidative degradation products frequently and strongly deter feeding by mezograzers (*Ampithoe longimana*) which often consume seaweed spores, zygotes, and juveniles [2]. Besides macroalgae also several microalgae (ca. 20 *Gomphonema* spp., *Asterionella formosa*; Diatomophyceae) produce C_{11} and C_8 hydrocarbons along with a toxic polar compound from the pool of highly unsaturated fatty acids (eicosanoids) [3]. In biosynthetic studies with cell free extracts of the diatom *G.*

parvulum (9*S*)-hydroperoxy-eicosatetraenoic (9*S*-HPETE) acid was shown to be converted by a novel type of a lyase into the defensive compound 9-oxo-nona-5*Z*,7*E*-dienoic acid along with cyclic and linear C_{11} hydrocarbons [4]. If the eicosanoid precursors are first converted into C_{12} hydroperoxides, subsequent lyase activity produces C_8 trienes together with 12-oxo-dodeca-5*Z*,8*Z*,10*E*-dienoic acid (*A. formosa*). Both unsaturated acids serve as efficient chemical defenses against attacking feeders and micro-organisms. The reaction cascade is triggered by damage of the diatom, initiating rapid release of the free fatty acid from phospholipids followed by lipoxygenation and oxidative cleavage into the unsaturated hydrocarbon and the oxo acid [5].

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11

CHARACTERIZATION OF THE 16S-23S ITS REGIONS FROM MULTIPLE rRNA OPERONS IN SEVERAL CYANOBACTERIAL GENERA.

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We amplified, TA cloned, and sequenced the 16S-23S ITS region from single isolates of a variety of cyanobacterial taxa: *Calothrix parietina, Scytonema hyalinum, Coelodesmium wrangelii, Tolypothrix distorta*, and a putative new genus in the Microchaetaceae. All isolates were found to carry ITS regions containing the sequences coding for two tRNA molecules ($tRNA^{Ile}$ and $tRNA^{Ala}$). We retrieved additional sequences without tRNA features from both *C. parietina* and *S. hyalinum*. Furthermore, in *S. hyalinum*, we found two of these non-tRNA-encoding regions identical in length, but different in sequence. This is the first report of ITS regions from a single cyanobacterial isolate different not only in configuration but also, within one configuration, different in sequence. The potential of the ITS region as a tool for studying molecular systematics and population genetics is significant, but the presence of multiple non-identical rRNA operons poses problems. Multiple non-identical rRNA operons may impact both studies which depend on comparisons of phylogenetically homologous sequences, and studies which employ restriction digests of PCR products.

12

CHARACTERIZATION OF THE 16S rRNA GENE AND ASSOCIATED 16S-23S ITS REGION IN *MICROCOLEUS* (CYANOPHYTA).

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Thirty-one strains of *Microcoleus* were isolated from diverse arid regions in the western United States. The 16S rRNA gene of all these strains was characterized. The 16S-23S internal transcribed spacer (ITS) region of many of the strains was sequenced as well. Detailed morphological characterization of strains was carried out on a subset of the strains when molecular data indicated that more than one species was represented. Combined molecular and morphological data reveal the presence of two morphologically similar taxa in our collections, *Microcoleus vaginatus* (Vaucher) Gomont and *Microcoleus steenstrupii* Boye-Petersen. These taxa are sufficiently dissimilar in their 16S rRNA sequence that we suspect the two taxa belong in separate genera. *M. vaginatus* is phylogenetically closest to *Trichodesmium* and *Arthrospira*. *M. steenstrupii* as defined in this paper includes *M. chtonoplastes*, and may have closest relatives among several genera in the Phormidiaceae. Configurations within the 16S-23S ITS regions are variable between the two taxa. *M. vaginatus* has both $tRNA^{Ile}$ and $tRNA^{Ala}$ genes in the ITS, just the $tRNA^{Ile}$ gene, a fragment of the $tRNA^{Ile}$ gene, or no tRNA genes. *M. steenstrupii* has rRNA operons with both $tRNA^{Ile}$ and $tRNA^{Ala}$ genes, the $tRNA^{Ile}$ gene only, or no tRNA genes in their associated ITS regions. It appears that *M. vaginatus* shows no subspecific level or population level variation within the combined morphological and molecular characterizations. *M. steenstrupii* did show considerable genetic variability, and so we suspect that our strains represent several cryptic genospecies. However, the genetic variability was not congruent with geography.

13

PHLOROTANNINS AS CHEMICAL DEFENSE AGAINST MACROALGAL EPIPHYTES ON *ASCOPHYLLUM NODOSUM.*

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Phlorotannins are a group of polyphenolic acids present in brown algae. They have been shown to act as defense against herbivores and UV-B radiation. They have also been suggested to function as a defense against epiphytes. In this study the effect of the presence of macroalgal epiphytes on the phlorotannin content in *Ascophyllum nodosum* was analysed. Six adult plants of *A. nodosum* were collected from each of four sites in the archipelago on the west coast of Sweden. Four apical parts from each of the 24 plants were analysed for their phlorotannin content. For each plant, two samples were without any visible macroalgal epiphyte growth and two samples were 50-100% covered by epiphytes, mainly of the orders Ceramiales and Ectocarpales. We were not able to detect any general effect of epiphytes on the phlorotannin content. There was, however, a significant interaction between the two factors algal individual and epiphytes, where apical parts with epiphytes present had either higher or lower levels of phlorotannin than samples without epiphytes. Furthermore, there was a general, significant difference in the phlorotannin content among sites. The results suggest that the relationship between macroalgal epiphytes and phlorotannin production in *A. nodosum* is complex and influenced by several interacting factors.

14

BENCH–SCALE TESTING OF IMMOBILIZED MICROALGAL BIOMASS FOR HEAVY METAL BINDING

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This bioresin technology utilizes immobilized non-living biomass derived from algae to bind heavy metals from dilute solutions. There is significant need for such technology as contamination of water supplies is a broad area of concern in former mining operations, industrial sites, groundwaters and surface waters. Bench scale-tests used immobilized biomass to assess heavy metal binding. Column efficiency was high with 2-10 ppb of copper being passed through these columns in the first 25-50 bed volumes of influent water with 10 ppm Cu^{2+}. Reproducibility within batches and over time was adequate, although measurable variations among batches of bioresins were evident. HCl concentrations of 0.0024 N and above were required to elute copper (pH $\leq$ 2.6) from the columns, and the less expensive sulfuric acid was also demonstrated to be effective. Binding of Cu^{2+} at 10 ppm was largely unaffected by roughly equimolar Fe^{2+}, as copper had a particularly high affinity for the tested bioresin. The process appeared to be cation exchange as the oxyanion selenate was not bound. Cost of processing/1000 gal. was calculated to be $0.42, of which $0.31 was system capital and the remainder operating cost for a base case of 10 ppm Cu^{2+} and 600 gpm.

15

EVOLUTIONARY MORPHOLOGY OF THE EUGLENID CHLOROPLAST: TAXONOMIC IMPLICATIONS.

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The euglenophytes are a monophyletic assemblage within the Euglenida. The use of chloroplast morphology as a tool for taxonomy is nearly as old as euglenid systematics. Different taxa have been united based on the morphology of the chloroplast margin, the structure of the pyrenoid (if present), and the relative size and number of plastids. The previous taxonomies based on chloroplast morphology have not, however, had the benefit of modern ultrastructural and molecular biological techniques to aid in their evaluation. The current study uses morphological characters as discerned from confocal laser scanning microscopy (CLSM) and transmission electron microscopy (TEM) to discern monophyletic sub-groups within the Euglenophyta (sensu- stricto). These hypotheses are being independently tested with molecular characters in the form of intron content in the plastid encoded gene rbcL. The current project is a pilot study using chloroplast ultrastructure in six species of euglenophytes from four genera to assess the validity of a heretofore unidentified group. Initial observations suggest that a monophyletic group with an inward projecting pyrenoid, capped by a cupped shape paramylon grain exists within the euglenophytes. We propose to name this group the *Mucoglena* based on an apomorphic secretion of mucilage unlike that seen in other euglenophytes.

16

THE USE OF SEAWEEDS IN INTEGRATED AQUACULTURE: THE CHILEAN EXPERIENCE

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In Chile, salmon cultivation is well established, and produces over 200,000 tons yr-1. Integrated aquaculture has been proposed as an environmentally friendly way of recycling the waste of intensive aquaculture practices. An initial approach has been considered which contemplates the use of culture lines surrounding fish cages. Several studies have demonstrated under these conditions seaweeds can maintain higher production levels and present higher nitrogen contents as compared to cultures without the presence of fish cages. However, success has not been total, partly because the amount of seaweeds required to remove a significant proportion of the waste produced from intensive large-scale cultivation systems is very large. In this context, the use of a multi-layered seaweed cultivation of *Gracilaria* and *Macrocystis* is suggested. On the other hand, land-based systems have been proposed as a technological alternative for integrated aquaculture. The latter type of systems are technically feasible, although, the high level of investment required at present. Our results indicate that *Gracilaria* is capable of removing a significant proportion of the ammonium excreted by fish. These studies indicate that annual fish production can reach over 30 kg.m-3, with an associated *Gracilaria* production of 49 kg (wet). m-2. y-1. Finally, the environmental benefits associated with the development of integrated tank cultivation were assessed indicating that by integrating seaweed cultivation into a fish farm, the economic profitability of a commercial fish operation is not affected when internalizing the environmental costs which could be incurred for environmental compliance. (Grant: FONDEF D991/1101)

17

THE CYANOTOXINS-BIOACTIVE METABO-LITES OF CYANOBACTERIA: OCCURRENCE, ECOLOGICAL ROLE, TAXONOMIC CONCERNS AND EFFECTS ON HUMANS

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Cyanobacteria toxins (cyanotoxins) include cytotoxins and biotoxins with cytotoxins including about 60 compounds ranging from phytoalexins to animicrobials to enzyme inhibitors to compounds that can reverse multidrug resistance. Producer organisms include marine/brackish water *Cystoseira, Hormothamnin, Lyngbya, Nodularia* and *Synechocystis,* and the freshwater/terrestrial genera *Anabaena, Dichotrix, Fischerella, Hapalosiphon, Lyngbya, Microcystis, Nostoc, Oscillatoria, Planktothrix, Phormidium, Schizothrix, Scytonema, Spirulina, Stigonema* and *Symploca.* Since many of these compounds have been identified, not during ecological studies, but during drug discovery investigations, their ecological role is only speculative. Biotoxins are responsible for acute lethal, acute, chronic and sub-chronic poisonings of wild/domestic animals and humans. They include the neurotoxins; anatoxin-a, anatoxin-a(s) and saxitoxins plus the hepatotoxins; microcystins, nodularins and cylindrospermopsin. These compounds are included when referencing harmful algal blooms (HAB's) such as the more predominate marine PSP (paralytic shellfish poisoning), DSP (diarrhetic shellfish poisoning), NSP (neurotoxic shellfish poisoning), ASP (amnesic shellfish poisoning) and EAS (estuary associated syndrome). The CTP (cyanobacteria toxin poisoning) organisms occur in freshwater lakes, ponds, rivers and reservoirs throughout the world. Organisms responsible for CTP's are *Anabaena, Aphanizomenon, Cylindrospermopsis, Microcystis, Nodularia, Nostoc Oscillatoria (Planktothrix), Trichodesmium* and certain picoplanktic genera. Concern for animal and human health impairments arises from animal poisonings, associated with cyanobacteria waterblooms, beginning with the later part of the 1800's. It was not until the 1950's that we began to understand that cyanobacteria could indeed produce highly toxic compounds. A recent 1998 compilation of all available information on toxic cyanobacteria was published by the World Health Organization. This increasing focus on the role of cyanobacteria metabolites in chemical ecology, drug discovery and toxinology has placed new importance on using correct taxonomy for communication of responsible organisms.

18

EXPLORING *PORPHYRA* SPECIES FOR USE AS NITROGEN SCRUBBERS IN INTEGRATED AQUACULTURE

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Finfish mariculture along the Northeast US coast continues to develop into a strong industry. At a regional level, mariculture can be a significant contributor to nutrient loading in coastal waters. Since macroalgae are able to concentrate nutrients and grow at high rates, they can be an useful tool for alleviating this problem. In addition, seaweed mariculture is by itself a multi-billion dollar industry, with the red alga *Porphyra* (nori) valued at over $US 1.8 billion. Local species and strains of *Porphyra* from the Northeast U.S.A. are being studied to determine their capacity as nutrient scrubbers under different nutrient and temperature conditions. *P. purpurea* was grown under two N sources (NO3- vs. NH4+). The fastest growth (up to 13% d-1) and greatest N content (ca. 7% DW) were measured in plants grown at 300 µM NH4+. Short-term NH4+ uptake by *P. purpurea* (strains from Maine and Long Island Sound) and by *P. amplissima* was not saturated at 150 µM, the highest concentration tested. The *P. purpurea* isolate from Maine took up NH4+ faster than did the Long Island Sound isolate. NH^{4+} uptake by *P. amplissima* was faster than uptake by either *P. purpurea* strain. The high growth rates obtained and the ability for N uptake and tissue accumulation make these species suitable for using as a biological nutrient removal system.

19

POPULATION GENETICS OF *PHORMIDIUM RETZII* (CYANOBACTERIA) MATS IN A HEADWATER STREAM USING *RAPD* ANALYSIS.

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Unidirectional flow of water in lotic ecosystems greatly impacts the gene flow of species, especially non-motile organisms. This research addresses the genetic variation of the cosmopolitan cyanobacterium *Phormidium retzii* on a spatial and temporal scale in a headwater stream. Ten permanent plots were demarcated, with the first five spaced 1m apart and the second five 20m from the first set. Individual mats of *P. retzii* were collected from each plot throughout the growing season (3 sample trips). Random amplified polymorphic DNA (RAPD) markers were employed to examine genetic similarity. The eight primers utilized yielded 87 bands among 29 samples. Distance analysis revealed five groupings, which were not necessarily correlated with sample date or by stream location (upstream vs. downstream). Eight of the 10 samples from the final collection date did group together, to the exclusion of all others. Principle Coordinate Analysis (PCO) revealed two clusters, one of which included all of the samples from the last collecting date to the exclusion of others. Analysis of Molecular Variance (AMOVA) did not show any significant differences between upstream vs. downstream sites, but detected significant ($p<0.05$) differences in genetic variation between the three sample dates. The genetic variance among dates was 86% and within a given date 14%. We speculate that the genetic similarity of mats on the final date may be from a dominant population upstream with a greater growth rate and samples from the first two dates represent cryptic populations from the previous year.

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PHRAGMOPLAST-MEDIATED CYTOKINESIS IN NON-CHAROPHYCEAN GREEN ALGAE—IS IT HOMOLOGOUS TO THE PROCESS IN STREPTOPHYTES?

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Phragmoplast-mediated cytokinesis, basic to all land plants, occurs in three orders of charophycean algae--the Coleochaetales, the Charales, and the Zygnematales--but not in the Chlorokybales and Klebsormidiales. Phragmoplast-mediated cytokinesis has also been found in two genera of the Trentepohliales, an anomaly for chlorophycean algae. Within the green algal orders where phragmoplasts have been found, not all genera studied have this mode of cytokinesis. Among the genera that do have it, there is significant variation in the mechanisms involved. In the Trentepohliales, the temporal separation of karyokinesis and cytokinesis, the massive "double-cone" microtubule array, and the centripetal fusion of cell plate vesicles are distinct features that indicate that on a structural basis the phragmoplast-mediated cytokinesis is not homologous to that in the charophycean green algae or the land plants. However, given the structural differences in this process among the charophycean green algae and between them and the land plants, one wonders if structural variations are relevant to the question of homology. We propose that in the Trentepohliales, differences in the details of the vegetative cytokinetic processes and the microtubular structures from those found in the streptophytes are great enough to indicate parallel evolution of phragmoplast-mediated cytokinesis in these groups. The putative combination of cell wall infurrowing with a central phragmoplast in *Phycopeltis* can be interpreted as support for the nonhomology of the Trentepohlialean phragmoplast-mediated cytokinesis and that in the streptophytes. Evolutionary and developmental studies within the chlorophycean and streptophyte evolutionary lineages are needed.

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MOLECULAR EVIDENCE IDENTIFIES BLOOM-FORMING *PHAEOCYSTIS* (PRYMNESIOPHYTA) FROM COASTAL WATERS OF SOUTHEAST CHINA AS *PHAEOCYSTIS GLOBOSA*

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Sequence data from the 18S small subunit ribosomal RNA gene and rDNA ITS regions have been used to identify the species of a *Phaeocystis* (Prymnesiophyta) that caused harmful algae blooms in the coastal waters of southeast China. This *Phaeocystis* has morphological and physiological features that differ from those previously described for either *P. globosa* Scherffel or *P. pouchetii* (Hariot)Lagerheim. However, the sequence comparison of the *Phaeocystis* 18S rDNA and rDNA ITS clearly showed that it was remarkably similar to several isolates of *P. globosa*. Thus, the species isolated from the southeast coast of China is identified as *P. globosa* rather than *P. cf. pouchetii* or another species. Our results also demonstrate that phenotypes of different members of the genus *Phaeocystis* are variable, apparently changing in response to environmental conditions. It is concluded that, on the basis of this phylogenetic analysis, the bloom forming southeast China coast species of *Phaeocystis* most likely originated from an endemic warm-water, rather than a foreign source.

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AGROBACTERIUM-MEDIATED GENETIC TRANSFORMATION IN THE MACROSCOPIC MARINE RED ALGA *PORPHYRA YEZOENSIS*

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Amongst marine algae, stable genetic transformation has only been accomplished in a few unicellular species (i.e. a few diatom and dinoflagellate species). We report on the stable genetic transformation of a multicellular marine red alga or seaweed, *Porphyra yezoensis*. *Porphyra*, also known as nori, which is one of the most widely eaten and cultivated seaweeds in the world. It is also one of the most ancient seaweeds and multicellular eukaryotes that exists today, being at least 500 million years old. Our method of gene transfer is unusual in that it utilizes *Agrobacterium tumefaciens*, which has not heretofore been used to transform any alga. Transformation has been confirmed using both GUS (with and without an intron) and GFP reporter genes coupled to either a heterologous cauliflower mosaic virus (CaMV) 35S promoter gene or one of two homologous reporter genes (RPB1 and GAPDH). Transgene expression has been observed in primary transformants for more than an eight-month period, as well as in progeny through the T1 and T2 generations. Evidence of stable genetic transformation will be presented. This gene transfer system permits for the first time the study of red algal promoter gene structure and function. (Supported by NSF)

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ANATOMICAL AND NUCLEAR SSU rDNA SEQUENCE DATA INDICATE THAT THE DASYACEAE AND DELESSERIACEAE ARE POLYPHYLETIC

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The aim of the current investigation was to test the general convention that the Dasyaceae, Delesseriaceae and Rhodomelaceae (Ceramiales, Rhodophyta) are all monophyletic. Phylogenetic relationships among 45 ceramialean taxa, including eight ceramiacean, 18 dasyacean, nine delesseriacean, eight rhodomelacean species and two of uncertain affiliation, based on 34 anatomical characters and nuclear small-subunit (SSU) rDNA sequences were determined. Results from our 'total-evidence' approach were consistent with the notion that the Dasyaceae, Delesseriaceae and Rhodomelaceae have evolved from a common ancestor within a paraphyletic Ceramiaceae. Our data indicate, however, that the Rhodomelaceae alone was monophyletic with the Dasyaceae unequivocally polyphyletic and the Delesseriaceae either polyphyletic, or paraphyletic in excluding the *Dasya*-group. Based on our results the Heterosiphoniaceae fam. nov. H.-G. Choi, Kraft, I.K. Lee et G.W. Saunders is proposed for the *Heterosiphonia*-group, previously placed in the family Dasyaceae. Although the Dasyaceae *sensu stricto* is a natural group, it is in need of a thorough systematic investigation at the generic level. Our analyses indicate that the genus *Dasya* is polyphyletic in excluding *Dasysiphonia*, *Eupogodon* and *Rhodoptilum,* and *Heterosiphonia japonica* has affinities to this group as well.

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UNDERUTILIZED TOOLS: SEAWEEDS AS BIOREMEDIATION AND DIVERSIFICATION TOOLS AND BIOINDICATORS FOR INTEGRATED AQUACULTURE AND COASTAL MANAGEMENT

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On a regional scale, finfish aquaculture can be one of the significant contributors to coastal nutrification. Contrary to common belief, even in regions of exceptional tidal and apparent "flushing" regimes like the Bay of Fundy, water mixing and transport may be limited and water residency time can be locally prolonged. Hence, nutrient bio-availability remains significant for a relatively long period of time in some areas. Understanding the assimilative capacity of coastal ecosystems under cumulative pressure, then, becomes critical. To avoid pronounced shifts in coastal processes, conversion, not dilution, is the solution by integrating fed aquaculture (finfish) with organic and inorganic extractive aquaculture (shellfish and seaweed) so that the "wastes" of one resource user become a resource for the others. Such a bioremediative approach provides mutual benefits to co-cultured organisms, and economic diversification and increased profitability per cultivation unit for the aquaculture industry. These concepts will be discussed and illustrated by the results of our on-going projects and we will demonstrate that seaweeds can also be excellent bio-indicators of nutrification/eutrophication revealing symptoms of environmental stress and measuring the zone of influence of an aquaculture site. The aquaculture industry is here to stay in our "coastal scape": it has its place in the global seafood supply and demand, and in the economy of coastal communities. To help ensure its sustainability, it needs, however, to responsibly change its too often monotrophic practices by adopting polytrophic ones to become better integrated into a broader coastal management framework.

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RESPONSE OF FRESHWATER FLAGELLATES TO ENVIRONMENTAL GRADIENTS

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Like plants, photolithotrophic flagellates require light, inorganic carbon, nutrients and a suitable oxygen concentration to survive and yet, paradoxically, like animals they are motile with the potential to select their position within the water column. As essential resources become spatially separated in stratified lakes, this motility, combined with an ability to perceive environmental gradients, gives flagellates the opportunity to position themselves at a favourable depth. Using preference chambers and cell-track analysis this study investigated the ability of five phylogenetically contrasting species of flagellates to detect and respond to gradients of light, temperature, oxygen, carbon dioxide, pH and phosphate. Measurement of population distribution, swimming speed, swimming direction and kinetic reactions determined the sensory response of cells and established a series of optimal preferences. Finally, to complete the study, multi-factorial experiments testing the response of flagellates to combinations of gradients were carried out in an attempt to assess preference dominance and determine if flagellate distribution in stratified lakes may be explained by behavioural trends found in laboratory experiments.

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CYANOBACTERIAL TOXINS: THEIR ACTIONS AND MULTIPLE FATES IN MICROBES, ANIMALS AND PLANTS

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Understanding of the significance of cyanobacterial toxins in natural and anthropogenic environments and of the possible roles of these compounds in the life of the producer-cells is currently advancing. This is partly because of increasing knowledge of the occurrence, molecular structure and toxicity (largely in animal vertebrate systems) of the toxins. However, as more investigations are performed on the effects and fates of cyanobacterial toxins in aquatic, terrestrial and aerial laboratory and environmental systems, so our understanding of their significance and multiple fates is increasing. Cyanobacterial poisoning in animal vertebrates is relatively well recognised. This is due to improved case descriptions and methods for the extraction of the toxins, including microcystins, nodularins, saxitoxins, anatoxins and cylindrospermopsins, from exposure sources and exposed material (fish, birds and mammals, including humans). However, there are now wider indications of the significance of cyanobacterial toxins to microbes, lower animals, the early developmental stages of vertebrates and in plants. Examples of outcomes of exposure to cyanobacterial toxins, at environmentally relevant concentrations, are presented, including: adverse effects on the avoidance, respiration and growth of protozoa (Paramecium, Tetrahymena); stress and developmental aberrations in fish and amphibian eggs, embryos and juveniles; uptake of microcystins by rooted and non-rooted aquatic plants (*Ceratophyllum*, *Phragmites*) and terrestrial plants (*Phaseolus*, *Zea*, Sinapis), and inhibitory effects on plant photosynthesis. In addition to causing a wide range of adverse effects, cyanobacterial toxins can undergo various fates including biodegradation in aquatic bacteria, and enzymic detoxication. Individual cyanobacterial toxins may have multiple roles in terms of the producer-cells. Whether their primary functions are due to their toxicity is unclear. To some eyes, the molecules include metal chelators and cell signalling messengers and, for microcystins, there is evidence for both of these possibilities.

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CAN DNA SEQUENCING TELL US ANYTHING ABOUT HOW FRESHWATER ALGAE TRAVEL?

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It seems widely accepted that most freshwater algal genera and species are worldwide in distribution. An opportunity to assess what this really means arose from an analysis of mating affinity, nuclear ribosomal DNA Internal Transcribed Spacer sequence, and geographic distribution of more than 100 worldwide isolates of *Pandorina*, *Volvulina* and *Yamagishiella* The *Pandorina morum* clade (including also additional species names such as *P. smithii*, and *P. colemaniae*), though morphologically uniform, encompasses an evolutionary span, as determined by comparison of ITS, greater than either of the multispecies genera *Gonium* and *Eudorina*. There are at least 30 *Pandorina*/*Volvulina syngens*, sexually isolated groups, so far as can be determined, among the current collection of strains. In addition, as in other Volvocacaean genera, two clones are homothallic, capable of forming zygotes within a genetic clone. Within each syngen, genetic distance increases with geographic distance between collection sites. At least half the isolates studied must have been introduced since the Pleistocene, from the south, not the west. The study of all these organisms, distributed throughout the world presumably by the activities of shorebirds, suggests that a morphological species CAN be worldwide in distribution, but that interfertile populations are geographically localized. The driving force is that any body of freshwater is, in a sense, an island subject to its own evolutionary course. The study provides initial data on the rate of appearance of new mating groups versus their rate of distribution over the earth.

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CHLAMYDOMONAS REINHARDTII MUTANTS THAT REQUIRE ELEVATED CO_2 FOR OPTIMAL GROWTH.

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Chlamydomonas reinhardtii possesses a CO_2 concentrating mechanism that enables it to grow at very low CO_2 concentrations. In order to identify genes required for growth at low CO_2, an extensive screening of *C. reinhardtii* insertional mutants was conducted. The D66 strain (*nit2*$^-$, *cw15*, *mt*$^+$) was transformed by electroporation using a plasmid containing a gene that confers resistance to the antibiotic Zeocin. About 42,000 primary transformants were screened. These transformants were first grown on acetate in the dark. The colonies were then tested for their ability to grow photosynthetically on elevated CO_2 or very low levels of CO_2 (100 ppm). About 80 mutants were identified which were unable to grow well at low CO_2 concentrations. Another 80 mutants were unable to grow photoautotrophically at high or low CO_2 concentrations. The location of the inserted DNA is being determined using a combination of inverse PCR and TAIL PCR. Using these methods, one can rapidly locate the inserted DNA in the genome and identify the gene that has been disrupted by the insertion. One of the mutants has an insertion in the gene that encodes Rubisco Activase. Our evidence suggests that Rubisco Activase is required for growth under low levels of CO_2. Supported by NSF grant IBN-09904425 to JVM and a postdoctoral fellowship to SLC from Fund Antorchas.

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ULTRASTRUCTURE OF THE ENIGMATIC CHAROPHYCEAN GREEN ALGA *ENTRANSIA FIMBRIATA*

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Based on molecular, ultrastructural, and biochemical data, the charophycean green algae are considered to be the closest living relatives of plants. Hence, comparative studies of these algae may increase our understanding of ancestral characteristics in the streptophyte lineage (charophyceans + embryophytes). Recent molecular phylogenetic analysis suggests that *Entransia fimbriata* is a deeply branching charophycean that is perhaps affiliated with the order Klebsormidiales (Karol and Delwiche unpublished). Transmission electron microscope examination of *Entransia* reveals that cytokinesis occurs by means of furrowing that does not appear to involve microtubules, a characteristic also reported in cytokinesis of *Klebsormidium flaccidum*. Both taxa are unbranched filaments of cells with a single starch-filled chloroplast that contains a pyrenoid with traversing membranes. No plasmodesmata are found in either organism. Thus, preliminary TEM findings support the molecular evidence indicating that *Entransia* may be affiliated with Klebsormidiales. Among the characteristics that distinguish *Entransia* from *Klebsormidium* are a highly lobed chloroplast and some unusual wall features. In many cases, the ingrowing septum edges of *Entransia* do not meet in the most direct way, forming cross walls with large protuberances. Thick cross walls in *Entransia* are apparently also created by short cells that die and collapse, functioning as necridia.

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SIMILARITIES BETWEEN BENTHIC CYANOBACTERIA AND EUKARYOTIC ALGAE: CHEMICAL DEFENSES AND THE ANIMALS WHO LOVE THEM

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About 400 compounds have been isolated from marine cyanobacteria worldwide, but their ecological roles remain largely unknown. In the coral reefs of the tropical Pacific, large filamentous cyanobacteria are common components of the benthic community. Many of these benthic cyanobacteria produce secondary metabolites that serve as chemical defenses against large grazers. However, diverse small consumers have evolved to cope with cyanobacterial metabolites and preferentially use cyanobacterial mats as food and shelter. While studies have focused mainly on the negative effects of cyanobacteria on benthic communities, the positive roles of cyanobacteria for benthic animals have rarely been addressed. Interactions between filamentous cyanobacteria and their associated fauna are diverse and include sequestration of cyanobacterial metabolites by gastropods, facultative and specialized consumption of cyanobacteria by grazers, temporary shelter for small predators, and symbiosis between cyanobacteria and some crustaceans. Although examples of similar interactions between chemically-defended eukaryotic algae and their associates are known, these interactions have not been previously addressed for cyanobacteria. The number of organisms that preferentially associates with benthic filamentous cyanobacteria suggests that these prokaryotes can significantly influence local biodiversity patterns. This research indicates strong parallels on the ecology and evolution of consumer-prey interactions between eukaryotic and prokaryotic algae in coral reefs.

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BENTHIC MARINE ALGAL HERBARIUM OF LONG ISLAND SOUND DIGITAL COLLECTION

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University of Connecticut presents the "Benthic Marine Algal Herbarium of Long Island Sound Digital Collection."

When Phase One of this project is completed, this collection will include an online herbarium of all Long Island Sound macroalgae species. The database will be on the web and open to the public. The taxonomy and descriptive text are part of a collaboration between the University of Connecticut and NEAS. Database features include the ability to create searches and generate sets based on subjects, division, class, order, family, genus, habitat, species, keyword, location, etc. Each record will be cataloged according to Dublin Core cataloging guidelines. All species have a thumbnail image and a larger image for full viewing. TIFF files will be archived and available in the future (10mg images). Certain species will be marked as a teaching collection and made available upon request on a CDrom for teaching purposes. Future plans include expanding the database geographically in the Northeast. The database is located and maintained on a server at the Homer Babbidge Library on the UConn Storrs campus. The information is in Microsoft Access, and is made available for viewing and searching on the web through ColdFusion. This online collection is in the process of being created, a test site is now available at: http://norman.lib.uconn.edu:6550/algae/algaesearch.cfm (hit enter for all records or use search term: lamin)

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BIOREMEDIATIVE POTENTIAL OF *CHROMULINA FREIBURGENSIS* IN CULTURE FROM THE BERKELEY PIT

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The Berkeley Pit, part of the largest Superfund site in the United States, is an open-pit copper mine that operated from 1955 through 1982. Today, the Berkeley Pit contains approximately 1200 billion liters of metal laden water with an average pH of 2.7, and 12 grams/liter of dissolved solids. The principle dissolved ions include aluminum, arsenic, calcium, cadmium, copper, iron, potassium, magnesium, manganese, sulfates, and zinc. A species from Division Chrysophyta--*Chromulina freiburgensis* Dofl. was isolated from this extreme environment. This species has been tested in the laboratory through a series of controlled experiments to determine bioremediative potential. Optimal temperature was determined by monitoring growth with cell counts at temperatures ranging from 5 C to 40 C . The optimal nutrient ratio was determined by varying nitrogen ($NaNO_3$) and phosphorus (Na_2HPO_4) levels. An experimental matrix varying nutrients was developed to test for bioremediative potential which included: initial and final pH measurements; initial and final Ion Chromatography Pairing–Atomic Emission Spectrometry (ICP-AES) for dissolved metals; and examination of final samples under Transmission Electron Microscopy (TEM). From these experiments, *Chromulina freiburgensis* was found to grow optimally in Berkeley Pit surface water with cell densities reaching ten million cells per milliliter at 10^0C with additions of 50 mg $NaNO_3$/L and 5 mg Na_2HPO_4/L. This large biomass was also found to increase diversity and abundance of heterotrophs. At the optimal nutrient level, this species was found to increase pH from 2.21 to 2.47 over 90 days. Significant removal of calcium, iron, nickel, and silica was observed.

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CONTRIBUTIONS TO UNDERSTANDING SEAWEEDS IN COASTAL COMMUNITIES

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The goal of my presentation is to review several studies that have enhanced our understanding of the effects of abiotic factors on coastal and estuarine seaweed populations. Accordingly, I will introduce a few key papers dealing with five major abiotic factors- i.e. salinity, temperature, desiccation, water motion, and illumination. Foremost, the salinity tolerance studies of Russell and Bolton (1975) have broad applicability to estuarine seaweeds, while the osmoregulatory studies of Bisson and Kirst (1979) are also significant. Biebl’s (1972) review of his earlier studies on temperature tolerances in diverse seaweeds were pivotal. Johnston and Raven’s (1986) studied the effects of desiccation on the fucoid brown alga *Ascophyllum nodosum*, while similar studies on the saccate brown seaweed *Colpomenia peregrina* were conducted by Oates (1985). Lewis (1968) conducted early synoptic evaluations of the effects of water movement on rocky shore communities, while Kitching and Ebling (1967) gave detailed assessments of seaweed populations within estuarine tidal rapids in Ireland. Basically estuarine tidal rapids represent areas of enhanced nutrients, oxygen and light availability, plus reduced sedimentation (Mathieson *et al*. (1983). The physiological effects of light have probably been evaluated more than any single abiotic factor and two areas of importance are cited here. The critical papers by Levring (1947) on submarine illumination and those of Ramus (1978) and Littler and Littler (1980) on algal form and light response. Several areas of future studies are also suggested, which may further enhance our understanding of seaweed adaptations. In summary, five major abiotic factors affecting coastal and estuarine seaweed populations will be discussed, their importance to seaweeds noted, and “key” findings for several significant papers summarized.

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MOLECULAR PHYLOGENETICS AND THE DINOFLAGELLATE CHLOROPLAST

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The chloroplasts of dinoflagellates have been among the last chloroplasts to be studied with molecular phylogenetic methods. DNA sequences from peridinin-containing chloroplasts are now available, and have begun to be examined with molecular phylogenetic methods. The high rate of sequence evolution in these genes increases noise and makes analyses difficult. We have evaluated phylogenetic signal in several dinoflagellate chloroplast genes using parametric bootstrapping and other methods. Although some aspects of the tree topologies found in phylogenetic analyses of dinoflagellate chloroplast genes are clearly artifactual, there are some features that seem to reflect genuine phylogenetic information. We find support for the concept of monophyletic chromophyte chloroplasts, but contradictory information concerning the relationships among the host cells that contain these plastids. The best hope for determining the evolutionary history of chromophytic plastids lies in the analysis of nuclear-encoded, chloroplast expressed genes.

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AMONG-SITE VARIATION IN THE EFFECTS OF TRAMPLING DISTURBANCE ON *SILVETIA COMPRESSA* (O. FUCALES) POPULATIONS

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Fleshy seaweeds and many invertebrates are vulnerable to trampling disturbance, particularly on rocky shores intensely used by people. The effects of human foot traffic on the canopy-forming fucoid *Silvetia compressa* (J. Ag.) Serrão, Cho, Boo et Brawl. were experimentally determined on three southern California rocky shores that receive different levels of abiotic (wave exposure) and visitor disturbance. At each site, 15 plots (0.5 m x 0.7 m) were randomly established in a well-developed *S. compressa* assemblage and then grouped into 5 blocks. Plots were randomly assigned to either 0 (Controls), 150, or 300 foot-step treatments within each block, and experimental trampling treatments were applied monthly for 16 consecutive months. *S. compressa* thalli were then harvested from 0.3 m x 0.5 m quadrats that were centrally positioned within each plot and returned to the laboratory for analysis. Thallus densities, vegetative and reproductive (i.e., receptacle) biomass, and the size (i.e., longest axis) of each harvested individual were determined. Trampling treatments resulted in significant reductions in vegetative and reproductive biomass, and the size structures of populations harvested from the trampled plots showed higher frequencies of smaller thalli. The population structure and degree of trampling damage varied among sites corresponding with existing levels of abiotic and human disturbance. These results indicate that even moderate levels of human foot traffic can alter the size structure of *S. compressa* populations and significantly reduce biomass and reproductive output. Additionally, the magnitude of trampling damage varied in a pattern corresponding with site disturbance regimes.

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RIVER PLANKTON AND WATER QUALITY OF THE LOWER RIO GRANDE/RIO BRAVO, TEXAS, USA

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Little if anything is known about the algae in the lower Rio Grande/Rio Bravo (Laredo to Brownsville, TX) and basic water quality information is limited. This river supplies drinking, irrigation and industrial use water in this US-Mexico border region. A long-term monitoring program was initiated in March 1999 and continues to the present. Data from March 1999 to March 2001 is presented. Water samples were collected from the river upstream (2 sites) and downstream (1 site) of an urban area (McAllen-Reynosa) and analyzed for nutrient concentrations (ammonium, nitrate-nitrite and phosphorus), chlorophyll a concentration and phytoplankton composition and quantity. During the study, average summer and winter temperatures in this river segment ranged from 28.4-30.1 ^{0}C and 17.5-21.8 ^{0}C, respectively. The river is oligotrophic upstream of the urban area and hypereutrophic downstream of the urban area. Average chlorophyll level was highest at the downstream site both years (1999- 14.6 µg/l, 2000-21.1µg/l). Each summer the downstream stretch experienced a cyanobacterial bloom consisting mostly of *Microcystis aeruginosa* although *Merismopedia* spp. and *Oscillatoria* spp. were common. Chlorophytes occurred at a low but consistent density in the warmer months. Diatoms were typically a minor component of the phytoplankton.

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PROBLEMS AND PROMISE IN CODING MORPHOLOGICAL CHARACTERS FOR PHYLOGENETIC ANALYSIS: AN EXAMPLE FROM *AULACOSEIRA* (BACILLARIOPHYCEAE)

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Understanding the phylogenetic relationships among species of *Aulacoseira* Thwaites (formerly *Melosira*, in part) and their relationships to other genera is important for several reasons. *Aulacoseira* is globally distributed and is often a large component of planktonic communities in lakes, ponds and large rivers. Consequently, it plays a significant ecological role as a primary producer in grazer-based aquatic food webs. Although species of *Aulacoseira* (along with other diatom species) are used profitably as tools for reconstruction of paleo-environments, ecological monitoring, and other ecological studies which have generated a large amount of data, much of this data is not really informative for a systematic understanding of the group. There are currently around 60 extant and 20 extinct

described species of *Aulacoseira*. However, there is no estimate of phylogeny within this genus that includes more than five species. Here we present the first comprehensive estimate of phylogeny for 30 species of *Aulacoseira* based on a cladistic analysis of morphological features gathered from the literature. Complications of gathering morphological data from the literature that was largely focused on refining species identifications for practical purposes of ecological identification will be addressed. Morphological characters chosen for the analysis and challenges associated with coding of characters will be discussed along with the resulting *Aulacoseira* phylogeny and our methods of its estimation.

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PHOTOBIOREACTORS FOR PHYCOLOGY: DESIGN AND FUNCTION OF A COMPUTER CONTROLLED, CULTURING APPARATUS OPTIMIZED FOR ANALYTICAL INVESTIGATIONS

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The majority of photobioreactor research has been directed at optimizing biomass yields. However, the potential for exploiting these highly controlled culturing environments for most physiological research has yet to be realized. The main reason for this is that photobioreactor technology has tended to be overly complex and somewhat cost prohibitive. A new generation of computer-controlled photobioreactor, designed and optimized for physiological research, has been developed using relatively inexpensive, easily obtained materials. The computer software responsible for controlling this apparatus allows simultaneous monitoring and control of pH, O_2, temperature and light for up to six photobioreactors operating concurrently. Additionally, the reactors can be operated as batch cultures, continuous cultures or in a self-regulating, turbidostat mode. All data pertaining to the reactors can be logged in real-time to the controlling computer. Most importantly, the control software can be freely distributed. Potential applications and design flexibilities will be discussed.

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IDENTIFICATION OF HARMFUL MARINE DINOFLAGELLATES

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An interactive, multimedia taxonomic reference guide of harmful marine dinoflagellate microalgae was developed containing species information and identification keys in the form of text, photographs and illustrations. This treatise represents a comprehensive, up-to-date multi functional dinoflagellate identification database featuring information on 68 species (15 taxa) of harmful dinoflagellates and associated species present in the world's oceans. There are 42 toxins-producing species, brevetoxins, saxitoxins and ciguatoxins, and seven nuisance species that produce 'red tides' often resulting in massive sea life mortalities. In addition to species description, information is available on the morphology, reproduction, ecology, toxicity, and habitat and type locality for each species. A comprehensive glossary of more than 120 related scientific terms, and an extensive literature reference collection containing about 500 citation is included. There are 600 figures illustrating each species via scanning electron micrographs (60%), light micrographs (20%), and line drawings (20%). The matrix-based identification key allows for multiple entry of characters to facilitate fast and effective diagnosis of a given species. All this data can be accessed easily on a CD-ROM, making this an excellent teaching tool and identification guide of harmful marine and associated dinoflagellate species. Here, I will present examples for several species of dinoflagellates in attempt to illustrate the usefulness of this identification guide in the lab or the field as a useful and valuable tool for the researcher, the instructor, as well as the students.

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MORPHOLOGY AND ECOLOGY OF *GLENODINIUM ARMATUM* LAVENDER (DINOPHYCEAE)

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Light and scanning electron microscopy were used to examine the morphology of *Glenodinium armatum* Levander, a small armored dinoflagellate from the Tvärminne Archipelago, Baltic Sea, Finland. This species is an important dinoflagellate in coastal rock pools. Cells are small, ovate, and dorso-ventrally flattened. The cell surface is rugose with small round pores. The plate tabulation is: Po, x, 4', 2a, 7", 6C, 5S, 5"', 2"". Two intercalary plates are hexagonal and adjacent. Epitheca conical and hypotheca round and two antapical spines are present. The cingulum wide, slightly displaced and descending, and comprised of six cingular plates. Sulcal platelets are five and do not reach the antapex. Often in the hypotheca, a red horseshoe-shaped stigma is present. Large nucleus situated posteriorly. Chloroplast golden brown and discoid. We suggest that *G. armatum* is a valid species with clear differences to *Peridinium umbonatum* Stein (syn. *P. inconspicuum* Lemmermann), but that it has not been always correctly identified. A new emended description is presented.

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THE *PSEUDODICTYOSPHAERIUM/MYCONASTES* SPECIES COMPLEX: COMMON PHYTOPLANK-

TERS FROM MINNESOTA AND NORTH DAKOTA LAKES.

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We have established numerous cultures of planktonic autosporic coccoid green algae from lakes of the Arrowwood National Wildlife Refuge, North Dakota, and Itasca State Park, Minnesota. Among these cultures, we have found many isolates that match the morphological descriptions of the genera *Pseudodictyosphaerium* and *Mychonastes*. Cells of these two genera are spherical and lack pyrenoids. *Pseudodictyosphaerium* is colonial in a common mucilagenous matrix and *Mychonastes* cells are solitary. Previous analyses of 18S rDNA sequences have indicated a close relationship between these genera within the Chlorophyceae. Preliminary analyses of the 18S rDNA sequence data from our isolates suggest that the colonial character may not define a monophyletic lineage. All of the isolates that we have analysed to date have 18S rDNA sequences that vary from that of *Mychonastes homosphaera* by no more than 7 bases. Based on these results, we propose the hypothesis that these asexual organisms form a complex of closely related clonal lineages that can not easily be described as "species" based on 18S rDNA sequences. Species designations should be based on genetic differentiation combined with consistent morphological, physiological, or ecological characters.

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EFFECTS OF LIGHT ON GRAZING AND CANNIBALISM OF *PFIESTERIA PISCICIDA* (DINOPHYCEAE)

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The toxic dinoflagellate *Pfiesteria piscicida* is primarily a heterotrophic organism, with photosynthetic potential through the 'stolen' chloroplasts of its algal prey ('kleptoplastidy'). Classical predator-prey oscillation was found in this dinoflagellate fed with *Rhodomonas sp*. We show that light has an influence on feeding and growth in *P. piscicida* that transcends its potential influence on kleptoplastidic photosynthesis. To our surprise, we found that light strongly stimulated grazing and heterotrophic growth in *P. piscicida*. In addition, under starvation conditions, *P. piscicida* population declined significantly more rapidly in the light, and further experiments suggest that the difference in rates of decline may be explained in part by light-stimulated cannibalism. The possible biochemical mechanism and ecological significance of light stimulation of grazing and cannibalism will be discussed.

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DEALING WITH THE SUN'S WORST SIDE: CYANOBACTERIAL SUNSCREENS

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Many cyanobacteria produce secondary metabolites that are assigned a role as sunscreens. I will briefly review the current knowledge on the chemical diversity, phylogenetic distribution, mode of action, and the biochemical pathways of synthesis and regulation of both lipid-soluble and water-soluble sunscreen secondary metabolites in cyanobacteria. Special attention will be given to the evolutionary implications of the use of sunscreens by cyanobacteria, as they relate to paleoenvironmental conditions.

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INTRAGENOMIC nrDNA POLYMORPHISM IN THE *STEPHANODISCUS NIAGARAE* COMPLEX (BACILLARIOPHYCEAE)

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Although concerted evolution generally acts to homogenize nuclear ribosomal DNA (rDNA) arrays, a certain amount of intragenomic variation is not uncommon. Despite its potential to seriously compromise phylogenetic reconstruction, this variation has largely been ignored by systematists. We examined levels of intragenomic ITS variation within the recently and rapidly derived *Stephanodiscus niagarae* complex, where speciation has almost certainly proceeded faster than concerted evolution. We examined multiple populations of *Stephanodiscus niagarae* and its closest relatives, included related species of *Stephanodiscus*, and sampled several species of *Cyclostephanos* and *Cyclotella*. PCR was performed on DNA extracted from monoclonal cultures, ITS fragments were cloned, and 6-10 amplicons selected for each of 14 populations for a total of about 125 sequences. Every species examined contains some level of intragenomic ITS polymorphism, ranging from multiple haplotypes differing by a few basepairs to highly diverged paralogues that probably represent independent loci. Phylogenetic analyses indicate several haplotypes shared among *S. niagarae* and its derivatives that probably predate the origin of the species complex. Despite extensive variation within other species, there appears in some cases to have been sufficient homogenization (and unambiguous clustering) of ITS types to clearly resolve deeper nodes in the family. These results suggest caution concerning the use of ITS sequences, which, unless sampled and analyzed carefully, may actually obscure species relationships and provide misleading information about population structure. We discuss the phylogenetic implications of the preliminary ITS gene tree and consider the use of fossil data to calibrate ITS evolution in diatoms.

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HYDRODYNAMICS AND ALGAL REPRODUCTION

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Environmental conditions, such as water motion, can influence fertilization success and spore dispersal in marine algae. Previous studies in fucoid algae showed that gamete release is restricted to periods of low water motion; however, few other algal taxa have been investigated, including species with an alternation of generations. We investigated gamete and spore release in *Alaria esculenta* (Phaeophyceae) and *Ulva lactuca* (Chlorophyta) to determine if water motion is inhibitory or stimulates propagule release. Sporophytes (sporophylls) and gametophytes (*U. lactuca* and *A. esculenta*) were assigned to either calm or shaken treatments inside a walk-in culture chamber. The number of zoospores (*A. esculenta*, *U. lactuca*) or gametes (*U. lactuca*) released each day was determined with a hemacytometer. Antheridial release was observed directly in *A. esculenta* after fixation. *A. esculenta* released a significantly higher number of zoospores under turbulent conditions (P= 0.0001, 2-way ANOVA) and a higher number of sperm under calm conditions (P = 0.001, 2-way ANOVA). Juvenile *A. esculenta* sporophytes were present in significantly higher numbers in calm treatments (P = 0.001 2-way ANOVA; contact time, male + female gametophytes = 150 min). Significantly higher release under turbulent conditions was observed for both U. lactuca gametes (P = 0.0021, 2-way ANOVA) and zoospores (P = 0.0028, 2-way ANOVA). These data demonstrate that different algal taxa have varying reproductive responses to water motion and have interest with respect to parthenogenesis, gametophyte and sporophyte ratios, and life-history adaptation.

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ALGAE AS A TEACHING TOOL IN ELEMENTARY CLASSROOMS

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In the University of Maine's National Science Foundation Graduate Teaching Fellows in K-12 Education program, 4th and 5th grade students at Lewis Libby Elementary School in Milford, Maine, were exposed to graduate-level science during the 2000-2001 academic year. This program brought cutting-edge research into the elementary classrooms, enriching the science curricula for students and providing professional development for teachers. Algae were used as subjects to learn microscopy, cell structure, the process of fertilization, and how living things are classified. Students made independent discoveries of the chloroplasts and nuclei in several algal genera, as well as the eyespot in *Euglena*. These discoveries prompted the students to think about the diversity of life, what similarities and differences exist between species, and the kinds of adaptations organisms have acquired. In addition, algae were used to help students understand the natural history and marine resources of Maine. Algae are ideally suited for use in elementary classrooms because of the ease by which they can be observed and the broad number of concepts they can be used to illustrate.

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COMMUNITY DYNAMICS OF PHYTOPLANKTON, PROTOPLANKTON AND BACTERIOPLANKTON IN SELECTED WISCONSIN LAKES

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As part of an NSF-sponsored Microbial Observatory program at the North Temperate Lakes Long-Term Ecological Research Site, microbial community dynamics are being monitored in three Wisconsin lakes, representing a trophic spectrum. Bacterioplankton diversity is assessed by automated ribosomal intergenic spacer analysis (ARISA) and dominant members identified by T-RFLP. Phytoplankton and protoplankton are enumerated in volumes of lake water varying from 5 to 25ml. Surrounded by the city of Madison, eutrophic Lake Mendota is dominated by cyanobacteria throughout the ice-free seasons. Our intensive counting procedures, however, have revealed an underlying diverse assemblage of chlorophytes, typical of less impacted waters, at low population levels. Dystrophic Crystal Bog in northern Wisconsin has a low diversity of phytoplankton and protoplankton and in summer becomes almost a pure culture of *Peridinium limbatum*, an event that may be correlated with a dramatic decline in bacterioplankton diversity. In winter the bog is dominated by heterotrophic production as biomass of protozoa exceeds that of phytoplankton. Nearby oligotrophic Crystal Lake has a high species diversity of both phytoplankton and protoplankton, and cyanobacteria and chlorophytes are very diverse. In autumn the lake supports an unusual planktonic grazer, a testate amoeba which captures bacterioplankton and phytoplankton in a net of pseudopodia. Preliminary results indicate that bacterioplankton dynamics may be strongly linked to algal dynamics in the eutrophic and dystrophic lakes but are uncoupled in the oligotrophic lake.

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EFFECTS OF SURFACE HYDROPHOBICITY AND LIGHT ON SPORE SETTLEMENT IN *HINCKSIA IRREGULARIS* (PHAEOPHYCEAE)

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We examined the effects of light and surface hydrophobicity individually and in tandem on *Hincksia irregularis* spore settlement. *H. irregularis* spores were determined to be negatively phototactic by the use of computer-assisted motion analysis. Spore settlement was significantly influenced by surface hydrophobicity and by light, individually and in tandem. Settlement experiments revealed significantly higher settlement on hydrophobic surfaces and in dark environments when compared to negatively charged surfaces and lighted environments. Experimental light/dark boundaries elicited distinct spore settlement responses, with spores displaying dissimilar settlement patterns on plates with different surface hydrophobicities. Complex responses of *H. irregularis* spores to light and charge may affect their dispersal and probably aids in their detection of suitable settlement locations in marine microenvironments. The behavioral responses exhibited by *H. irregularis* suggest that light and charge should be investigated individually and in tandem in studies of other fouling organisms. (MASGC R/MT 40)

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rRNA PROBES FOR IDENTIFICATION AND CHARACTERIZATION OF MARINE PHYTOPLANKTON: THEIR POTENTIAL APPLICATION FOR DNA MICROCHIPS

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A fast and reliable identification of nano- and picoplankton by light microscopy is often difficult because of the lack of usable morphological characteristics, whereas electron microscopy and biochemical methods are very time consuming. Identification of toxic algae also requires a great deal of taxonomic experrtise so that false positives are not recorded. One solution is to use taxon specific rRNA probes. For this purpose we designed probes for phytoplankton taxa, including toxic algae. These probes were either labelled with Digoxigenin (DIG) and used in DNA dot blot experiments, or labelled with fluorochromes and used in whole-cell hybridisations with fluorescence microscopy or flow cytometric detection. Specific probes could be used over a broad taxonomic range from higher groups (i.e. the class of dinoflagellates) to species level (i.e. *Prorocentrum lima*). These probes were be used in the EU MAST project AIMS for the development of an automated identification system for marine phytoplankton in combination with flow cytometry and artificial neural networks (ANNs), in the EU MAST DETAL and in the German national project (TEPS) for the development of an early warning system for harmful algal blooms. Results using Digoxigenin (DIG)-labelled probes on picoplankton samples taken from several water bodies indicate that hierarchial re-probing of spotted samples can be achieved and this suggests that probes can be adapted to DNA microchips. Preliminary field results for a hand-held DNA microchip reader are presented. This work was supported by the German BMBF TEPS 03F0161 and the EU AIMS MAS3-CT97-0080 and EU DETAL Q5RS-2000-30778 projects.

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THE *CHLAMYDOMONAS* GENOME PROJECT.

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The *Chlamydomonas* genome project involves 1) sequencing of cDNAs isolated from cells exposed to various environmental conditions, 2) construction of a high density DNA microarray, 3) construction of genomic contigs that nucleate around specific physical and genetic markers, 4) generation of a complete chloroplast genome sequence and analyses of chloroplast gene expression, and 5) placement of the genomic information on the network in a user friendly format. The aspects of the project emphasized by our group at Stanford involves the generation of normalized cDNA libraries, sequencing of the cDNAs to generate both contigs and unigene families and the use of this information to construct a high density cDNA microarray. I will discuss the techniques involved in securing cDNA sequence information and the ways in which that information is assembled and analyzed. I will also discuss the use of the different cDNA libraries to identifying differentially expressed genes (by in silico subtractions) and strategies for constructing high-density cDNA microarrays. Finally, if completed, the first global expression studies, and the use of the microarrays to elucidate global expression in mutant strains will be discussed.

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MACROALGAL BLOOMS IN FLORIDA'S COASTAL WATERS: *CODIUM ISTHMOCLADUM*

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During the past decade, reports of macroalgal blooms along the reefs of southeast Florida have increased. These blooms may become harmful due to negative impacts associated with excessive biomass, smothering of corals, sponges, and other reef organisms, and benthic hypoxia and anoxia. Since 1989-1990, recurrent blooms of *Codium isthmocladum* have occurred on reefs off Palm Beach and Broward Counties in depths between 20 and 60 m. The objectives of this study were to determine the spatial and temporal variability in *C. isthmocladum* biomass and tissue nutrient (C:N:P) ratios. Studies conducted primarily on populations in northern Palm Beach County showed distinct seasonal and spatial patterns in biomass. The growth of individual thalli followed the classic sigmoidal growth curve, with linear increases in biomass occurring during the summer, and growth terminating in late September/October. During peak growth, thalli doubled ca. every week (i.e., specific growth rate = ca. 0.10 doublings d-1). Tissue nutrient analyses supported the hypothesis that nitrogen was limiting the growth of this alga. Growth of *C. isthmocladum* is largely a result of its relationship with light, temperature, and nutrients; when conditions are favorable, and in the absence of physical disturbance, the alga is capable of explosive growth. In the last two years, blooms of *Caulerpa verticillata* have been occurring on the same reefs where *C. isthmocladum* is still abundant. Anthropogenic nutrients may be an important part of the reason why macroalgal blooms have been occurring for over a decade in south Florida's coastal waters.

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A DISTRIBUTIONAL ANALYSIS OF BENTHIC MARINE ALGAE COLLECTED IN SOUTH-CENTRAL ALASKA DURING THE CHIA STUDIES OF EVOS

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During the University of Alaska's Coastal Habitat Injury Assessment (CHIA) study of the Exxon Valdez oil spill, intensive investigations were carried out on the intertidal algal communities of Prince William Sound, the Kenai Peninsula, the Kodiak Islands, and the Alaska Peninsula. As a part of these investigations, thorough voucher collections were made of the benthic marine algae present in more than 100 transect areas used for the study. The 7,300 collected specimens were pressed, identified to species, curated, and cataloged. For this study, we have utilized these data as well as information on the habitat types and species life histories to prepare regional checklists and distributional analyses of the species discovered. Environmental conditions in south central Alaska vary regionally as well as seasonally, and the narrow vs. broad ranges of these species appear to reflect closely their ability to tolerate these diverse conditions. The algal biodiversity, distribution, and habitat data made available through this study provide crucially needed background information for future environmental impact studies and for restoration and conservation efforts inAlaska.

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FAMILY AND GENERIC DELINEATION WITHIN THE ACROCHAETIALES (RHODOPHYTA)

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Recent SSU- and LSU-based molecular systematic investigations have confirmed the close relationships between the members of the Acrochaetiales-Nemaliales-Palmariales complex. While the Nemaliales and Palmariales have consistently been resolved as monophyletic, the Acrochaetiales is represented by two divergent and strongly supported groups -Acrochaetiales I and II. While the Nemaliales has been resolved as sister to the Acrochaetiales and Palmariales, the relationships between the remaining taxa are equivocal. Recognition of two families within the Acrochaetiales is proposed - an emended Acrochaetiaceae and Colaconemataceae fam. nov. Molecular data have confirmed the placement of *Acrochaetium*, *Audouinella*, and *Rhodochorton* within the Acrochaetiaceae and *Colaconema* within the Colaconemataceae. Based on morphological and anatomical grounds, the genus *Liagorophila* is placed in synonymy with *Acrochaetium*. The Colaconemataceae houses a diverse assemblage of marine acrochaetioid taxa, and will most certainly be represented by multiple genera following further investigation. Key features of taxa within this group will be presented, as well as a review of the valid generic names available for colaconemataceaen species - *Grania* and *Kylinia*.

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SEAWEED CHEMICAL ECOLOGY: ITS IMPORTANCE TO INDIVIDUALS, POPULATIONS, AND COMMUNITIES.

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Seaweeds use defensive chemicals to deter a wide range of generalist consumers. Once seaweeds deter larger consumers like fishes and urchins, these seaweeds become sites of evolutionary opportunity for small, sedentary herbivores that escape or deter their own consumers by associating with toxic algae. In many cases, the compounds that deter fishes are the specific compounds that small, specialist herbivores use to locate or recognize their hosts. In numerous cases, it appears that "feeding" specialization is really "habitat" specialization, and is driven by the need for small sedentary herbivores to escape predation rather than by nutritional traits of the host. Similarly, palatable seaweeds can avoid herbivores by associating with unpalatable competitors; in some situations, palatable species may depend on defended competitors to prevent their local extinction due to herbivory. Seaweed chemical defenses also show strong geographic patterns (more in tropical than in temperate species), local patterns (more in herbivore-rich than in herbivore-poor sites), and within thallus patterns (concentrated in parts of the plant, or at times of the season or even day). Some intraspecific variance in defenses is caused by variance in physical stress (desiccation, UV) or induction of defenses following attack by small herbivores. In some cased, geographic variance is herbivore feeding choices, behavior, and evolution is driven by the indirect effects of seaweed defenses. Thus, chemical defenses often produce cascading effects that impact the ecology and evolution of marine systems as a whole.

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SYSTEMATICS OF *ULVA* AND *ENTEROMORPHA* (ULVALES), WITH EMPHASIS ON SPECIES FROM THE NORTHEAST PACIFIC

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Ulva L. and *Enteromorpha* Link are two of the most common macroalgal genera world wide and are well known for their role in “green tides” and marine biofouling. Traditionally, these genera have been distinguished from one another by their different morphologies: *Ulva* species have distromatic blades, and *Enteromorpha* species form monostromatic tubes. Despite the distinct morphological difference between these two genera, morphological simplicity and plasticity of algae within each of them have confounded systematic and taxonomic efforts. Recent molecular studies of primarily European taxa indicate that *Ulva* and *Enteromorpha* are not reciprocally monophyletic yet within the *Ulva/Enteromorpha* assemblage several species can be identified. A study of Pacific *Ulva* and *Enteromorpha* was undertaken to reconstruct phylogeny and test species hypotheses based on morphology, development and hybridization experiments. Multiple samples of the eleven currently recognized *Ulva* and *Enteromorpha* species along the Pacific coast of North America were collected from Alaska to southern California. Collections were identified using existing morphological descriptions, and when possible, growth in culture. Sequences for the internal transcribed spacers of nuclear ribosomal DNA and the chloroplast-encoded gene for the large subunit of RUBISCO were determined for these samples and additional ones from Australia, Chile, Hawaii and Japan. Sequences for Pacific collections were combined with those for European taxa in phylogenetic analyses using the maximum parsimony criterion. The results provide improved understanding of species diversity and biogeography in *Ulva* and *Enteromorpha* and additional evidence for the polyphyly of these genera.

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TRANSFERRING AND EXPRESSION OF GLUCOSE OXIDASE GENE - *GLUC* IN *PORPHYRA YEZOENSIS*

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The disease-resistant gene *gluc*, encoding glucose oxidase, has been used to increase advance crops’ resistance to diseases. Now the *gluc* gene has been reconstructed in the plasmid pYP1203 between *gus* and cm$^+$(*cat*) gene, a reporter gene and an antibiotics resistance labeling gene within the RB (right border) and LB (left border). With PEG and electroporation methods, the pYP1203 with *gluc* gene and *gus*, *cat* genes was transferred into the cells from thalli of *Porphyra yezoensis*. After 24-72 hours, the GUS detection in transgenic cells of *Porphyra yezoensis* was positive. Cell transformation efficiency could reach as high as about 3-4%. The expression of *gus* gene in the filaments regenerated from transgenic cells was also detected. Furthermore with the chloromycetin resistance screening, the transgenic cells of *Porphyra* survived over one month in the 50 ug/ml of chloromycetin concentration, while the wild cells could not survive in the same chloromycetin concentration. That results indicated that the *gluc* gene might be integrated into the chromosomes of *Porphyra yezoensis*. The molecular test of these transgenic algae and their disease resistance analysis are continuing.

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SALINITY AFFECTS HIGH TEMPERTURE TOLERANCE IN BROADLY HALOTOLERANT ISOLATES OF *NANNOCHLORIS* AND *DUNALIELLA*.

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I studied the effect of growth salinity on short-term high temperature stress in new strains of broadly halotolerant *Nannochloris* and *Dunaliella* isolated from the Salt Plains National Wildlife Refuge in Oklahoma, USA. Photochemical yield (pulse modulated fluorescence) was unaffected by growth salinity in both algae. *Dunaliella* was progressively more heat tolerant (2 h at 41.5 °C) with increasing salinity. Photochemical yield of cells at 100 and 50 g/L was inhibited by about 15 and 40%, respectively, and largely recovered within 30 min after return to 23 °C. Thermal inhibition (2 h at 45 °C) of photochemical yield in *Nannochloris* was about 45% at both 50 and 100 g/L, but recovery was slower at 100 g/L, and slower at both salinities compared to *Dunaliella*. At 20 g/L, both species were almost 90% inhibited by high temperature and required more than 24 h to recover. Salinity alone had little effect on in vivo absorption spectra, ambient and 77 K fluorescence excitation and emission spectra. However, 2 h heating increased the photosystem (PS) I:PS II emission ratio (714:690 nm) at all salinities. This ratio largely recovered within 24 h in *Dunaliella* at 50 and 100 g/L salinity, and partially recovered in *Nannochloris* at 100 g/L. Cells of both species heated at 20 g/L exhibited chlorosis the next day, but regreened after several days.

These results are consistent with steady-state temperature-salinity experiments with *Nannochloris*.

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NICHE ADAPTATION OF *PROCHLOROCOCCUS* ECOTYPES: INSIGHTS THROUGH COMPARATIVE GENOMICS

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Prochlorococcus is an extremely small, chlorophyll *b*-containing oceanic cyanobacterium. Specific ecotypes of *Prochlorococcus* have adapted to different ecological conditions with regard to factors as light or the available nutrients. Such differentially adapted ecotypes are less than 3% divergent in their 16S rRNA sequences, which invites speculation as to how their specific gene content has diverged to reflect the particular niche of each strain. Complete genome sequences have been determined of two *Prochlorococcus* strains, MED4 and MIT9313. These two strains are representatives of high and low light-adapted ecotypes. Intriguing similarities between both genomes include their small size and compact organization (MED4: 1.7 Mbp and MIT9313: 2.3 Mbp), a gene cluster for RubisCo and carboxysomal proteins that is of obviously non-cyanobacterial origin, or genes for two different lycopene cyclases explaining how *Prochlorococcus* synthesizes alpha-carotene, a carotenoid that is not common to cyanobacteria. Several genes and operons which in cyanobacteria are involved in light harvesting, nitrate utilization or in the generation of circadian rhythms have been reduced to a different degree in the two compared genomes. MED4 has many more genes encoding high light inducible proteins and photolyases. In contrast, MIT9313 possesses more genes to build up more complex light harvesting structures, including a gene cluster to produce chromophorylated phycoerythrin. The latter represents an intermediate between the phycobiliproteins of non-chlorophyll *b* containing cyanobacteria and a degenerated phycoerythrin present in MED4. Screening of natural samples from the Red Sea suggests that a highly similar phycoerythrin form is wide-spread among high light-adapted ecotypes.

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CHARACTERIZATION OF THE ADHESIVE MUCILAGES SECRETED BY LIVE DIATOM CELLS USING ATOMIC FORCE MICROSCOPY

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Atomic Force Microscopy (AFM) resolved the topography and mechanical properties of the adhesive mucilages secreted by living cells of the fouling diatom Craspedostauros australis. Tapping mode images of live cells revealed a soft, cohesive mucilage layer that encased most of the diatom silica cell wall. Force curves revealed that this type of mucilage had an adhesion strength of 71.2 mN m-1 and a compressibility of 5.7 x 106 N m-2. High force contact mode imaging (i.e., increased set-point voltage) resulted in the 'raking' of the cohesive mucilage to reveal the underlying, silicified wall structures. Further imaging of cantilever 'cleaned' cell walls of live cells in stationary growth phase revealed the active secretion of soft mucilage via pore openings in the girdle bands. A second adhesive mucilage in the form of distinct strands was found to project through the silica valve walls at the raphes, and to be involved in cell adhesion and motility. Single force spectroscopy measurements revealed that an adhesive strand (or strands) was resistant to breaking forces greater than 40 nN and could only be detached from the AFM cantilever probe using mechanical retraction of the piezo. Using the Worm-like Chain model (WLC) the chain length and persistence length of the strands were calculated to be 3μm and 0.04Å, respectively. These values imply that the strands are very adhesive and strong compared to other single biological polymers, and could account for the complex interactions between diatom cells and their substratum.

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EFFECTS OF BARLEY STRAW EXTRACT ON GROWTH OF FIVE SPECIES OF PLANKTONIC ALGAE

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The effects of exposure to barley straw extract and the timing of exposure on the growth of four common cyanophyte species and one species of green algae were investigated in two laboratory experiments. Clonal cultures of *Anabaena cylindrica*, *Cylindrospermum* sp., *Gloeocapsa* sp., *Eucapsis* sp., and *Chlorella vulgaris* were obtained from culture collections. In both experiments, the algae were cultured in Guillard's WC medium at 20 °C on a 12:12 L/D photoperiod. In the first experiment, the algae were dosed with four concentrations of barley straw extract at the beginning of the experiment (day 0) and growth was monitored every second day using fluorometric detection of chlorophyll *a* for 14 d. In the second experiment, the algae were dosed with the same extract concentrations, but the extract was not added until the algae were in exponential growth phase (day 6). Both experiments also had control treatments (i.e. no extract) and each extract and control treatment was replicated five times. Growth of *C. vulgaris* was inhibited by all doses in both experiments, but inhibition was 22% greater when the extract was added on day 0. Growth *Gleocapsa* sp. was slightly inhibited by all doses when the extract was added on day 0, but not when it was added on day 6. No other species were inhibited, regardless of dose or timing of dose. The results of this study and other bioassay studies suggest that differential susceptibility to barley straw among algae is common and may reduce the effectiveness of barley straw as an algal control technique.

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BIOGEOGRAPHY OF MARINE RED ALGAE: MYTHS AND REALITIES

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Theories about the geographical distribution of marine algae fall roughly into two categories: (1) a concept of biogeographical regions in which algal distribution is determined primarily by growth, reproductive and lethal temperature boundaries (Setchell, van den Hoek, Breeman, Lüning) and (2) an historical perspective in which distribution is determined primarily by patterns of dispersal and the establishment of barriers to dispersal (vicariance biogeography) (Svedelius, Garbary, Lindstrom, Hommersand). Setchell proposed the 5° isotherm rule in 1920, and in 1924 Svedelius advocated a worldwide distribution for tropical and subtropical groups followed by discontinuous distribution upon closure of the connection between the Indian Ocean and Mediterranean Sea and, later, between North and South America (Wegener's theory). Transarctic dispersal routes have received special attention in recent years (Lindstrom, Lüning, van Oppen, Olsen, Stam), as have special relationships between Australasia, South Africa and South America (Hommersand). Less well understood are the climatic changes that have taken place in the Cenozoic which are strategic to an understanding vicariant biogeography. The advent of molecular methods combined with the tools of phylogenetic systematics now make it possible to identify ancestral taxa, test the consistency of tree topologies, and calculate mean branch lengths between sister lineages diverging from an interior node of a tree. With such methods it may be possible to infer ancestral areas, identify dispersal pathways, determine the chronology of isolating events, assess the impact of multiple invasions, and generally relate dispersal and vicariance models to phylogenetic hypotheses for red, brown and green algal taxa.

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PRODUCTION AND RELEASE OF GEOSMIN BY THE CYANOBACTERIUM *OSCILLATORIA SPLENDIDA* ISOLATED FROM A PHOENIX WATER SOURCE

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Geosmin is a common component of the off-flavors detected in the drinking water supply sources of metropolitan Phoenix (Arizona). A cyanobacterium, *Oscillatoria splendida*, was isolated from source water during incidents of elevated geosmin production and was implicated as a cause of earthy/musty off-flavors in the drinking water. Production of geosmin was found to be constitutive in *O. splendida* during all growth stages. Effects of environmental parameters on the growth characteristics, and on production and release of geosmin by *O. splendida*, was studied under laboratory conditions. The specific growth rate and cell-bound geosmin increased with increasing temperature from 12 to 26 ^{0}C, the range of water temperatures that occur in the drinking water supply. On a per-chlorophyll *a* basis, however, more geosmin was released from the cells at lower temperatures. An inverse relationship was evident between light intensity and *O. splendida* growth and the release of geosmin. Cell-bound geosmin, however, was higher at higher light intensities. Dark incubation initially stimulated the biosynthesis of geosmin, whereas a prolonged period of darkness (2-3 weeks) resulted in massive release of geosmin into the culture medium from lysis and cellular decomposition. Dissolved nitrogen appeared to be the limiting nutrient for *O. splendida* in the local water supply source. When nitrate was added to laboratory cultures, both growth and geosmin production increased. These results will be discussed in context with episodes of off-flavors in drinking waters in metropolitan Phoenix, Arizona.

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DO DIATOM DIETS ACT AS BIRTH CONTROL PILLS FOR MARINE COPEPODS?

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Traditionally, diatoms have been regarded as providing the bulk of the food that sustains the marine food chain to top consumers and important fisheries. However, this view has recently been challenged on the basis of laboratory studies showing that these small, unicellular algae possess anti-mitotic properties similar to the cytotoxic compounds isolated from numerous marine and terrestrial higher plants. In fact, when copepods, the principal predators of diatoms, are fed diatom diets, they produce abnormal eggs that either fail to develop to hatching or hatch into malformed nauplii that die soon afterwards. The aldehydes responsible for this anti-cell growth activity have recently been isolated and these compounds have been shown to arrest not only the development of copepod and sea urchin embryos, but also the proliferation of human carcinoma cells. In terrestrial environments, there are many reports of secondary metabolites produced by plants that interfere with the reproductive capacity of grazing animals, and which act as a form of population control. But this type of biological model is new for the marine environment where most of the attention on plant-animal interactions has focused on feeding deterrents and poisoning compounds. Such "birth-control" compounds may discourage herbivory by sabotaging future generations of grazers, thereby allowing diatom blooms to persist when grazing pressure would normally have caused them to crash.

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PRELIMINARY RESULTS ON SECONDARY METABOLITES FROM ANTARCTIC BROWN ALGAE AND THEIR ECOLOGICAL RELEVANCE

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Brown algae (Phaeophyceae) are often dominant members in abundance and biomass of Antarctic macroalgal communities. They are ecologically important in providing habitat structure and shelter to many invertebrates and fish. However, as live algae they often appear to be of little importance as food for herbivores. Palatability of thalli of eight abundant Antarctic brown algae was tested against two sympatric algal grazers, the fish *Notothenia* coriiceps and the sea star *Odontaster validus*. Polar and non-polar organic extracts of six brown algae were also tested against these grazers and against the omnivorous amphipod *Gondogeneia antarctica*. Almost all thalli and at least one of the extract types per algal species were unpalatable to the grazers, suggesting that chemical defenses are probably present in Antarctic brown algae. We further tested biological activity of brown algal extracts in antimicrobial assays as well as antifouling assays with sympatric bacteria and diatoms, respectively. We chemically screened organic brown algal extracts for the occurrence of secondary metabolites. Acetogeno-terpenes were detected in *Desmarestia menziesii* and *D. anceps*. Phlorotannins were present in all brown algal species analyzed, but at different concentrations between species as well as between different thallus parts within individuals. Phlorotannin extracts were size fractionated to examine distribution and abundance of various size classes between species as well as between different thallus parts within individuals.

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THREE POSSIBLE NEW SPECIES OF *NITZSCHIA HASSALL* (BACILLARIOPHYTA) FROM THE SHEYENNE RIVER, ND.

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The Sheyenne River is a third order river that flows through central to eastern North Dakota. The first study of this river's periphyton was completed in the summers of 1998-1999. One hundred sixty diatom species were identified. *Navicula* and *Nitzschia* were the most abundant genera. Among this group were three Nitzschia that appear to be new species. These three species were studied extensively using light microscopy. During this examination, one hundred specimens were measured. Preliminary results indicate that *Nitzschia* sp.1 belongs to the sect. Obtusae and *Nitzschia* sp. 4 and sp. 5 belong in the sect. Lanceolatae. These specimens will be examined using scanning electron microscopy in a future study.

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DISTRIBUTION OF EPIPHYTIC BACTERIA ON CELLS OF *PSEUDO-NITZSCHIA* MULTI-SERIES IN CULTURE

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Toxicity of the domoic-acid-producing pennate diatom *Pseudo-nitzschia multiseries* is enhanced by bacterial association. Yet, apart from initial molecular studies nearly a decade ago, the diversity of bacteria in cultures of *P. multiseries* has not been explored. The goal of this study was to examine the frequency and distribution pattern of epiphytic bacteria growing on cultured cells of *P. multiseries*. Diatoms were grown in flasks of f/2 medium for 31 days. Every 5-12 days, 5 one-mL subsamples were transferred to a filtration apparatus and grown undisturbed for 2 days in order to maintain natural associations between the diatom host and its bacterial epiphytes. The subsamples were then gently prepared for SEM examination. From 15-40% of the diatom cells had attached bacteria. Most diatom cells carried only 1-5 bacteria, although some had up to 74 bacteria. Even in late stationary phase, many diatom cells were bacteria-free. A morphologically diverse bacterial flora was observed attached to the host diatoms, including: 1) stalked bacteria, 2) rod-shaped bacteria attached by their narrow side, 3) rods attached along their long side, and 4) cocci. Stalked bacteria were most common on the girdle band junctions of the cell. They were also attached to the

raphe and striae. Rods resting on their long side were most common on the striae of the valve face. These preliminary results indicate that cultured diatoms co-exist with a diverse bacterial epiphytic flora, having a specific distribution pattern and frequency. Their effect on domoic acid production has yet to be determined.

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ISOLATION OF PHASE-SPECIFIC COMPLEMENTARY DNAS FROM CARPOSPOROPHYTES OF *GRACILARIOPSIS LEMA-NEIFORMIS* USING MAGNETIC BEADS AND PCR.

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Although the morphology and developmental patterns of carposporophyte stage have been investigated well, there are few molecular, genetic, or biochemical data about this stage. The greatest obstacle to this research has been that the conventional methods to isolate tissue-specific genes require a lot of tissues, but the carposporophyte is very tiny and mostly embedded in female gametophyte tissues. Recent advanced techniques have allowed the subtractive cloning of differentially expressed genes from small amounts of tissue or cells. We applied the subtractive hybridization method using magnetic beads and PCR to the analysis of phase-specific cDNAs from carpo-sporophytes of *Gracilariopsis lemaneiformis* (Graci-lariales, Rhodophyta). A hundred cystocarps were dissected to isolate gonimoblast tissues, and total RNAs were extracted from the gonimoblast tissues and the female gametophyte branches, respectively. Messenger RNAs were captured on paramagnetic oligo-dT beads, followed by first-strand cDNA synthesis on the beads. Three rounds of subtractive hybridization between the amplified second-strand carposporophyte cDNA in solution and the first-strand gametophyte cDNA attached to magnetic beads were sufficient to remove common genes present in both gametophyte and carposporophyte stages. A specific PCR product from the nuclear GAPDH gene was readily amplified from gametophyte and carposporophyte cDNA, but no amplification was observed using the subtracted carposporophyte cDNA as template. This control PCR product demonstrates that the hybridization steps successfully removed the common GAPDH cDNA, which is found in both stages, giving confidence that the remaining genes cloned from the subtracted carposporophyte cDNA library are stage-specific.

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PHYLOGENETIC ANALYSES OF TRIBE NITELLEAE (CHARACEAE) USING RBCL SEQUENCE DATA.

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The tribe Nitelleae (Characeae) currently includes two genera, *Tolypella* and *Nitella*. Wood and Imahori's monograph of the Characeae divides *Tolypella* into two sections and *Nitella* into three subgenera with seventeen sections. We determined sequence data for the gene rbcL from representatives of both sections of *Tolypella* and the three subgenera of *Nitella* to test the monophyly of these groups. Phylogenetic analyses support the monophyly of the Nitelleae as well as *Tolypella* and *Nitella*. Both sections of *Tolypella* were also found to be monophyletic. Within *Nitella*, subgenera *Hyella* and *Tieffallenia* formed monophyletic groups sister to each other. Subgenus *Nitella*, however, formed two paraphyletic lineages, one including *Nitella* acuminata, *N. bastinii*, *N. clavata* and *N. praelonga* and the other including *N. opaca* and *N. stuartii*. Subgenus *Nitella* is defined by one-celled dactyls terminating the branchlet rays, while subgenera *Hyella* and *Tieffallenia* exhibit more complex dactyl morphology. The molecular data appear to resolve certain inconsistencies in Wood's classification. For example, in the rbcL analysis, *N. hookeri*, a multiple-dactyl species that has nevertheless been placed in subgenus *Nitella*, falls in a clade characterized by multiple-dactyl species. Thus, although the traditional sectional classification of Wood does not divide the genus into monophyletic groups, a combined morphological and molecular approach holds promise for erecting a natural classification of *Nitella*.

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EFFECT OF LOADING RATES ON NITROGEN REMOVAL EFFICIENCY AND NITRIFICATION WITHIN ALGAL TURF SCRUBBERS

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A potential alternative to land application of livestock manures for crop production is the production of algae to recover the nitrogen (N) and phosphorus (P) present in the manure. The specific objectives of these experiments were to test the effects of different loading rates of anaerobically digested dairy manure on nitrogen removal efficiency and nitrification within algal turf scrubbers (ATS). Laboratory-scale ATS units were operated by continuously recycling 220 l of wastewater and adding manure effluents daily. The algal turfs contained mixed indigenous assemblages of benthic algae. The most abundant genera were *Ulothrix*, *Oedogonium*, and *Rhizoclonium.* Weekly harvest of algal biomass, and wastewater samples were analysed for total

Kjeldahl nitrogen (TKN), ammonium (NH_4-N), nitrate, and elemental composition. In previous experiments with loading rates of 0.6–0.96 g TN m^{-2} d^{-1}, algal nitrogen accounted for 42–50 % of input NH_4-N. Nitrate production accounted for the bulk of remaining input NH_4-N. Lower loading rates (0.24 and 0.48 g TN m^{-2} d^{-1}) tested here showed a higher removal rate where algal nitrogen accounted for 71% of input NH_4-N (56% of TN), and nitrate production was negligible. The disappearance of NH_4-N from wastewater in scrubbers measured over 4–5 hours showed initial rates of about 6 mg NH_4-N hr^{-1} g^{-1} DW algae, corresponding to 3.7 g NH_4-N d^{-1} m^{-2}. This was followed by slightly lower rates, probably indicating limitation in carbon and/or micronutrients.

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VERTICAL MIGRATION OF *EUGLENA VIRIDIS* ON SCRIPPS BEACH, LA JOLLA, CALIFORNIA

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The upward migration of millions of benthic *Euglena viridis* cells forms expansive green patches covering Scripps Beach during daytime low tides. Scripps Beach is an open ocean beach characterized by high-energy wave action. As the rising tide approaches, the green patches fade as the population migrates down into the sediment. Between July 18 and July 23, 2000, light experiments were conducted to evaluate the effect of incident irradiance on the vertical migration of *E. viridis*. Neutral-density filters were used to reduce the incident light levels to 56%, 22%, and 2% of incident irradiance. Opaque film canisters were used as a dark treatment. Microscopic examination of surface samples was used to tally the number of *E. viridis* under 5 replicates of each light treatment arranged in a Latin square design. Sediment weights were used to calculate the number of *E. viridis* per mg of sand collected. One-way ANOVA revealed a statistically significant effect of light. The 100% incident irradiance and dark treatments resulted in significantly lower surface cell densities. Sediment cores were collected and sectioned to determine the vertical distribution of the population during high and low tides. The cell counts of the core sections revealed that large numbers of viable cells were found 15 cm below the surface during high tide. The vertical migration pattern of the Scripps Beach population of *E. viridis* will be contrasted with the pattern observed in diurnally migrating *E. viridis* populations living on the banks of small streams in central North Carolina.

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MORPHOLOGICAL AND MOLECULAR CHARACTERIZATION OF FOUR *NOSTOC* SPECIES ISOLATED FROM DESERT SOILS

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Four strains of *Nostoc* were isolated and characterized with regards to morphology and life cycle. Three strains were isolated from microbiotic soil crusts in the Mojave Desert of Southern California, and one strain was isolated from the lichen *Collema tenax* from crusts in Canyonlands National Park. All four strains were found to be morphologically distinct, with differences in: macrocolony appearance, microcolony shape and size, density of trichomes within the microcolony, and hormogonia production. The most commonly reported species from desert soils is *Nostoc commune* Vaucher. However, our strains all differ from that taxon in the size and shape of akinetes and heterocysts. *Collema tenax* is reported to have *Nostoc commune* as its phycobiont, but our observations on the phycobiont indicate that it is very distinct from that species and other *Nostoc* we have observed in desert soils. We subsequently sequenced a partial sequence of the 16SrRNA gene and the associated internal transcribed spacer (ITS) between the 16SrRNA and 23SrRNA genes for all four strains. Evidence of three multiple rRNA operons each was recovered for two strains, based upon configuration and sequence differences in the ITS regions. ITS with the following configurations were observed: no tRNA genes, $tRNA^{Ile}$ and $tRNA^{Ala}$ genes, and the beginning portion of $tRNA^{Ile}$ immediately followed by the end portion of $tRNA^{Ala}$. Although our strains appear distinct from the descriptions of species in Geitler (1932) and Starmach (1966), further molecular characterization of species in this genus is probably warranted before they are assigned to existing or new species.

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GENETIC VARIABILITY AND STRUCTURE IN POPULATIONS OF THE KELP *POSTELSIA PALMAEFORMIS*

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Available experimental and observational data suggest that the sea palm, *Postelsia palmaeformis*, has short spore dispersal distances. We hypothesized that this dispersal pattern would produce substantial genetic structuring in local populations and identifiable genetic lineages . We sampled individuals from populations 30 m to 2 km apart using Amplified Fragment Length Polymorphism (AFLP). We present methods for successfully carrying out AFLP on *Postelsia*, and give an overview of our initial analyses. As predicted by the life-history of *Postelsia*, we find evidence of significant geographic differentiation (Fst) of *Postelsia* populations from as little as 30 m apart. In addition, we outline an experiment to determine parent-offspring relationships.

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PHOTOSYNTHETIC ABILITIES OF FUCOID ALGAE DURING LOW TIDE: DIFFERING RESPONSES OF ADULTS AND YOUNG STAGES

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On the intertidal rocky shores of the Gaspé Peninsula (Québec), three species of fucoid algae (*Ascophyllum nodosum*, *Fucus vesiculosus*, *Fucus distichus*) form distinct zones. One possible underlying cause of this pattern is that the upper limit of each species determined by physiological stresses, especially that experienced by younger stages which are likely to be more susceptible. To assess this possibility, we investigated the photosynthesis activity of these macroalgae using chlorophyll *a* fluorescence as measured by pulse-amplitude modulated fluorometry (PAM). Measurements of the responses of both adult and early stages (3-wk old) were made in the field during low tide under different meteorological regimes, including sunny and windy conditions. In general, algae became more photoinhibited (i.e., reduced photosynthetic efficiency, Fv/Fm) with increasing levels of insolation and desiccation, and early stages showed a more dramatic response than did the adults. Recovery was rapid, especially for the embryos. During sunny, calm conditions adults of the species found lowest on the shore (*F. distichus*) became photoinhibited more rapidly than those at higher levels. In contrast, the embryos of the highest species (*A. nodosum*) were the most susceptible to stresses experienced during emersion, especially high levels of insolation. Thus, whereas the physiological characteristics of the adults were consistent with their patterns of vertical distribution, those of the early post-settlement stage were not, suggesting possible ontogenetic shifts in their capacities to response to environmental conditions.

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FLOW-INDUCED CHANGES IN DINOFLAGELLATE MEMBRANE FLUIDITY

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Dinoflagellates are among the most flow-sensitive organisms known, with physiological responses including changes in swimming, population growth, morphology, and the stimulation of bioluminescence. The mechanotransduction pathways involved in flow sensing by dinoflagellates are unknown. Studies of flow-sensitive mammalian cells have suggested that the plasma membrane is a primary mechanosensor and that flow-induced changes in membrane fluidity activate signal transduction pathways. In the present study, flow-induced changes in membrane fluidity of the dinoflagellate, *Lingulodinium polyedrum*, were monitored using the dye DCVJ (9-(dicyanovinyl)-julolidine), which has a fluorescence yield that is proportional to viscosity. Flow exposure resulted in a reversible decrease in DCVJ fluorescence consistent with an increase in membrane fluidity. The change in fluorescence occurred on the time scale of seconds suggesting it was not due to changes in the cytoskeleton. The magnitude of fluorescence decrease was proportional to flow rate as previously described for mammalian endothelial cells. The similar flow-induced changes in membrane fluidity for dinoflagellates and mammalian cells suggest shared mechanotransduction mechanisms.

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AN UNUSUAL POLYUNSATURATED C-27 HYDROCARBON FROM THE MARINE DINOFLAGELLATE *PYROCYSTIS LUNULA*

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Studies of the lipids of different algal species have revealed a diversity of fatty acids, sterols, and hydrocarbons, of which several are considered useful biomarkers, with potential for characterizing phytoplankton community composition. To extend this approach and characterize the lipids and lipid classes of laboratory-cultured marine dinoflagellates, a silicic acid fractionation system was developed to obtain compositional data for sterols and hydrocarbons of over forty species. In the course of this work, a neutral fraction obtained from a lipid extract of *Pyrocystis lunula* was found to contain an abundant quantity of a long-chain polyunsaturated hydrocarbon, along with previously reported keto-steranes. The hydrocarbon molecular weight (364) and retention time obtained by gas chromatography/mass spectrometry analysis suggested a C_{27} compound, which was confirmed by reduction (Adams catalyst) to give the straight chain alkane, *n*-heptacosane. The presence of eight double bonds was established by deuteration to give a product with molecular weight 396. While the positions of double bonds have not been established, the carbon number of this hydrocarbon and the number of double bonds strongly suggest formation by decarboxylation of the recently described, long-chain polyunsaturated C_{28} fatty acid shown to be a constituent of phospholipids. This hydrocarbon was not found in any other genus of the examined dinoflagellates, and appears to be one of the first identifications of a hydrocarbon in this class of algae. The function(s) of this compound in *P. lunula* is currently unclear.

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POPULATION DYNAMICS AND THE TOXICITY OF BLUE-GREEN ALGAE IN THE NAKTONG RIVER, KOREA

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Seasonal and spatial variations of phytoplankton community were monitored at 16 sites along the Naktong River. Blue-green algae appeared from May through November with dominant genera of *Microcystis, Anabaena, Oscillatoria* and *Gomphosphaeria.* Frequency and biomass of the genera became greater at lower reaches. The *Microcystis* were observed from May to October up to 85,750cells/ml. Six species of *Microcystis* were identified with morphological characteristics and *M. aeruginosa* was most dominant. There were significant relationships between biomass of *Microcystis* and NO_3^-, TP and pH in water column. However, NH_4^+, PO_4^{-3} and N/P were not critical in successions to the *Microcystis* dominated community. *Microcystis* blooms were notable at water temperature higher than 25¡É. *Microcystis* density in sediment was 3 orders of magnitude higher than water column. *Anabaena* density ranged up to 11,220cells/ml. Four species of *Anabaena* were identified and *A. flos-aquae* was most dominant. *Anabaena* biomass was not related to temperature, NO_3^-, TN, PO_4^{-3}, TP and N/P of water column and the frequency of trichome with akinete and/or heterocyte were not related to these parameters. Microcystins were detected from May to November with yearly fluctuations. Microcystin-RR was most dominant. Total 84.2% of algal materials with *Microcystis* exhibited toxicity of microcystin with maximum of 3,292µg/g dry wt. Total 12.5% of water column with *Microcystis* exhibited dissolved microcystin up to 3.3µg/l. Microcystin concentrations were positively related to *Microcystis* biomass and pH of water column. Anatoxin-a was determined by FD-HPLC analysis with NBD-F and all concentrations were below the detection limit of 0.1µg/l.

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THE END-PERMIAN MASS EXTINCTION WAS SYNCHRONOUS COINCIDED WITH THE EVOLUTION OF TOXIC ALGAE

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The cause of the end-Permian mass extinction, the greatest of all mass extinctions, is one of the most intriguing mysteries in the history of life. The end-Permian mass extinction was primarily a marine event, resulting principally in the elimination of sessile filter-feeding organisms. Based on two methods, molecular clocks and correlation with ancient atmospheric CO_2, the algae derived from secondary endosymbioses are believed to have originated in the middle to late Permian. All of the toxic marine algae in today's oceans are derived from secondary endosymbioses. Therefore it appears likely that the end-Permian extinction was due to the evolution of toxic algae in the phytoplankton of late-Permian seas. Sieving of the toxic algae in the phytoplankton likely resulted in the decline and eventual elimination of a large portion of the Paleozoic fauna during the end-Permian mass extinction.

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TWO CHLOROPLAST TRANSFER RNA INTRONS FOUND IN *CHAETOSPHAERIDIUM* SUPPORT A MONOPHYLETIC COLEOCHAETALES.

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The evolutionary relationships of the algal genera *Mesostigma* and *Chaetosphaeridium* to other algae and land plants are currently controversial. A close evolutionary relationship between land plants and two orders of the charophycean algae, the Charales and Coleochaetales, is supported by morphological, ultrastructural, biochemical, genomic, and phylogenetic data. A number of phylogenetic analyses support a monophyletic Coleochaetales, with Coleochaete and *Chaetosphaeridum* as sister groups. *Mesostigma* was traditionally viewed as a member of the prasinophytes and has recently been considered as a lineage possibly basal to the charophycean algae, or sister to all green algae. By contrast, recent analyses of small subunit ribosomal RNA gene sequences have been interpreted as evidence of an alternative classification with *Mesostigma* forming a clade with Chaetosphaeridium to the exclusion of *Coleochaete* and other charophycean lineages. The shared presence of introns in two chloroplast tRNA genes (tRNAAla and tRNAIle) among charophytes *Coleochaete* and *Nitella* and the liverwort *Marchantia* supports a monophyletic group containing the Coleochaetales, the Charales, and land plants. Through isolation and sequence analysis of the tRNAAla and tRNAIle genes in *Chaetosphaeridium*, we have identified introns similar in sequence and position to those found in *Coleochaete*. These data and the published absence of these introns in *Mesostigma* lend new support to a monophyletic Coleochaetales including the genera *Coleochaete* and *Chaetosphaeridium.*

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A NOVEL GENE TRANSFER IN *PORPHYRA*

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Foreign genes have been successfully transferred into both animal and plant by transgenesis, but introduction of foreign genes into many important algae species is still in its infancy. Although nuclear and organellar transformations in *Chlamydomonas reinhardtii*, a green microalgal species, have been well established, attempts to develop transformation methods for microalgae, notably the colonial green alga *Volvox carteri* and several diatom species have just begun. Currently, there is no report of success of genetic transformation in the commercially important red algae, *Porphyra*. To facilitate applications of seaweed in both basic research and integrated aquaculture biotechnology, we have undertaken initiatives to develop gene transfer technology for *Porphyra*, and made significant progress. A plasmid DNA with a selection marker was transferred into archaeospores of *P. leucosticta* by electroporation. Total DNA was then prepared from those electroporated samples at various times of incubation including 24 hrs, one week, two weeks and up to eight weeks. PCR analysis showed the consistent presence of plasmid DNA in all samples tested, strongly suggesting the success of gene transfer in *Porphyra*. The thalli developed from those transformed archaeospores grow well under continuous presence of selection pressure. These observations provide us with great confidence that we will be able to perform gene transfer studies for *Porphyra* and produce transgenic *Porphyra* thalli.

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CLONING AND DIFFERENTIAL EXPRESSION OF PROLIFERATING CELL NUCLEAR ANTIGEN IN THE COCCOLITHORID PHYTOPLANKTON *PLEUROCHRYSIS CARTERAE*

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Proliferating cell nuclear antigen (PCNA) is an auxiliary protein for DNA polymerase- and - , and is potentially a useful marker for cell cycle and growth rate studies for marine phytoplankton. In this study, the gene coding for PCNA (*pcna*) was identified in the marine coccolithophorid microalga *Pleurochrysis carterae*. Two mRNA forms, designated as *Pcpcna*1 and *Pcpcna*2, contained an identical coding region for 222 amino acid residues followed by an untranslated sequence of 302 bp (Ut1) and 246 bp (Ut2), respectively. Genomic DNA sequencing revealed six exons and five introns in this gene, an unusual structure first described for *pcna*. The coding region of *Pcpcna* is highly similar to counterparts in other organisms. The PC10 antibody strongly cross-reacted with the fusion protein of *Pcpcna* over-expressed in *E. coli* as well as crude protein extract from the alga. As detected with quantitative reverse transcription-polymerase chain reaction (RT-PCR) using specific primers and western blotting using PC10, PCNA in this alga is expressed significantly more abundantly during the exponential growth phase than the stationary growth phase. Furthermore, the two mRNAs appeared to be expressed differentially; *Pcpcna*1 decreased and dis-appeared before *Pcpcna*2 when cultures entered stationary growth phase both at salinities of 28%o and 15 %o. Immunofluorescence labeling demonstrated that this protein was present in the nucleus. These results demonstrate that PCNA is unique in terms of structure in this coccolithorid phytoplankton and the expression of the two mRNA species is differentially regulated relative to growth stages.

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INFLUENCE OF SALINITY AND TEMPERATURE ON GROWTH AND PHOTOSYNTHESIS IN THE EXTREMOPHILIC CHLOROPHYTE, *NANNOCHLORIS* SP.

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Preliminary data suggest *Nannochloris* sp., isolated from the Great Salt Plains National Wildlife Refuge, is a true extremophile. This alga is able to withstand salinities ranging from 0 to 150 $^{o}/_{oo}$ and temperatures up to 45^{o}C. To test the hypothesis that acclimation to high salinity confers tolerance to high temperature, experimental cultures were acclimated to salinities of 25 and 100 $^{o}/_{oo}$ and/or temperatures of 23 and 38^{o}C; irradiance (500 mol photons m^{-2} s^{-1}) was saturating for both growth and photosynthesis. Cells acclimated to low salt and low temperature exhibited high photosynthetic performance in terms of both light-saturated photosynthesis (P_{max}; 45.0 fmol O_2 cell^{-1} h^{-1}) and light-harvesting efficiency (; 0.103 fmol O_2 cell^{-1} h^{-1}/ mol photons m^{-2} s^{-1}). However, high-salinity cells exhibited values for net P_{max} (18.1 fmol O_2 cell^{-1} h^{-1}), (0.107 fmol O_2 cell^{-1} h^{-1}/ mol photons m^{-2} s^{-1}) and growth rates (*ca*. 0.4 d^{-1}) that were equal to, or higher than, those of low-salinity cells when acclimated to high temperature. Both the amount of light required to achieve net photosynthesis (I_c) and that required to achieve light-saturated photosynthesis (I_k) were lower in high-salinity cells than those exhibited by low-salinity cells grown at high temperature; reductions in I_c and I_k were primarily due to increases in light-harvesting efficiency. We propose that an increase in growth temperature might release *Nannochloris* sp. from energy constraints associated with osmolyte production and low-temperature effects on enzyme activity. These data are consistent with effects of short-term temperature stress on Chl *a* fluorescence kinetics in this alga.

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TRACE GAS PRODUCTION BY SEAWEEDS: DEFENSE, OXIDATIVE STRESS, SIGNALLING AND ATMOSPHERIC SIGNIFICANCE

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For some years it has been known that seawater contains a range of volatile halogenated compounds which are produced by phytoplankton and macroalgae. The total production for some of these compounds is estimated to be of the order of Tg per year, and the resulting flux of trace gases to the air is highly significant for global atmospheric chemistry, since the volatiles are an important source of halogen free radicals which impact on oxidant chemistry and participate in ozone destruction reactions. Ongoing research seeks to determine the physiological basis of the production of halogens by seaweeds. However, debate continues as to whether the algal production of these trace gases has a particular functional value or should be considered an 'accident' of photoautotrophy which serves to sequester reactive oxygen species. This talk will focus on our recent research on the production of volatile iodinated halocarbons and inorganic iodine species by sporophytes of *Laminaria digitata*. In this species elicitor compounds such as Oligoguluronates (oligomeric degradation products of cell wall alginate), trigger a strong, transient burst of active oxygen species, which is recognised as a ubiquitous defence characteristic in animal and higher plant phyla. In *Laminaria digitata*, which accumulates very high levels of iodine the oxidative burst rapidly induces a massive efflux of inorganic iodine which is accompanied by elevated production of volatile iodinated compounds. Our data also suggest that the iodine release is a component of the seaweeds defence system against pathogens.

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CHANGES IN ALGAL COMMUNITY SUCCESSION IN DIFFERENT NUTRIENT REGIMES

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The purpose of our experiment was to assess changes in relative abundances of algal species during community development under different nutrient concentrations. In artificial streams N and P concentrations were manipulated, when P and N were saturated respectively. Mean algal relative abundance per treatment was assessed. Algal species with relative abundances at least 10 % in any treatment were analyzed in detail. More detailed response was discerned based on the species composition of the benthic algal community than results from total density.

Cymbella affinis Kutz. had significantly high (ANOVA, $p<0.05$) presence in all low nutrient concentrations. The highest relative abundance of this species was 33% at 64 μg/L N and 43% at 2 μg /L P and decreased with increase in the nutrient concentrations. Along the N gradient we had significant ($p<0.001$) algal species response to the six treatments. *Microcystis aeruginosa* Kuetz., was present at low and median N concentrations. Some green filamentous algae and known euthrophic diatom species like, *Nitzschia palea* (Kutz.) W. Smith, were dominant at high N concentrations. Along the P gradient the response of the community was again significant ($p<0.001$) in all treatments. Diatoms had the highest relative abundances overall; coccoid bluegreen and filamentous green were present when P was greater than 32 μg /L. Following cause-effect relationships in relative algal abundance in known N and P regimes in artificial streams contributes to our understanding of algal succession when N and P co-vary in nature.

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MOLECULAR PHYLOGENY OF *CHLAMYDOMONAS REINHARDTII* INFERRED FROM 3'UTR SEQUENCE COMPARISONS OF GAMETO-LYSIN GENE

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In *Chlamydomonas reinhardtii*, wild-type strains of three principal stock lines have been widely used for genetic, biochemical and morphological studies (Harris 1989). All have been identified as descendants of a mating pair (plus and minus) of clones derived from a zygospore isolated by G. M. Smith in 1945. Recently we noticed that there are several sequence differences between gametolysin cDNAs of 11-32b plus and G-9 minus strains. A clear RFLP was found between the two mating-types when the 3'UTR was used as a probe to hybridize with genomic DNAs restricted with *Sal*I. The 3'UTR differed by the deletion of 12 nucleotides in the minus DNA. We therefore obtained representative strains of all three descendant lines of Smith's isolate and examined the RFLP and 3'UTR. All the plus strains of the Sager line (21gr, G-8) and the Cambridge line (UTEX90, 11-32b, C-239) had the long (L)-type 3'UTR, while the minus strains of the two lines (6145c, C-9, UTEX89, 11-32a, G-238) had the short (S)-type one. As for the Ebersold/Levine line stocks, both plus (137C, CC-125, CC-620) and minus (137C, CC-124, CC-621) strains had only the L-type 3'UTR. We also examined 9 wild-type strains of *C. reinhardtii* isolated from the wild (all obtained from CGC) and one strain of *C. smithii* (CC-1373 plus). Surprisingly, the 3'UTR of *C. smithii* was 100% identical with the S-type one of laboratory strains. A phylogenic tree indicates that the A-type laboratory strains are included in a different clade from that of the B-type/*C. smithii*.

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MOLECULAR PHYLOGENY OF THE TRIBE CHAREAE (CHARACEAE) BASED ON RBCL SEQUENCES

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Sequences for the large subunit of the gene encoding RUBISCO (rbcL) were obtained for approximately 60 individuals in all six extant genera of the Characeae in two tribes: Tribe Chareae--*Chara*, *Lamprothamnium*, *Lychnothamnus*, *Nitellopsis*; Tribe Nitelleae--*Nitella*, *Tolypella*. Parsimony analysis (1,000 bootstrap replicates) tested monophyly of species isolates and genera of Chara, and tribes in the family. Species, genera, and tribes were strongly supported as monophyletic, except for paraphyly of a monoecious/dioecious species pair (*C. globularis* and *C. connivens*). Branch lengths within the Nitelleae were longer than in the Chareae, and bootstrap support for *Nitella* monophyly was less than for the other genera. The rbcL data support a monophyletic Chareae and Nitelleae. Within the genus, Chara sections sensu Wood are not monophyletic and in some cases are clearly paraphyletic (e.g., Grovesia). In contrast, subsections are generally well supported monophyletic groups (e.g., subsections Willdenowia and Wallmania). The data support the contention of Proctor that Wood's classification, derived from Braun's of the 19th century, is artificial and that the character defining the two sections (stipulode arrangement) is homoplasious. Mode of cortication (1-, 2-, or 3-corticate), while exhibiting some homoplasy, appears to be a synapomorphic character state for some sub-sections. Utility of characters across taxa in the family varies and is not an all-or-none categorization.

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THE IMPORTANCE OF INTEGRATING ALGAL SYSTEMS INTO A BALANCE MANAGEMENT APPROACH TO AQUACULTURE

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The National Oceanic and Atmospheric Administration (NOAA), through its National Marine Fisheries Service (NMFS) and National Sea Grant College Program (NSGCP) has developed a vision for how seafood will be produced in the USA which includes the proper management of natural fisheries, aquaculture and the management and involvement by human coastal communities. The concept of balanced ecosystem management is not only being talked about at national levels but active research programs are being planned and supported. The recent $5 million National Marine Aquaculture Initiative (NMAI) specifically called for proposals that study the "trophic level consequences of marine aquaculture and marine species enhancement". Recent workshops at the World Aquaculture Society meeting in Orlando and at the regional aquaculture meeting held at Boston on the topic of "Aquaculture and the Environment" have focused on a balanced approach to both aquaculture and fisheries management. All of the workshops focused on the important role of plants in the aquatic community. The basic premise about a balanced ecosystem approach is to incorporate the biological functions of a diverse group of plants and animals into a unified system that maintains the natural interactions of species and allows an ecosystem to function. Models are useful in understanding the energy and nutrient flow within an ecosystem; as are GIS technologies that allow us to map biological and ecological regimes. Macroalgae and phytoplankton both convert nutrients to plant material and transform carbon dioxide to oxygen. In contrast, animals derive much of their nutrition from plants, in one way or another and transform oxygen to carbon dioxide. This presentation will discuss the need to incorporate the use of plants in ecosystem maintenance such that there is balance between the animal, including humans, and plant communities in coastal areas. This will all be related to new NOAA programs and funding opportunities for research support in this area.

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THE GENOME OF THE FILAMENTOUS NITROGEN-FIXING, SYMBIOTIC CYANOBACT-ERIUM *NOSTOC PUNCTIFORME*

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Nostoc punctiforme is a terrestrial oxygenic photoautotrophic cyanobacterium with a complex life style as exemplified by multiple developmental alternatives of its vegetative cells, its ability to establish a symbiosis with plants, its capacity for facultative heterotrophic growth, and it is amenable to genetic analysis. These multiple phenotypic characteristics are indicative of the breadth of genetic information that is likely to be present in its genome. The shot gun sequencing phase of the more than 9.76 Mb genome of *N. punctiforme* ATCC 29133 was completed in July, 2000 by the Joint Genome Institute with support of the DOE (http://www.jgi.doe.gov/). Preliminary annotation of a 9.13 Mb database identified 7,432 putative protein encoding ORFs. Seventy-one percent (5,324) of the putative genes have sequence similarity to previously recognized genes in the current database of all organisms, while twenty-nine percent (2,164) are unique to *N. punctiforme*. Forty-five percent (3,328) of the genes putatively encode proteins with known or probable known function. This latter value is two-three times the number of known genes in the unicellular cyanobacteria *Synechocystis* PCC6803 and *Synechococcus* WH8102. Some of the excess coding capacity in *N. punctiforme* is seen in multiple members of gene families encoding sensory transduction systems, putative transcriptional regulatory proteins, transporters, transposases and very large multidomain proteins with peptide synthetase and polyketide synthase domains. The genetic potential detectable in the currently unfinished genome of *N. punctiforme* encompasses nearly all characteristics that define cyanobacteria as a phylogenetic group and the data will be presented in that context.

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RELATING PHYTOPLANKTON DYNAMICS AND PRODUCTION TO SEDIMENT RESUSPENSION IN SOUTHERN LAKE MICHIGAN

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Sediment resuspension is an annually recurrent feature during spring holomixis in southern Lake Michigan. Relationships between resuspension events and phytoplankton biomass, compositional dynamics, and production were evaluated during 1998 and 1999. Increased water-column light attenuation (K_{PAR}) and suspended particulate matter (SPM) concentrations corresponded with resuspension events within nearshore regions. However, neither K_{PAR} nor SPM corresponded with chlorophyll (Chl) *a* concentrations, indicating no impact of resuspension on instantaneous biomass accumulation. Diatoms and cryptophytes dominated phytoplankton assemblages and together typically comprised greater than 85% of the Chl a. The associations of SPM/K_{PAR} with diatom Chl *a*, and the inverse relationship between relative diatom and cryptophyte Chl *a* corresponded with the dominance of diatoms and cryptophytes in near- and offshore waters, respectively. Moreover, a spatial variation in species composition occurred during resuspension events; small, centric diatoms exhibiting meroplanktonic life histories and large, pennate diatoms considered benthic in origin were asso-ciated with sediment resuspension whereas large, net diatoms and cryptophytes typically comprising phytoplankton of the annual spring bloom and of optically-clear, offshore waters were not. The presence of viable diatom photopigments and the abundance of small centric diatoms within the surficial sediments, established this layer as the source of meroplankton. Integral production was dramatically reduced within sediment-impacted waters; however, nearshore assemblages appeared to have greater photosynthetic capacities than offshore assemblages. Although resuspension dramatically influenced near-shore phytoplankton assemblages, it appeared to have little, if any relationship with the compositional de-velopment of the annual spring bloom.

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USING ABSORBANCE AND FLUORESCENCE SPECTRA TO DISCRIMINATE MICROALGAL PHYLOGENTIC GROUPS AND TAXA

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Developing optical detection techniques for discriminating phytoplankton species in mixed assemblages has long been a goal of aquatic scientists. Previously, the authors described methods to discriminate the optical absorbance spectra for a red-tide dinoflagellate within both hypothetical mixed cultures and mixed natural assemblages. To this end, the utility of absorption and fluorescence-emission spectra for further discriminating among algal phylogenetic groups (and potentially taxa) and for discriminating phycobilin and non-phycobilin containing algae, respectively, was examined in laboratory cultures. A similarity index algorithm, in conjunction with fourth-derivative transformation of absorbance spectra, provided discrimination among/between the chlorophyll [Chl] *a*/phycobilins, Chl *a*/Chl *b*, Chl *a*/Chl *c*/fucoxanthin, Chl *a*/Chl *c*/peridinin spectral classes as well as closely-related phylogenetic groups within a class. Among the cyanobacteria, diatoms, and chlorophytes tested, absorbance spectra of taxa possessing dissimilar cell morphologies were discriminated with the greatest range of differentiation occurring among cyanobacteria. Interestingly, spectra for problematic cyanobacteria (including taste/odor metabolite- and toxin-producing species) were discriminated from spectra from each other and from other cyanobacteria. Fluorescence-emission spectra were distinct among algal spectral groups; similarity comparisons of fourth-derivative plots discriminated the increasing contribution of distinct taxa containing different phycobilin pigments and between phycobilin and non-phycobilin containing taxa within hypothetical mixed assemblages. The potential application for *in situ* instrumentation incorporating such approaches in monitoring programs, particularly those targeting harmful algal blooms, is elucidated.

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A NEW *LYNGBYA* SPECIES FROM AQUARENA SPRINGS, SAN MARCOS, TEXAS

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A large *Lyngbya* species was isolated from Aquarena Springs, a series of freshwater, nonmineral springs flowing from the Edwards Aquifer that are impounded to form Spring Lake. The species grows in rather large, loosely formed clumps at a depth of 2-3 meters, and consists of sheathed filaments from 55 to 60 um in thickness containing a single trichome that ranges from 45-50 um in thickness in each sheath. The sheath is 5-6um thick. Our isolate is similar to *Plectonema wollei* Farlow ex Gomont, a large, thick sheathed morphospecies which has recently been placed in *Lyngbya wollei* (Farlow ex Gomont) Onodera et al. It is also similar to *Blennothrix* species, another large-trichomed genus with a distinctive form of false branching. However, the *Lyngbya* from Aquarena Springs shows no evidence of false branching of any type, forms loose clusters rather than mats, and is larger than any of the species currently described in any of the genera in question. When we sequenced the 16SrRNA and associated 16S-23S internal transcribed spacer (ITS), phylogenetic analysis indicated the taxon was most similar to *Oscillatoria sanctae*. However, there are so few taxa in the Oscillatoriaceae *sensu stricto* that have been sequenced, phylogenetic placement of the taxon by molecular analysis alone is not possible at present. Two different ITS configurations were obtained, one with both the tRNA”$^{\text{Ile”}}$ gene and the tRNA”$^{\text{Ala”}}$ gene, the other with no tRNA genes. Our work represents the first molecular analysis of any form in the large-trichomed *Lyngbya* clade.

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BACTERIAL EFFECTS AND ALGAL BIOREMEDIATION BY *CHLORELLA ELLIPSOIDEA* GERNECK OF THE BERKELEY PIT LAKE SYSTEM

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The Berkeley Pit Lake System is one of the largest contaminated sites in North America and is located near the headwaters of the largest superfund site in the U.S. It is filling at a rate of about 28.7 million liters per day with metal laden, acidic (pH 2.7) water. *Chlorella ellipsoidea* is one of the first autochthonous species of algae from the Berkeley Pit Lake System to be tested for its bioremediative potential. An experimental matrix was designed for this experiment using a completely randomized design (CRD). The matrix was set up with tissue culture flasks having the following treatments: Na2HPO4 at 0, 25, 50, 75, 100 mg/l, inoculated vs. no-inoculated with algae (187,500 algae/ml final), and filtered (0.2Fm) vs. non filtered pit water as variables, $NaNO_3$ amount was fixed at53 mg/ml. Three replicates were made of each and the experiment lasted 60 days. The results of this experiment demonstrated significant remediation of most metals. These results will be presented. In addition, filtering seemed to negatively impact the bioremediative potential of the cultures. As a result, axenic cultures of *Chlorella ellipsoidea* will be tested in Berkeley pit water to determine if it is bacteria or filtration that caused these results.

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A NEW PARADIGM FOR FRESHWATER FRAGILARIOID DIATOM CLASSIFICATION? A CRITIQUE OF LANGE-BERTALOT’S NEW SYSTEM

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In a recent study of freshwater diatoms from South America (Rumrich et al. 2000), Lange-Bertalot introduced a new paradigm for the classification of fragilarioid diatoms. This new system is antagonistic to that presented by Williams and Round (1987) because Lange-Bertalot recognizes a marked variability in the characters chosen and a supposed overall continuity of morphological features among the genera created by his English counterparts. Lange-Bertalot then proposes a partitioning of *Fragilaria* into two genera: *Fragilaria* and *Staurosira* mainly based on the presence/absence of rimoportulae and areolate girdle bands. The newly defined Fragilaria includes relatively large phytoplankters such as *F. capucina* and *F. crotonensis*. In turn, *Staurosira* includes, for the most part, small periphytic organisms, and contains several new species that were based on varieties of old *Fragilaria* taxa. This fragmentation of species and their varieties is based on a supposed morphological discontinuity. As a consequence an apparent increase in species diversity has occurred within the fragilarioid group.The present work analyzes Lange-Bertalot's new paradigm and confronts it with recent LM and SEM evidence. The incorporation of concepts such as plasticity, polymorphism, and parallel evolution in current classification systems is also discussed. It is concluded that Lange-Bertalot’s system represents a step backward from that of Williams and Round. Some adjustments in the latter scheme could be sufficient to accommodate the diversity of fragilarioids known at present.

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PROTEIN SEQUENCE ANALYSIS OF THE HISTONE-LIKE PROTEIN HCC FROM THE HETEROTROPHIC DINOFLAGELLATE *CRYPTHECODINIUM COHNII* (PYRROPHYTA)

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The major histone-like protein HCc was extracted from chromatin of the dinoflagellate *Crypthecodinium cohnii*, purified by carboxymethylcellulose (CMC) chromatography and high performance liquid chromatography (HPLC), for protein sequencing. Four fractions were identified by HPLC fractionation of the CMC 400 mM NaCl peak, which proved to be very similar in amino acid composition, differing by only several amino acids. These differences are of the same level as the differences in histone variants of typical eukaryotes. The fractions were analyzed by peptide mapping using V8 protease, which also showed very close similarity between the four proteins . Protein sequence information was obtained by sequencing overlapping peptides, to yield approximately 80% of the protein sequence for two of the variants. Sequence comparisons with HCc1 and HCc2 from C,cohnii as reported by Sala-Rovira et al. (Chromosoma 100, 510) suggest that these variants are similar, but not identical to HCc1 and HCc2.

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A MOLECULAR PHYLOGENY OF THE BANGIOPHYCIDAE (RHODOPHYTA) AND THE ORIGIN OF SECONDARY PLASTIDS

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We sequenced the nuclear small subunit ribosomal DNA coding region from 20 members of the Bangiophycidae and from two members of the Florideophycidae to gain insights into red algal evolution. A combined alignment of plastid small subunit rDNA, rbcL, and psbA sequences were also studied to complement the understanding of bangiophyte phylogeny and to address red algal secondary symbiosis. Our results are consistent with a monophyletic origin of the Florideophycidae, which form a sister-group to the Bangiales. Bangiales monophyly is strongly supported, although *Porphyra* is polyphyletic within Bangia. Bangiophycidae orders such as the Porphyridiales are distributed over three independent red algal lineages. The Compsopogonales sensu stricto, consisting of two freshwater families, Compsopogonaceae and Boldiaceae, forms a well-supported monophyletic grouping. The single taxon within the Rhodochaetales, *Rhodochaete parvula*, is positioned within a cluster containing members of the *Erythropeltidales*. Analyses of plastid sequences show that the plastids of the heterokonts are most closely related to members of the Cyanidiales and are not directly related to cryptophyte and haptophyte plastids. Different data partitions support conflicting topologies regarding the positions of the cryptophytes and haptophytes

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LONG-TERM CHANGES IN ROCKY INTERTIDAL SEAWEED POPULATIONS IN URBAN SOUTHERN CALIFORNIA

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Long-term changes in rocky intertidal macrophyte populations were studied at nine southern California sites in Orange and Los Angeles Counties. These sites were first studied by E. Y. Dawson in the late 1950s and were subsequently resurveyed in the late 1960s and1970s, and many again in the early 1980s. The photographs and field notes of previous investigators were obtained and used to relocate each study site. Using either line transect or plot-based sampling methods, frequency and cover data were obtained for all macrophyte populations. Collected data were then compared with records from previous studies to determine changes in species composition and abundances. During the past fifty years, the human population increased greatly throughout southern California, resulting in extensive modifications of the coastal landscape and increased inputs of non-point and point-source discharges, particularly in rapidly growing Orange County. In addition, shifts in oceanographic climate have taken place over the five decadal period of study. Our results reveal that significant changes in macrophyte abundances have occurred throughout the region during this period. The abundances of disturbance-resistant seaweeds have increased and of large, fleshy species have decreased. Macrophyte diversity has declined and most sites are now dominated by low-producing crustose and articulated coralline algae, and small, turf-forming algae. Increases in these forms, together with observed decreases in larger, and generally more-productive fleshy seaweeds have resulted in a significant decline in the availability of frondose canopy habitat, changes in primary productivity, and possible shifts in food web structures in southern California rocky intertidal communities.

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THE CYANOBACTERIAL FLORA OF MICROBIOTIC CRUSTS OF NATURAL BRIDGES NATIONAL MONUMENT, UTAH.

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We examined the cyanobacterial flora of microbiotic crusts in two sites of Natural Bridges national Monument. Composite soil samples were subsampled, dilution plated on Z8 media, and incubated 3-5 weeks. Cyanobacteria were characterized directly from plates, and isolations were made of all morphologically distinct strains. The cyanobacterial flora of soils of this site differed markedly from cyanobacterial floras observed elsewhere in the soils of the Colorado Plateau. Oscillatoriales were well represented, with *Leptolyngbya* (4 spp.), *Microcoleus* (3 spp.), *Schizothrix calcicola*, *Phormidium rubroterricola*, and *Oscillatoria coerulescens* all present. Coccoid cyanobacteria were notably absent, and heterocystous species were limited in number (only *Scytonema javanicum* and *Calothrix elenkinii*) and very rare. *Nostoc* species were unusually rare, and seen only in moistened soil samples. *Tolypothrix* was also seen in the moistened soils. Two especially interesting strains are likely new taxa. A *Schizothrix* morphospecies was recovered with distinctively constricted cells and a yellow-brown calyptra that fits no description of *Schizothrix* or *Leptolyngbya*. An especially large form of *Microcoleus vaginatus* was found with a trichome width of 5-9.6 ìm. Sequence of the 16S rRNA provides some evidence that both of these morphospecies may be distinct from other members in their genera.

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USE OF AN EDIBLE RED SEAWEED TO IMPROVE EFFLUENT FROM SHRIMP FARMS

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An integrated shrimp and seaweed farm in Hawaii demonstrates the advantages of using algae to improve effluent quality from animal aquaculture operations. *Gracilaria parvispora* grown in the effluent channel of a commercial shrimp farm *(Penaeus vannamei)* absorbed significant quantities of nitrogenous and other waste compounds. Wild populations of the edible red seaweed were transferred into the effluent channel and grew at a Relative Growth Rate of 4.7% per day and increased it's nitrogen content from 1% to 3%. The weekly harvest of 100 kg from the effluent canal represented a removal of 3 kg of nitrogen from the effluent stream. An additional benefit was that seaweed could be fertilized in the ditch. Stocks of seaweed placed in the effluent ditch for five days would increase their nitrogen content from 1% to 3% and C:N ratio changed from 30:1 to 10:1. These algae would then be placed in cages in the ocean and would achieve growth rates of 9% per day compared to inorganically fertilized algae with RGR of 4.6% and unfertilized algae with RGR of 1.7%. Inorganic fertilizers that were formulated to mimic the concentrations in the effluent canal could not support the growth rate of the organic fertilization, implying that some micro-nutrients or other factors were benefiting the algae growth.

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SPECIES COMPOSITION AND CONTROLS OF ULVOID ALGAL BLOOMS IN WASHINGTON STATE

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Blooms of green macroalgae can devastate important finfish and shellfish habitats. In earlier work, blooms in the San Juan Islands, Washington State were shown to be composed primarily of *Ulva fenestrata* in the intertidal and *Ulvaria obscura* subtidally. *Ulva* is more desiccation-tolerant than *Ulvaria*, allowing it to dominate the intertidal. *Ulvaria* does not have a growth or photo-synthetic advantage when grown subtidally. This study examined ulvoid algal blooms throughout Washington State to see if the intertidal versus subtidal distribution of these two species was consistent over a larger geographical scale. We also tested grazer preferences using a suite of herbivores to see if top-down effects might allow *Ulvaria* to dominate the subtidal. All tested sites in Puget Sound had the highest relative *Ulvaria* biomass in the subtidal and the highest relative *Ulva* biomass in the intertidal. *Ulvaria* was completely absent from sites in Hood Canal, Grays Harbor, and Willapa Bay. At several sites *Enteromorpha* spp. contributed significantly to the ulvoid bloom. Grazing preferences were examined with two species of gastropod (*Lacuna variegata* and *Lacuna vincta*), an urchin (*Strongylocentrotus droebachiensis*), and an isopod (*Idotea woesnesenskii*). All tested grazers are more active in the subtidal and preferred *Ulva* to *Ulvaria*. In conclusion, *Ulvaria* dominates the intertidal and *Ulva* the subtidal whenever the two co-occur. While not the only plausible explanation, it appears that subtidal grazers may allow *Ulvaria* to dominate the subtidal.

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CYCLOPHEOPHORBIDE *A* IN SUSPENDED PARTICLES IN THE META- AND HYPOLIMNION DURING OXYGEN DEPLETION IN A SUBTROPICAL LAKE

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Cyclopheophorbide a (CPP) was the most abundant degradation product of chlorophyll a (Chl a) in the hypolimimnion, benthic boundary layer and sedimenting particles in Lake Kinneret in June-July 1997, when a prolonged period of residual oxygen concentration was found below the thermocline. The highest concentration of CPP was found in the benthic boundary layer, where it reached as high a concentration as 2 *g liter-1. In sediment traps positioned below the thermocline CPP constituted from about 5 to 20% of the Chl a concentration and in bottom sediments from 5 to 185%. CPP was not detected in epilimnetic water samples, but was found in sediment traps located within the oxygenated water column, within the range of surface wave action. Comparison of regular traps, with traps poisoned by formaldehyde indicates that the importance of CPP-containing particles in the sedimentation flux is relatively low, and that the bulk of CPP found in the bottom sediment is apparently a result of Chl a transformation in situ by biological activity. The timing and location of CPP appearance coincided with the lake compartments where denitrification apparently took place. This suggests that CPP can be used as a target pigment to trace organic matter processing under transient conditions in the water column and on the bottom sediments.

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EVALUATING CARBON METABOLISM OF *PORPHYRA* BASED ON IRRADIANCE, TEMPERATURE AND NUTRIENT CONCENTRATIONS

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Fish aquaculture produces a nutrient-rich effluent. One means to remediate the discharge of these effluents is to couple seaweed culture with that of fine-fish. Seaweeds assimilate inorganic nutrients and some may produce tissue that can be sold. *Porphyra* culture is a multi-billion dollar global industry producing edible biomass and high-value biochemicals. To identify fast growers, we have examined the carbon metabolism of *Porphyra purpurea, P.umbilicalis* and *P.leucosticta* by measuring photosynthetic production at light levels ranging from 17 µmol photon m^{-2} s^{-1} (sub-saturating) up to 315 µmol photon m^{-2} s^{-1} (saturating). These experiments identified species that are efficient at low (higher á) and high irradiances (P_{max}). The three species were also evaluated at growth temperatures 5°C, 15°C and 20° C to determine optimal growth temperatures along with varying nutrient concentrations. Results of these experiments will help choose *Porphyra* species for maximum growth and biomass under varying light, nutrient concentration and temperature conditions.

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SPATIAL PARADIGMS OF LOTIC DIATOM DISTRIBUTION: A LANDSCAPE ECOLOGY PERSPECTIVE

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On August 27, 1999, diatoms were sampled and current velocity and depth were measured on a regular square sampling grid at 81 locations in an unshaded, cobble-bottom reach of White Creek, NY. Diatom relative abundance was subjected to detrended correspondence analysis. The first axis (DCA1) explained 51% of the variance and separated the samples according to current regimes. The spatial autocorrelation of DCA1 sample scores was determined by Moran's I statistic, which showed that communities' patch length was > 3.1 m while their patch width was 0.5 to 1 m. *Achnanthes minutissima* and *Fragilaria capucina,* the two species with the highest relative abundance, displayed spatially structured patches of low abundance and comparatively random patches of high abundance, suggesting broad scale abiotic control of species performance in low abundance regions and finer scale biotic control of high abundance areas. The spatial complexity in low vs. high velocity transects was determined by calculating the respective fractal dimension (D). D of DCA1 was higher in the higher current velocity transects, demonstrating that spatial complexity of diatom communities increased in faster currents, probably due to impeded immigration. Partial canonical correspondence analysis on diatom, environmental and spatial data revealed that 38% of the variance of species data was explained exclusively by current velocity, while space alone contributed only 10%. This result indicates that 1) current velocity was the major factor controlling diatom distribution and 2) there were other spatially dependent variables, most likely biotic, but their role in shaping diatom communities was minor.

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CURRENT VARIABILITY SHAPES MORPHOLOGICAL COMPLEXITY IN COLONIAL STREAM DIATOMS

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On August 27, 1999 diatoms were sampled, and current velocity was measured at 81 locations on a regular square sampling grid in an unshaded, cobble-bottom reach of White Creek, NY. The grid had an extent of 16 m^2, interval, the distance between neighboring sampling points, of 0.5 m, and grain size, the size of the elementary sampling unit, of 0.01 m^2. Six of the seven dominant benthic diatoms were colonial forms, including *Diatoma vulgaris, Fragilaria capucina, F. crotonensis, Gomphoneis minuta, Melosira varians,* and *Synedra ulna*. Their morphology and distribution were investigated from the perspective of fractal geometry and stream ecology, respectively. Fractal dimension of diatom colonies, indicative of their shape complexity, ranged from 1.06 to 1.54, demonstrating vast morphological variation from simple geometric shapes to complex outlines. The relative abundance of the six diatoms was regressed against current regime, which ranged from 0.03 to 0.66 mÌs^{-1}. All regression models were significant at $P < 0.05$ and explained between 55% and 94% of the variation in diatom distribution. The diatom niche breadth, i.e. the amount of environmental variability a species can tolerate, was defined from these models and showed substantial variation, from 5 to 14. The regression model of fractal dimension against niche breadth was significant and explained 76% of colonial shape variation, revealing a strong relationship between diatom colonial complexity and habitat variability. This finding suggests that environmental variability could create highly complex colonial morphologies in benthic diatoms as an evolutionary strategy for survival in unpredictable environments.

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SEAWEED CHEMICAL DEFENSES: LINKING DEMOGRAPHY TO INTRAPLANT VARIATION IN DEFENSES

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Optimal defense theory (ODT) predicts that plants should allocate more resources to production of defense chemicals in parts that have the highest value in terms of fitness. In previous studies, assumptions about differences in fitness value among plant parts have been based on general reasoning, e.g. that reproductive tissue is more valuable then vegetative tissue since fitness ultimately depends on reproduction. Here we present an example of how demographic elasticity analysis can be used to generate species-specific estimates of the fitness value among different plant parts. These estimates were used to make predictions about intraplant variation in chemical defenses of the intertidal seaweed *Ascophyllum nodosum*, within the framework of the ODT. We show that the pattern of variation in defence levels, as well as susceptibility to herbivory, among different tissue types of Ascophyllum is in accordance with these predictions. A stronger focus on variation in life history among species could lead to a better understanding of intraplant variation in defense levels, in seaweeds as well as vascular plants.

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TAXONOMIC RESOLUTION OF *LEPTOLYNGBYA* (CYANOPHYTA) UTILIZING THE 16S rRNA GENE SEQUENCE

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Partial 16SrRNA sequence data were obtained from 17 strains of desert soil *Leptolyngbya* from three sites in the western United States to determine phylogenetic relationships. The resulting sequence data was aligned and analyzed with currently published sequence data from other cyanobacteria within the genus *Microcoleus* (outgroup) and other *Leptolyngbya*. Phylogenetic analysis using the optimality criteria of parsimony, distance, and maximum likelihood all gave trees with nearly identical topology that were well supported by their respective boot-strap analyses. A large well-supported clade contained 11 of our *Leptolyngbya* strains and the two *Leptolyngbya* sequences on GenBank. One unusual, calyptrate *Leptolyngbya* was molecularly similar to *Microcoleus*. One strain, with the classic sheath morphology of *Schizothrix*, was distinct in its 16S rRNA sequence from both *Leptolyngbya* and *Microcoleus*. Two other strains were distinct from all strains in our tree. We suspect that *Leptolyngbya* as currently delineated contains multiple genera and many cryptic genospecies with highly similar morphology. Additional phenotypic characterization using other character sets is warranted. There was some evidence for limited operon-specific variability in the 16S rRNA gene in our strains.

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EFFECTS OF NITRATE AND PHOSPHATE DISCHARGE FROM A FERTILIZER PRODUCTION PLANT IN A FJORD

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“Hydro Agri Glomfjord” a fertilizer producer has been discharging large quantities of ammonium, nitrate and phosphate into Glomfjord (in northern Norway, N 66,48°, E 13,57°) since 1947. The effects of the nutrient load to the Fjord have resulted in classical eutrophication symptoms. Elevated nutrient levels have resulted in frequent plankton blooms and seasonally reduced O_2 levels in the deep-water bodies as well as an eradication of the normal littoral assemblages. The fucoid belt had been replaced by various green algae 6 km from the outlet and outward the fjord. The effect is only seen on the northern side of the Fjord. After some improvement in the discharge loads in the 1980’s, the condition in the pelagic column improved with regard to plankton blooms and deep water O_2 concentration. The littoral communities didn’t, show any indication of recovery and a pollution indication index based on algal composition, showed even increased eutrophication.

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STUDIES ON THE LIFE CYCLE OF *PORPHYRA DIOICA* AND *PORPHYRA UMBILICALIS* FROM PORTUGAL

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Porphyra is a well known genus, as it is one of the most important maricultured seaweeds. It has an annual retail

value of US $1.8 billion principally grown in Japan, Korea and China. In the past 10 years, expanding markets in North America and Europe have put new demands on the import of the red alga. After some attempts trying to grow Asian species, efforts are now centered on domestication of native species, both in North America and Europe. With our current understanding of this genus, we believe that their are at least six *Porphyra* species along the Portuguese coastline. We have initially identified two species, *P. dioica* and *P. umbilicalis*, as potentially promising for aquaculture development. We are studying the life cycles and phenology of these species, with special note on morphological variability and on the environmental control of each phase of the life cycle. *Porphyra dioica* carpospores germinate best at 15-20ºC and 25-75 micro mol photons m^{-2} s^{-1}. Best conditions for conchocelis vegetative growth and the development of conchosporangia are at 15º C and 25-75 micro mol photons m^{-2} s^{-1}. *P. dioica* conchocelis growth appears to be under temperature controlled, rather than by photoperiod. For *P.umbilicalis*, 15º C and 25 micro mol photons m^{-2} s^{-1} are the best conditions to obtain conchocelis and for formation of conchosporangia. At these conditions carpospore survival rate is about 33.4 %. Conchospores are released at 10 and 15º C and at 25-75 micro mol m^{-2} s^{-1}. For both species, formation of conchosporangia seems to be more related with time and culture conditions, rather than with size or development of the conchocelis.

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GROWTH OF CULTURED ABALONE, *HALIOTIS FULGENS*, USING NATURAL ALGAL DIETS

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Algal diets have been evaluated and used extensively for the aquaculture of abalone, especially in Japan. In other areas little is known about the nutritional value of the algae that the local abalone consume. In Mexico, regional hatcheries use *Macrocystis pyrifera* as a main source of natural food. Kelp availability, year round, has not generally been a problem for abalone aquaculturists. Most difficulties occur during severe storms, which may prevent access to kelp beds. El Niño also caused widespread destruction of *M. pyrifera*. In this study, growth rates of juvenile green abalone *Haliotis fulgens*, 31.7 " 1.5 mm shell length and 2.5 " 0.2 g body weight were evaluated during 136 days. Juveniles were fed with some of the algae used in regional hatcheries, *Macrocystis pyrifera*, *Egregia menziesii*, *Eisenia arborea*, *Porphyra* sp. and *Ulva* sp. Shell length growth rates varied between 2 mm day-1 for Ulva sp. and 18 mm day-1 for *M. pyrifera*. Body weight rates ranged from no growth for Ulva sp. to 14 mg day-1for E. menziesii. The percent of survival was between 46 and 75 %. Ulva sp. showed the lowest protein content followed by *E. menziesii*, *E. arborea*, *M. pyrifera* and *Porphyra* sp.

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NITROGEN AND PHOSPHORUS REMOVAL RATES WITHIN SUBUNITS OF ALGAL TURF SCRUBBERS GROWN ON DAIRY MANURE

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Conservation and reuse of nitrogen (N) and phosphorus (P) from animal manure is increasingly important as producers try to minimize transport of these nutrients off-farm. An alternative to land spreading is to grow crops of algae on the N and P present in the manure. The general goals of our research are to assess one algal production technology, termed algal turf scrubbers (ATS) to recover nutrients from animal manures. The specific objectives of these experiments were to test different loading rates of anaerobically digested dairy manure on nitrogen removal rates. Algal turfs were grown in a laboratory-scale ATS unit (1 m^{2}) operated by recycling wastewater and adding manure effluents daily. The most abundant genera of benthic algae in the ATS unit were *Ulothrix*, *Oedegonium* and *Rhizoclonium*. Replicate subsamples (0.04 m^{2}) of algal turfs of the same age were removed from the ATS unit and treated with different loads of manure containing 5–40 mg l^{-1} ammonium-N (NH_4-N). During the experiments, the pH was maintained between 7–7.5 to prevent ammonia volatilization. Ammonium-N removal rates were biphasic, with a fast rate of 3.0–4.7 mg-NH_4-N hr^{-1}g^{-1} DW for the first 20–30 minutes, followed by a slower rate of 0.53–0.96 mg-NH_4-N hr^{-1}g^{-1} DW for the remainder of the 2 hour incubation period. The initial rates are comparable to laboratory scale ATS units and correspond to calculated removal rates of about 3 g NH_4-N m^{-2}d^{-1}.

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STORAGE OF NITROGEN IN THE FORM OF PROTEIN BODIES IN THE KELP *LAMINARIA SOLIDUNGULA*

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Proteinaceous cytoplasmic inclusions with characteristic anatomical localization patterns are common in marine algae, but the function of these cell structures has not been demonstrated. Microscopic examination of the endemic Arctic kelp, *Laminaria solidungula* J. Agardh, grown under nitrogen-limited and nitrogen-replete culture conditions revealed a variety of unusual cell structures. Some were present in both experimental treatments, but others occurred intact only under N-replete conditions. Cortex cells of N-replete specimens contained abundant single membrane-bounded cytoplasmic inclusions, 1-10 µm in diameter. Cytochemical staining procedures showed that these inclusions were proteinaceous but they did not reveal a carbohydrate component. Intact proteinaceous inclusions were absent from cells of the N-starved samples; however, structures presumed to be depleted protein bodies had an anatomical distribution comparable to that of the protein bodies in the N-replete algae. The proteinaceous inclusions in *L. solidungula* may represent a nitrogen store, which like the stores of nitrate and amino acids present in many algae, can be utilized during the summer months when water column nitrate concentrations are low but when photosynthetic carbon fixation must be maximized. Storage of nitrogen in the form of protein bodies has the advantage of being osmotically inactive.

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PHYSCOMITRELLA, A NEW MODEL SYSTEM FOR GENOMIC STUDIES

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Model systems have played essential roles in elucidating complex processes in a large number of different organisms. With the emergence of genomics as a tool, the use of model systems can now be extended to study interacting networks of genes and their products that make up cells, organisms and populations (see: Quatrano, R. S., ed. 2001). Plant Genomics: Emerging Tools (American Society of Plant Biologists). Using molecular and genetic approaches in the moss *Physcomitrella* patens, we are taking advantage of an established genetic system in which transformation and homologous recombination can be reproducibly utilized to address the question of how apolar cell is established and expressed. In collaboration with Dr. D. Cove,an EST project and BAC library construction are now in progress to identify genomic sequences in moss that are similar to those in seed plants (http://www.moss.leeds.ac.uk). Such sequences will be used for gene disruption and gene replacement studies to determine cellular phenotypes(function) of these genes. We are trying to complement mutants with interesting developmental phenotypes related to cellular polarity, as well as to generate a tagged (insertional) library for purposes of identifying lines with similar phenotypes that would allow us to isolate the genes involved. Hopefully, our approach to study polarity in the moss model system can be seen as one approach that might be applied to key algal species for genomic studies and a further understanding complex phenomena in algae.

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TOXICITY IN *PERIDINIUM ACICULIFERUM* - AN ADAPTIVE STRATEGY TO OUTCOMPETE OTHER WINTER PHYTOPLANKTON?

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Freshwater dinoflagellates may form dense blooms during winter in ice-covered lakes. Unlike their marine counterparts freshwater dinoflagellates are rarely considered as potential toxin producers. Here wetested if the winter species *Peridinium aciculiferum* produces a toxin(s)and investigated the potential adaptive function of that toxin - i.e.predator defense or inhibition of competitors (allelopathy). We found that *P. aciculiferum* produces a toxin(s). Using traditional toxicity bioassays(*Artemia* toxicity test and haemolytic activity assay), we detected the production of a toxic substance by *P. aciculiferum* cells from both the field and from laboratory cultures. Cultures deprived of phosphorus showed most toxicity. Potential predators, such as *Daphnia galeata* (Cladocera) and *Eudiaptomus graciloides* (Copepoda), were apparently not harmed by *P. aciculiferum* toxicity. However, a competitor, the flagellate *Rhodomonas lacustris* (Cryptophyceae), which co-occurs with *P. aciculiferum* was killed (allelopathy) by the *P. aciculiferum* toxin. The toxin(s) caused the cells of *R. lacustris* to swell and burst. We concluded that allelopathy may be an adaptive strategy of winter dinoflagellates, allowing them to outcompete other phytoplankton species, and thereby dominate the algal biomass.

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SYMBIODINIUM SPP. ISOLATES FROM STONY CORAL: ISOLATION, GROWTH CHARACTERISTICS AND EFFECTS OF UV IRRADIATION.

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Symbiodinium species were isolated from *Montipora capitata, Acropora palmata* and two field samples of *Porites porites*. Cultures were grown and maintained on L1 medium prepared with 0.22 µm filtered seawater (34 ppt). Cultures of *P. porites* (JR02) contained both stationary cocoid cells, cysts and motile gymnodinoid cells. Cocoid cells ranged in size from small cells (10 – 12 µm) to large cysts (23 – 25 µm). Mature large cysts contained eight gymnodinoid cells. Cocoid cells of intermediate sizes contained two or four cells which ulti-mately divided to form the eight-gymnodinoid cells found in the large cysts. In log growth cultures, new cells were derived from the release of the eight-gymnodinoid cells. Large cocoid cysts were not often observed in mature cultures. Gymnodinoid cells were motile and became attached to the container surface where they rapidly spun as do most *Symbiodinium* sp. At some point the gymnodinoid cells would lose their motility and sink to the base of the container where they would convert to cocoid cells. Cultures of the other isolates contained similar cell types. A laboratory exposure system was developed and exposures to UV light were conducted in a solar simulator over a range of UV doses. Cultured *Symbiodinium* species demonstrated higher sensitivity to UV than their analogous coral/zooxanthellae complexes.

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DEVELOPMENT OF THE CONCHOCELIS STAGE OF *PORPHYRA PERFORATA* FROM THE TEMPERATE MEXICAN PACIFIC

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The conditions governing the reproduction of the conchocelis stage of *Porphyra perforata* were studied *in vitro*. Carpospores and conchocelis development were observed under a factorial of light and temperature with four levels of irradiance 15, 50, 100 and $275 \mu Em^{-2}s^{-1}$, with 8:16,12:12,16:8 photoperiods and four temperature levels 10,15, 20 y 25 ^{0}C. *Conchocelis* stage and conchosporangial structure developed without shells. Filaments were present at 10-20^{0}C and $15\text{-}270 \mu Em^{-2}s^{-1}$ with 8:16, 12:12 ,16:8 photoperiods. The initiation of the conchosporangia was present at $50\text{-}100 \mu Em^{-2}s^{-1}$. Release and germination of conchospores occurred only when the photoperiod changed from 8: 16 to 12:12. Filaments exposed to $270 \mu Em^{-2}s^{-1}$ (16:8) persisted with discolored apices that did not develop conchosporangia. Filaments not exposed to variations of photoperiod formed monospores. Our results are notoriously different from previous studies of *P. perforata* from Washington USA (cold region) were *Conchocelis* was vegetative at 15^{0}C and $5\text{-}100 \mu Em^{-2}s^{-1}$, and conchosporangial initiation was occurred at $10\text{-}25 \mu Em^{-2}s^{-1}$ with a 8:16 photoperiod and only in shells as substrate. Differences were found between both regions in the levels of temperature, irradiance and photoperiod, which promoted the development of the reproductive structure of the *Conchocelis* stage. These results show that the conchocelis stage in plants from the temperate region develop and produce reproductive structures at higher light and temperature than plants from the cold region, which suggests ecophysiological differences of this species between both regions.

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USING FISH TO SAMPLE DIATOM COMPOSITION IN STREAMS: ARE INTESTINAL FLORAS REPRESENTATIVE OF NATURAL SUBSTRATES?

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The diatom composition of natural substrates at a single site were compared with that of the gut contents of three species of fish: stoneroller minnows, bluntnose minnows, and common shiners. Seven samples of each type were collected on a single day from a single pool in a headwater stream. Cluster analysis showed that diatom species composition clustered by fish species, with common shiners being a totally exclusive cluster, while natural substrates were scattered within the stoneroller and bluntnose clusters. Both richness and Shannon diversity were higher in fish gut samples than in the samples collected by humans, with stonerollers having the greatest diversity. When water quality indices calculated from the diatom floras recovered in these samples were compared, it was surprising to see that these indices consistently were better in fish gut samples than in samples collected by researchers. Common shiners particularly differed, a result possibly due to the fact that they apparently fed off of *Cladophora* and the highly oxygenated rock wall where the stream cascaded into the pool. In no case was the difference between stonerollers and natural substrates significantly different. Furthermore, samples from natural substrates had an internal similarity not significantly different from their similarity to samples in stoneroller and bluntnose guts. Our data thus consistently indicate that stoneroller minnows are excellent samplers that efficiently collect diverse, representative diatom samples.

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SYSTEMATICS OF *PRASIOLA* AND PHYLOGENETIC POSITIONING OF THE PRASIOLALES (CHLOROPHYTA)

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The systematics of the green algal genus *Prasiola* was investigated through a combination of morphometric and molecular analyses, and the phylogenetic position of the order Prasiolales was re-examined with molecular tools. Thirty-six herbarium sheets of *Prasiola* as well as 39 field collections and 12 type specimens were measured for several cellular and thallus characters. Means of each character were used in both cluster (UPGMA algorithm) and principal components analyses (PCA) to examine relationships among collections. Significant differences among means from the resulting groups were tested using one-way analysis of variance (ANOVA; $p<0.05$). Both cluster analysis and PCA resolved five groups of specimens, of which four groups were associated with at least one type specimen. Parsimony and distance analyses of the first half of the 18S rRNA gene support the Prasiolales as being a sister clade to the Trebouxiophyceae. Uncorrected sequence divergence values within the Prasiolales range from 0-1.7%, and within *Prasiola* range from 0-1.4%. Analyses of the *rbc*L gene were not well resolved at the ordinal level, but revealed some interesting trends within and among the genera, such as sequence identity among several morphologically distinct marine species of *Prasiola*, and large sequence divergence values between marine and freshwater *Prasiola*. Inclusion of representatives of the other members of the Prasiolales (*Rosenvingiella*, *Prasiococcus* and *Prasiolopsis*) confirmed that the Prasiolales is a monophyletic group.

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ULTRASTRUCTURE OF THE BASAL BODY COMPLEX AND PUTATIVE VESTIGIAL FEEDING APPARATUS IN *PHACUS PLEURONECTES* (EUGLENOPHYCEAE)

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Phacus pleuronectes (O. F. Müller) Dujardin is a phototrophic euglenoid with small discoid chloroplasts, a flat, rigid body, and longitudinally arranged pellicular strips. The flagellar apparatus consisted of two basal bodies and three flagellar roots typical of many phototrophic euglenoids, but also had a large striated fiber that connected the two basal bodies and associated with the ventral root. The three roots, in combination with the dorsal microtubular band, extended anteriorly and formed the major cytoskeletal elements supporting the reservoir membrane and ultimately the pellicle. A cytoplasmic pocket arose in the reservoir/canal transition region. It was supported by the ventral root and a C-shaped band of electron-opaque material that lined the cytoplasmic side of the pocket. A large striated fiber extended from this C-shaped band toward the reservoir membrane. The presence of striated fibers in the basal apparatus and associated with the microtubule reinforced pocket suggested that *P. pleuronectes* may be at the base of the *Phacus* lineage and may be more closely related to the phagotrophic euglenoids than to *Phacus* species which are ovoid in shape and have thicker pellicle strips.

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ARE CYOTPLASMIC POCKETS (MTR/POCKET) PRESENT IN ALL PHOTOSYNTHETIC EUGLENOID GENERA?

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In 1985, Willey and Wibel described the existence of a cytoplasmic pocket formed from the reservoir membrane in *Colacium*. A band of microtubules derived from the ventral flagellar root (MTR) lined the pocket and a dense fibrillar mesh was associated with the membrane. A comparison of bodonid cytostomes, colorless euglenoid cytostomes, and the reservoir pocket found in *Colacium* suggested that the three structures were homologous and that photosynthetic euglenoids arose from phagotrophic ancestors. MTR/pockets have since been reported in other photosynthetic euglenoids including *Euglena, Eutreptia, Eutreptiella, Cryptoglena*, and *Tetreutreptia*. We now report on MTR/pockets in *Lepocinclis*, *Trachelomonas*, *Strombomonas* and *Phacus* thereby demonstrating the presence of this complex in representatives of all of the major photosynthetic genera. A comparison of the MTR/complex across genera indicates a reduction in structural complexity that is consistent with recent phylogenetic schemes based on molecular characters.

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A PHYSICAL MAP OF THE *CHLAMYDOMONAS REINHARDTII* NUCLEAR GENOME

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We will report an ongoing project to construct a physical map aligned with the genetic map of the *Chlamydomonas* genome. The project builds on a previously assembled molecular map consisting of more than 250 unique molecular markers placed on the 17 linkage groups of the genetic map with an average spacing of 4-5 cM (400 - 500 kb). Those markers have been used as hybridization probes to screen a BAC library constructed from *C. reinhardtii* DNA. The positive BAC clones have been assembled into "contigs" of approximately 130 kb by comparing a "fingerprint" of restriction fragments of each clone. The gaps between the mapped contigs are filled by screening the BAC library with unique probes derived from EST sequences and by "walking" from the ends of contigs. The use of the physical map to facilitate map-based cloning of genes identified by mutation will be discussed.

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USING INCIDENT LIGHT TO MAXIMIZE RESOLUTION OF STRUCTURE ON DIATOM VALVES WITH A LIGHT MICROSCOPE

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Because of their abundance in a diverse array of aquatic habitats and chemical conditions, and the fact that their species-specific siliceous outer coverings remain for millennia in sediments, diatoms have become one of the most powerful organismal groups used in the reconstruction of historical environmental conditions. Although SEM is now needed to identify some species, the bulk of the identifications and data collection is still accomplished with a light microscope. In this paper I will demonstrate the use of interference reflection contrast (incident light) for the examination of diatoms that significantly improves the resolution of structural detail. Using incident light one can routinely distinguish between structures close to the theoretical limit of resolution for visible light, and about 70 nm closer together than resolvable with standard transmitted light optics. Examples of how the incident light technique can improve data collection with light microscopy will be given. Most research light microscopes could be easily and inexpensively outfitted to use this technique.

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TO DIVE OR NOT TO DIVE: SCUBA VERSUS ROV SAMPLING OF MACROALGAE AT 30M DEPTH

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Remotely Operated Vehicles (ROVs) and enriched air Nitrox SCUBA diving have recently become available to researchers for studying the deep-water environment. Each use a different technique for collecting macroalgal abundance data: ROVs use collections and high-resolution digital video which can be quantified using an integrative laser and computer imagery program (high tech), while divers often count the densities of individuals and use a point contact method for sampling percent (%) cover in situ (low tech). While the types of data collected by both techniques are the same, the effects of the different sampling methods on data resolution are unknown. As part of a larger study on deep-water macroalgae in central California, I compared the abundance of common macroalgae (% cover of macroalgal groups and individuals/m2) collected by divers and the ROV Ventana at a depth of 30m at 3 locations in central California. Generally, there were no significant differences between diver and ROV data in the % cover of coralline Rhodophyta, non-coralline Rhodophyta, and *Pleurophycus gardneri*/m2. The use of a laser-calibrated computer imagery program and an ROV with user-controlled lighting greatly decreased lab analysis time, and a method for sampling macroalgal layers with the ROV was developed. Thus, ROVs with high-resolution digital video and supplemental macroalgal collections can be used to quantify deep-water algae as accurately as in situ divers, but without the limited dive time, depth limits, and physical demands of the latter.

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THE OTHER SIDE OF THE COIN: POSITIVE EFFECTS OF ALGAL CHEMICALS ON ANIMALS

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Phycologists and marine ecologists have long recognized the ecological and evolutionary importance of conflict between seaweeds and herbivorous animals. However, we have generally been slow to recognize the importance of positive interactions among plants and animals. From the rest of the talks in this symposium it should be evident that algae employ noxious chemicals to reduce damage by herbivores. However, the direct and indirect positive effects of those chemicals on animal populations and communities are generally underappreciated. For example, in the same way that structurally complex organisms like corals or kelp provide a physical refuge for many species by excluding or confusing predators, the chemical defenses produced by many algae repel larger herbivores and predators, making these algae ideal safe sites for smaller invertebrates and algae that lack chemical defenses. Such “associational refuges” increase the persistence of species susceptible to predation, often resulting in a local increase in diversity, but the consequences of such interactions for the host species are often unclear. However, there are a growing number of taxonomically diverse examples (including some crustaceans, gastropods, sea urchins, and polychaete worms) in which small invertebrates use chemically defended algae as shelter yet feed on other species. In some cases these herbivores benefit the plant by aiding in its attachment to the substrate, facilitating dispersal, or mediating interactions with competitors. I will discuss these and other examples from recent work to illustrate how algal chemistry can mediate strong positive interactions with important community-level consequences.

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CHEMICAL MEDIATION OF INTERACTIONS AT SEAWEED SURFACES

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Chemical mediation of surface colonization of seaweeds by epibiota is much less well understood than analogous seaweed/herbivore interactions. Fundamental to studies of colonization is the localization and quantification of metabolites on or near seaweed surfaces. Halogenated furanones from the Australian alga *Delisea pulchra* are present at the surface of the alga in concentrations which strongly inhibit settlement of epibiota , and are examples of "natural antifoulants". However, many other chemically rich seaweeds do not appear to use natural products as antifoulants, either because the compounds are not present at the thallus surface, or because they do not persist there in high enough concentrations. We suggest that natural antifoulants in seaweeds will most commonly be non-polar metabolites, and restricted to species with morphologies that allow for their release. In contrast, primary (and often water soluble) metabolites appear to be more common as inducers of settlement. Several examples of inducers will be briefly presented, including our own research on the floridoside - isethionic acid complex from red algae which induces metamorphosis of larvae of the echinoid *Holopneustes purpurascens*. The leaky nature of such metabolites in seaweeds suggests that delivery systems for positive cues will be less important. These and other suggested generalities regarding the production of deterrents vs. inducers of colonization will be considered.

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CHELATED IRON AND GAMETOGENESIS IN *MACROCYSTIS*

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Gametophytes of *Macrocystis* can be vegetatively propagated indefinitely in the laboratory in media that lacked chelated iron. When clones of vegetatively propagated female gametophytes of *Macrocystis pyrifera* (L.) C. Ag. were exposed to varying levels of chelated iron in both enriched natural and artificial seawater, low levels of chelated iron (1-5 micromoles/liter) stimulated the onset of gametogenesis as measured by egg production. In contrast higher iron concentrations strongly inhibited egg production. Gametophyte growth as measured by the increase in cell number was not affected by iron concentration in the medium. At all iron concentrations there was a tendency for the larger gametophytic fragments to become fertile earlier. The results suggest that iron may play a critical role in the development of *Macrocystis* gametophytes.

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THE RNA POLYMERASE II C-TERMINAL DOMAIN AND THE EVOLUTION OF RED ALGAE.

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In recent years a great deal of biochemical and genetic research relating to RNA polymerase II transcription has focused on the C-terminal domain (CTD) of the pol II largest subunit (RPB1). In animals and yeast, where pol II function is most well characterized, this domain of tandemly repeated heptapeptides has been linked functionally to important steps in the initiation and processing of messenger RNA. Despite its essential role throughout the pol II transcription cycle, a tandemly repeated CTD has not been found in growing number of diverse eukaryotic organisms. Phylogenetic analyses of RPB1 sequences, excluding C-terminal regions, show that those taxa in which the pol II CTD is strongly conserved make up a unique monophyletic group. The Rhodophyta is not a member of this "CTD-clade," nor are tandem heptad repeats present in most red algae. In addition, genetic complementation studies indicate that the evolutionary differences found in rhodophyte RPB1 C-terminal sequences are incompatible with CTD-based pol II transcription in yeast. The results of these combined analyses suggest that the CTD lies at the heart of a key innovation in the evolution of RNA polymerase II that helped to confer more intricate control over gene expression. The apparent absence of these control mechanisms in red algae may help to explain their lack of true tissue differentiation.

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PHYTOPLANKTON CHEMICAL SIGNALS INFLUENCE HERBIVORY BY PROTIST GRAZERS.

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Deterrence of herbivores through chemical signaling - plant chemical defense - is known to be widespread throughout terrestrial and marine benthic ecosystems. The occurrence and significance of chemical defenses produced by unicellular marine phytoplankton has not, however, been systematically explored. We are investigating the role of the phytoplankton-produced compounds dimethylsulfoniopropionate (DMSP) and its cleavage products dimethylsulfide (DMS) and acrylate in reducing herbivory by protist grazers. DMSP is produced in high intracellular concentrations by numerous phytoplankton species, especially notorious bloom-forming dinoflagellates and prymnesiophytes, and is cleaved by the enzyme DMSP lyase during grazing, physiological stress or cell lysis. Using four different strains of *Emiliania huxleyi*, a coccolithophorid (prymnesiophyte), we have shown that strains with high DMSP lyase activities experience consistently reduced levels of herbivory in comparison with low lyase strains. Curiously, the products of the cleavage reaction (DMS and acrylate) do not affect rates of herbivory, indicating that this is not an activated chemical defense. Feeding rates are, however, proportionally and substantially reduced by the reaction precursor, DMSP. We also have preliminary evidence that high lyase strains release more DMSP to seawater than do low lyase strains. DMSP, while not evidently toxic to protist grazers, appears to act as a chemical signal that reduces grazing. We hypothesize that these unicellular herbivores may have evolved to recognize and respond to a compound (DMSP) that is a precursor of the potentially deleterious cleavage product acrylate. This and related chemical signaling processes may be important in promoting formation of phytoplankton blooms in the sea.

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IMPACT OF ENVIRONMENTAL CHANGES ON PRODUCTION OF BROWN ALGAL PHLOROTANNINS

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Ecological theories such as resource availability theory (e.g. carbon-nutrient balance hypothesis) and growth differentiation balance theory have attempted to provide a theoretical framework in which to balance putative improvements in plant fitness resulting from secondary metabolite production with the metabolic costs of these compounds. However, experimental tests of these theories have yielded mixed results, in part because they have relied upon static measures of phlorotannin concentrations and have incorrectly assumed phlorotannins to be immobile defenses with negligible rates of turnover. To test these theories more effectively, *in situ* rates of phlorotannin biosynthesis were examined in several species of brown algae as they responded to manipulations of their surrounding environmental conditions (light, nutrients, and simulated herbivory). The rates of phlorotannin synthesis measured for damaged *Fucus vesiculosus* from Maine (USA) were dramatically in-creased relative to controls. Carbon resources required for this response were supplied via wound-induced increases in photosynthetic rates, rather than changes in patterns of internal resource allocation. The lack of internal resource trade-offs indicates that phlorotannin production may not always generate allocation costs in these plants. Shading decreased the magnitude of the induced response but growth *vs.* defenses trade-offs were not identified. For *Sargassum hystrix* var. *buxifolium* from the Bahamas, rates of phlorotannin synthesis and total phenolic contents were decreased by both shading and nutrient enrichment. Overall, rates of phlorotannin synthesis were often, but not always, correlated with the eventual changes in total phlorotannin concentrations. Where synthesis and accumulations were uncoupled, differential rates of phlorotannin turnover are expected.

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LATE-QUATERNARY SETTING FOR THE EVOLUTION OF AN ENDEMIC DIATOM IN YELLOWSTONE LAKE, USA

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The diatom *Stephanodiscus yellowstonensis* Theriot and Stoermer is endemic to Yellowstone Lake, where it can be an important component of the summer phytoplankton assemblage. It is closely related to *Stephanodiscus niagarae* Ehrenberg which is abundant in nearby Heart Lake, Lewis Lake, Jackson Lake and several regional reservoirs. Here we use the stratigraphic record of Yellowstone Lake to investigate the evolution of *S. niagarae* to *S. yellowstonensis* and to describe the limnologic and climatic conditions associated with its evolution. A dramatic morphological shift takes place between about 10,000 and 9,000 years before present (ybp). Coincident with the morphological change in the *S. niagarae/S. yellowstonensis* complex are changes in the diatom species assemblage, biogenic silica concentrations, sediment lithology, and regional vegetation (as indicated by pollen analyses), which suggests environmental change as an influence on speciation. The increase in biogenic silica concentrations indicates elevated siliceous algal biomass in the lake driven by increased nutrient availability, particularly Si and N. Paleoclimatic recon-structions for southern regions of Yellowstone Park at this time suggest increased winter precipitation, as a result of wastage of the Laurentide ice sheet and the northward migration of the winter jet. Thus, the expansion of *A. formosa* and total diatom biomass in Yellowstone Lake was likely fostered by both increased N and Si loading driven by increased winter precipitation and earlier ice out caused by warming. The finely laminated sediments suggest elevated lake level and long periods of stable water-column stratification, also associated with the elevated precipitation and regional warming.

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WATER-BORNE CHEMICAL CUES AS ELICITORS OF ALGAL DEFENSES

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It is well established that water-borne chemical cues from predators or predator-wounded conspecifics can induce defensive changes of aquatic prey animals, but few examples of such inducible defenses have been reported for aquatic algae. We have found that water-borne cues from actively feeding gastropods (*Littorina obtusata*) can induce increased concentrations of phlorotannins in the intertidal brown seaweed *Ascophyllum nodosum*. Elicited *A. nodosum* plants are also less susceptible to further grazing bygastropods compared to undamaged plants. Since seaweeds have poorly developed internal transport systems and may not be able to elicit systemic induced chemical defenses through conveyance of internal signals, induction through water-borne cues ensures that seaweeds can anticipate future periwinkle attacks without receiving direct damage by herbivores. We have also found that water-borne cues from a parasite (*Parvilucifera infectans*), can serve as signals to induce morphological defence in a toxic dinoflagellate (*Alexandrium ostenfeldii*). The dinoflagellates formed temporary cysts when exposed to filtrates from cultures with infected conspecifics. The effect of the filtrate disappeared after 8 h, indicating that the chemical cues are short-lived. Furthermore, the cysts were resistant to parasite infections for several weeks after exposure to direct parasite contact, indicating that they are an effective defense. We suggest that induction of defenses in marine algae through water-borne cues may be a common phenomenon and that more examples will be revealed as this field of science progresses.

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EUGLENOIDS OF NEW JERSEY: GENUS *EUGLENA*

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Euglena gracilis is one of the most intensively studied algal species and research based on this organism has led to many significant discoveries in the fields of ecology, cell biology and physiology. Despite the importance of euglenoids to evolutionary biologists, ecologists, and the research community in general, the vast majority of euglenoids are poorly known even among biologists who regularly encounter them in a multitude of environments (fresh water lakes, marine and estuarine habitats, ephemeral pools and commensal symbionts of vertebrates). For the past few years we have surveyed the freshwater photosynthetic euglenoids of New Jersey. Our previous investigations of euglenoid phylogeny based on rRNA sequences have provided a solid framework upon which phylogenetic relationships can be inferred and morphological characters can be evaluated. The first survey focuses on the genus *Euglena*. Twelve freshwater bodies located in the piedmont region of central New Jersey have been regularly sampled for the past three years. These ponds range from mildly to highly eutrophic. Twenty-six species have been identified to date: three from the Subgenus *Euglena*, thirteen from the Subgenus *Calliglena*, and ten from the Subgenus *Discoglena*. Taxa from field collections are identified in the lab, videotaped, photographed and logged into the database for the appropriate collecting site. Morphological features, cell measurements, habitat, photomicrographs, video clips and identification notes are then recorded on a cd-rom which can be used as a visual key to the species.

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A WEB-BASED DIGITAL KEY TO EUGLENOIDS

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We are using Lucid Professional software to construct digital keys for the identification of Euglenoids. Digital keys offer a number of unique advantages over traditional paper keys, including: 1) users can describe characters in any sequence, and may skip uncertain characters, 2) the key program, by scoring as the user progresses, can “guess” what the result will be, and direct the user to the most informative characters, or narrow the field of likely taxa, 3) terms, characters, or taxa may be linked to descriptive pages that can include video clips, animations, microscopic images, and diagrammatic illustrations, as well as text. In additional to the advantages that a digital key offers over a traditional paper key, the Lucid key format includes html support, and this internet capability provides additional advantages over stand-alone digital keys. For example, the key becomes as accessible as the internet, allowing distribution to users with a broad range of interests. When run on-line, the key can hyperlink to related pages in the home site or in remote sites, allowing users to customize their experiences. While novice biologists may be directed to pages that illustrate and explain biological terms, experts will move quickly to species descriptions, which will include images, original species diagnoses, and a summary of taxonomic debate, where applicable. Our key will be housed in the Euglenoid project website (www.bio.rutgers.edu/euglena). In collaboration with others, we hope to link our keys to other similar keys so that any protistan taxa may be identified from one web address.

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ARE BLOOM-FORMING GREEN ALGAE CHEMICALLY DEFENDED?

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Ulvoid green macroalgae, such as *Enteromorpha* and *Ulva*, can form large blooms that have deleterious impacts on the local biota. These algae are often assumed to be very palatable for most invertebrate and vertebrate herbivores because they lack obvious physical defenses, and because there have been few reports of their producing chemical defenses. However, in laboratory feeding preference assays, the ulvoid macroalgae *Enteromorpha linza* and *Ulva fenestrata* were low preference foods for green sea urchins, Strongylocentrotus droebachiensis. Both these algae and several other species of green algae produce large quantities of dimethylsulphoniopropionate (DMSP), which is enzymatically converted to dimethyl sulfide (DMS) and acrylic acid when the algae are physically damaged. In laboratory bioassays, both DMS and acrylic acid were potent feeding deterrents towards urchins at concentrations that the urchins would be likely to encounter in the field. The precursor in this system, DMSP, was a feeding attractant. Our data provide evidence that DMSP functions as a precursor in an acti-vated defense system in marine macroalgae and suggests a similar function in phytoplankton. The presence of this activated defense system may contribute to the persistence of macroalgal blooms by making these algae unpalatable to some species of herbivores.

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DIVERSITY OF EUKARYOTIC PICOPLANKTON IN COASTAL WATERS

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In comparison to prokaryotic picoplankton, our knowledge of the diversity of eukaryotic picoplankton is still very limited. Recently, clone libraries from the small sub-unit ribosomal RNA gene (rDNA) from oceanic samples have revealed the presence of many new eukaryotic groups, not represented in cultures (Lopez-Garcia et al. 2001. Unexpected diversity of small eukaryotes in deep-sea Antarctic plankton. *Nature* 409**:** 603-607; Moon-van der Staay et al.. 2001. Oceanic 18S rDNA sequences from picoplankton reveal new eukaryotic lineages. *Nature* 409**:** 607-610). In the framework of the European program PICODIV, we are using the same approach to characterize picoplankton in coastal waters. In addition to clone libraries, we are also attempting to isolate novel strains in culture and to quantify key groups with oligonucleotides probes detected by fluorescence *in situ* hybridization (FISH). Some of the novel groups already observed in the Pacific Ocean are found in coastal waters pointing out their ubiquity. One such group, probably heterotrophic, occupies a basal position within the stramenopiles. Two other major novel groups are part of the alveolates and appear to be intermediate between dinoflagellates and apicomplexa. Among the photosynthetic groups, Mamiellales (Prasinophyceae), cryptophytes, chrysophytes and dinoflagellates dominate coastal clone libraries. Some sequences can be relegated to the novel class Bolidophyceae, previously known only from offshore waters. Among heterotrophic groups, ciliates are particularly abundant. Finally, coastal waters seem to harbor some novel eukaryotic groups, not observed previously such as one that appears close to the Rhodophyta.

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PATTERNS OF ALGAL ASSEMBLAGES FROM COAL MINE IMPACTED STREAMS WITHIN THE UNGLACIATED WESTERN ALLEGHENY PLATEAU

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Macroalgae and periphyton assemblages from abandoned coal mines, reclaimed sites, and reference streams within the unglaciated Western Allegheny Plateau were examined in order to determine if sites could be classified by community structure. Fifty-six streams were visited in May and June 1999. To assess the impacts of coal mining on the lotic system the periphyton and macroinvertebrates (to ascertain herbivory influence) from riffles and macroalgae from a 20-meter stream segment were collected. In addition, 29 environmental parameters (e.g. pH, metals, sulfate) and habitat conditions were examined at each stream site. A total of 594 infrageneric algal taxa and 40 macroinvertebrate taxa were recorded from these stream sites. Diatoms were the most abundant and diverse algal organisms (359 taxa), followed by chlorophytes (121), Cyanobacteria (42), euglenophytes (31), xanthophytes (14), chrysophytes (12), rhodophytes (6), dinophytes (6), and cryptophytes (3). Based on canonical correspondence analyses (CCA), the first axis was strongly correlated with pH for both periphyton ($r=-0.93$) and macroalgal assemblages ($r=-0.85$). The second axis was influenced by specific conductance for both data sets ($r=0.64$ and 0.36). Multivariate techniques typically showed five major groupings of stream reaches that were defined by certain shared environmental and biotic attributes. For example, highly acidic sites were characterized by a dominant flora of *Euglena mutabilis*, *Eunotia exigua*, *Frustulia rhomboides*, *Klebsormidium rivulare*, and *Microspora tumidula*. Predictable relationships were observed between the water quality from abandoned and reclaimed mines and the biotic assemblages present, which may prove useful in the assessment and management of reclamation efforts.

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SYNTHESIS OF MOLECULAR RESEARCH ON *BATRACHOSPERMUM HELMINTHOSUM* (RHODOPHYTA) FROM STREAM REACHES IN EASTERN NORTH AMERICA

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The freshwater red alga *Batrachospermum helminthosum* was collected from eleven streams throughout the species range in eastern North America as follows: three stream reaches from Ohio, and one each from Michigan, Indiana, Tennessee, Louisiana, North Carolina, Connecticut, Rhode Island and Massachusetts. The molecular marker technique of inter-simple sequence repeats (ISSR) and sequence data from the plastid encoded rubisco large subunit gene (*rbc*L), the mitochondrial COX2-COX3 gene spacer region, and the nuclear region of ITS1-5.8S rDNA-ITS2 were employed to examine biogeographic trends in this alga. Analysis of the *rbc*L sequence revealed 5 genotypes with one genotype representing individuals from seven stream reaches. Data from the ISSR molecular markers gave a distinct banding pattern for each of 165 individuals examined. ISSR results showed all individuals within a reach clustered together but did not provide well-defined groupings based on stream reach. The sequence data for the COX2-COX3 gene spacer was invariant among individuals from a stream reach. The individuals from Connecticut, Rhode Island and 2 Ohio stream reaches were identical and similarly the individuals from the North Carolina and another Ohio location did not vary in sequence so that seven genotypes were recorded among the individuals from the eleven stream reaches. Analysis of the ITS1-5.8S rDNA-ITS2 region showed sequence variation not only among individuals from different streams but also among individuals from the same reach. The utility and congruency of these data sets to answer biogeographic questions will be discussed.

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INTEGRATING INTENSIVE AQUACULTURE OF *CHONDRACANTHUS EXASPERATUS*, THE TURKISH TOWEL SEAWEED

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A new, high value product from the Turkish Towel Seaweed, *Chondracanthus exasperatus,* was developed recently by a Seattle company. However, Washington State has a long term moratorium on commercial seaweed harvesting from wild populations so there is renewed interest in intensive cultivation of this species. The initial phase of this research was conducted at Mukilteo, Washington. There, strategies for long term cultivation in tanks were tested, and a custom cultivation tank design was developed for pilot scale cultivation research at a site on the shore of Clam Bay near Manchester, Washington. Long term cultivation is now being tested in tanks of up to 5000 L volume supplied with natural seawater, seawater supplemented with nutrients, and seawater effluent from nearby Pacific Halibut culture tanks. Seawater from Clam Bay is naturally rich in nutrients from tidal driven upwelling and nearby commercial salmon mariculture operations. Supplemental nutrients (commercially available "f/2" enrichment) and halibut culture tank effluent have both been tested for their ability to support *C. exasperatus* growth with relatively low seawater turnover rates. Compared to seawater at the site, Halibut tank effluent differs in both nutrient composition and quantities. Initial results indicate that halibut tank effluent is a satisfactory source of nutrients for *C. exasperatus* in intensive culture and that the Turkish Towel Seaweed scrubs significant quantities of nutrients from halibut tank effluent.

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VARIANTS OF THE HISTONE-LIKE PROTEIN HGM IN THE TOXIC MARINE DINOFLAGELLATE *GYMNODINIUM MIKIMOTOI* (DINOPHYTA)

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Gymnodinium mikimotoi, (formerly *G. nagasakiense*), is a unicellular toxin-producing marine dinoflagellate, that lacks a cell wall. The absence of a cell wall, along with some other characteristics, makes *G.mikimotoi* an ideal dinoflagellate for nuclear studies. Consequently, we have developed a nuclear isolation procedure for this dinoflagellate, and identified a major histone-like protein, HGm (Wargo & Rizzo, J. Phycol. 36, 584). Here, we present an electrophoretic analysis of HGm variants, including examination of these variants in response to certain environmental factors. The HGm protein group consists of four variants, termed HGm alpha 1, HGm alpha 2, HGm beta and HGm gamma. This determination was done using both one, and two-dimensional gels. The two-dimensional gel system utilized acetic acid/urea/Triton X-100 (AUT)-PAGE as the first dimension, and SDS-PAGE as the second dimension. A comparison utilizing peptide mapping with V8 protease showed some similarity between the two alpha variants, and also between the HGm beta and HGm gamma, but not between the two different groups. These HGm variants also responded in a similar fashion to various environmental conditions tested, with the exception of nitrogen stress. Under conditions of very low nitrogen, HGm alpha 2 levels dropped lower than normal as compared to HGm alpha1 and HGm gamma, which merely matched the drop in total nuclear protein. Under these same conditions, HGm beta levels did not drop as drastically as the other HGm components or total nuclear protein. Thus the relative levels of these histone-like protein variants do not appear to be influenced by the environmental conditions tested.

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ALGAL OFF-FLAVOR COMPOUNDS IN DRINKING WATER: CHEMICAL COMMUNICATION OR CHEMICAL WASTE?

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Algae produce a diversity of potent metabolites that can modify surface water chemistry, with major socio-economic implications. Although non-toxic to humans, many algal volatile organic compounds (VOCs) cause unpleasant taste and odour (T/O), which undermines consumer confidence and increases their use of alternative and often unregulated drinking water sources. Conventional treatment may fail to remove or even intensify odour, depending on the algal species, the VOCs, and the background levels of organic material present in the source water. Furthermore, some VOCs may signal the presence of potentially toxic algal taxa. On the other hand, many odour-causing compounds signal changes in growth or metabolism, in community composition, or in ecosystem function and health. Furthermore there is growing evidence that some of these VOCs act as chemical messengers or deterrents (semiochemicals). While many odour compounds are produced by a diversity of algal (and non-algal) species, there are some general patterns in VOC chemistry and production dynamics among major algal divisions related to cell composition and the metabolic pathways involved. Depending on the VOCs and taxa, production may be intra- and/or extracellular, and vary over population cycles with environmental conditions. T/O events therefore can provide opportunities to increase our understanding of chemical interactions among organisms, and how these may lead to, or reflect changes at the level of the individual, the population, and the community.

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A REASSESSMENT OF PHYLOGENETIC SIGNAL IN DNA SEQUENCES FROM THE ITS/5.8S REGION OF THE SYNUROPHYCEAE

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The objective of this study was to enhance the phylogenetic signal contained in the nuclear-encoded ITS/5.8S region in the Synurophyceae. DNA sequences were determined for internal transcribed spacers 1 and 2, the 5.8S coding region as well as the 18S and 26S flanking regions annealing to PCR primers. DNA sequences were obtained from 15 *Synura petersenii* strains, 5 additional *Synura* species, 3 *Mallomonas* species, *Tessellaria volvocina* and *Chrysodidymus synuroideus*. Sequences from the *S. petersenii* strains and *S. uvella* aligned unambiguously. A parsimony analysis using *S. uvella* as the designated outgroup sorted the *S. petersenii* strains into two well-supported clades or ITS variants. Sequence variation was low, and sequence divergence values ranged from 0-1.4% within the ITS variants, to 3%-4% between the two *S. petersenii* ITS variants and from 8.5%-9% between S. *uvella* and the fifteen *S. petersenii* isolates. Extensive length variation occurred among the remaining synurophycean sequences (e.g. 495 bp in *S. sphagnicola* - 598 bp in *Tessellaria*) and precluded reliable alignments. In the 5.8S coding region, changes at eight positions differed between *S. uvella* and all of the *S. petersenii* strains, suggesting that 5.8S sequences should contribute characters with low homoplasy for resolving interspecific relationships in the Synurophyceae. Other workers have used derived RNA transcript secondary structure to facilitate alignments and increase phylogenetic signal in ITS 1 and 2. This presentation summarizes the previous study at the intraspecific level and investigates the interspecific phylogenetic signal in the 5.8S region and in ITS 2 alignments ascertained from secondary structure.

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STRUCTURE-ACTIVITY RELATIONSHIPS OF OLIGOAGAR ELICITORS TOWARDS *GRACILARIA CONFERTA* (RHODOPHYTA)

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Gracilaria conferta (Schousboe ex Montagne) J. et G Feldmann is able to recognize oligosaccharides that are released from its cell wall due to the enzymatic activity of microorganisms. Elicitation of the alga with oligoagars resulted within minutes in an activation of molecular oxygen and a respiratory burst. After 1 h, two strains of *Cytophaga*-like bacteria that are usually associated with *G. conferta* were eliminated by 90 – 100 %. After some more hours, a tip bleaching response was observed. Neoagarooligosaccharides were prepared by partial enzyme hydrolysis and separated on Biogel P2 and P4, yielding neoagarosaccharide fractions with a disaccharide repetition degree ranging from 1 (neoagarobiose) to more than 8 (neoagarohexadecaose). These fractions were analyzed for their biological activity towards *G. conferta*, in terms of increase of oxygen consumption, release of hydrogen peroxide, elimination of epiphytic bacteria and induction of thallus tip bleaching. The structure-activity and dose-response relationships of neoagarosaccharides were very similar in the respiratory and oxidative burst responses as well as in the bactericidal response, with neoagarosaccharides consisting of 6 to 8 disaccharide repeating units being the most active. All these responses were competitively inhibited by the reduced form of neoagarohexaose, neoagarohexaitol. In contrast, the tip bleaching response was light-dependent, required much higher concentrations of neoagarosaccharides and was not inhibited, but induced by neoagarohexaitol, suggesting that it is an oxidative stress reaction that results from the activation of a distinct mechanism. Putative structural effects on the recognition of endogenous agar-oligosaccharide elicitors by *G. conferta* are discussed.

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CHARACTERIZATION OF A MYRCENE SYNTHASE FROM SUSPENSION CULTURES OF THE MARINE RED ALGA *OCHTODES SECUNDIRAMEA*

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A monoterpene synthase from suspension cultures of the marine red alga *Ochtodes secundiramea* is shown to

biosynthesize myrcene from geranyl diphosphate (GPP) using cell free extracts. This is the first in vitro characterization of a monoterpene synthase from a marine organism. Myrcene is the likely progenitor of the unusual halogenated monoterpenes characteristic of this marine alga and, as such, represents a key step in the biosynthetic pathway. Based on mechanistic considerations from reaction with the biologically relevant substrate GPP, as well as neryl diphosphate (the cis isomer of GPP) and linalyl diphosphate (LPP), the enzyme appears incapable of catalyzing the isomerization of GPP to LPP, a mechanistic feature of most terrestrial monoterpene synthases, perhaps reflecting its evolutionarily ancient origin. The ability to assay and quantitatively monitor the expression of this enzyme in suspension cultures, under strictly defined growth conditions, presents an unparalleled opportunity to delineate, at the molecular level, factors eliciting the biosynthesis of this class of secondary metabolites, to evaluate the metabolic pathway leading to halogenated monoterpenes and to investigate their role in the chemical ecology of marine algae.

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ECOLOGY AND MORPHOLOGY OF THE SOUTH CAROLINA RED TIDE DINOFLAGELLATE, *KRYPTOPERIDINIUM*.

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Since 1998 the estuarine waters of South Carolina have been monitored for the dinoflagellate, *Kryptoperidinium sp*. (formerly *Scrippsiella carolinium*). This harmful algal bloom (HAB) species was first discovered in South Carolina waters in April 1998 when a red tide occurred in Bulls Bay, near McClellanville. Throughout the spring and summer isolated blooms of *Kryptoperidinium* were detected in South Carolina estuaries from McClellanville to Charleston. In 1999 blooms of *Kryptoperidinium* were reported in estuaries from Georgetown to Hilton Head starting in April. Monitoring efforts in 2001 have already shown the presence of this dinoflagellate in Murrells Inlet and Georgetown estuaries beginning in February. Currently, investigations are being conducted to determine bloom dynamics. In past years, bloom initiation occurred following rain events, which increased dissolved organic material loads. During blooms, *Kryptoperidinium* populations increased with decreases in DOC, DON, and DOP concentrations and increases in DIC concentrations, suggesting high heterotrophic capability. *Kryptoperidinium* red tides have occurred over a widespread area in South Carolina and have been reported worldwide. Morphology, taxonomy, and genetic research is being conducted to determine if different strains of the same cell are responsible for these widespread outbreaks or if there are morphologically similar, yet genetically different genera or species.

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CYANOBACTERIAL SYSTEMATICS IN THE GENOMIC ERA

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Over the last thirty years, cyanobacteria have become widely represented in culture collections. This has facilitated description of taxa based on biochemical, morphological, and genetic bases. Sequence data for 16S rDNA,DNA:DNA hybridization, and multi-gene sequence data are permitting rapid, phylogenetically meaningful revision of large form-genera (e.g.*Synechococcus*, *Oscillatoria*), and demonstrating the difficulties of using the botanical system of classification for identifying cyanobacteria. At present, much effort is being devoted to combining genetic characterization with traditional polyphasic approaches based on morphology and physiology in order to develop a phylogenetic treatment of cyanobacterial taxonomy. In the meantime, whole genome sequences of representatives of at least eight cyanobacterial strains from a variety of morphological types are complete or nearly complete. The results of these genome projects are demonstrating that knowledge of the complete genome facilitates taxonomic comparisons in which subtle aspects of genetic relationships and evolutionary history can be inferred. Evolution of shared characters and evidence for character loss give insight into the ecological forces that shape diversification within a lineage as well as information about the molecular mechanisms of evolution. Given the relative ease with which prokaryotic genomes can be sequenced, the rapid pace of technological development in sequencing and bioinformatics, and the informative power of complete genome sequences, it is not unreasonable to strive for a taxonomy of the cyanobacteria that is ultimately based on complete genome sequences.

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OPTICAL BIOGEOGRAPHY OF *PROCHLOROCOCCUS* AND PHYCOERYTHRIN-CONTAINING PICOCYANOBACTERIA ON THE WEST FLORIDA SHELF

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Prochlorococcus and phycoerythrin-containing picocyanobacteria co-dominate oligotrophic waters in temperate and tropical areas. Less is known about their distribution in coastal areas where optical conditions lead to shallower euphotic zones and a different attenuation spectra for incident light. In this study we compare the distribution of *Prochlorococcus* and phycoerythrin-containing picocyanobacteria (mostly *Synechococcus*) with data on inherent and apparent optical properties in a subtropical continental shelf environment. Abundance and pigment diversity for these two major groups of picoplankton was determined by flow cytometry and several fluorescence approaches at 22 stations on the continental shelf off of Tampa Bay, Florida, in the Gulf of Mexico. Hydrographic and optical data indicated that a wide range of optical environments were sampled, ranging from turbid, shallow water influenced by exchange with Tampa Bay, to very transparent oceanic water on the outer shelf. The abundance of the two major groups of picoplanktonic cyanobacteria, *Synechococcus* and *Prochlorococcus*, was negatively correlated with *Prochlorococcus* reaching peak concentrations of ~105 ml^{-1} on the outer shelf and *Synechococcus* reaching peak concentrations of ~5 X 10^5 ml^{-1} on the inner shelf. The high abundance of *Synechococcus* in turbid, green waters on the inner shelf indicates that this taxon includes a number of strains that are adapted to coastal environments and that, as a group, they do equally well in neritic waters as in oceanic regions. In contrast, *Prochlorococcus* appears to be an open-ocean specialist.

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ALGAL SYSTEMATISTS AND SYSTEMATICS OF THE RECENT PAST, WITH AN EMPHASIS ON LIFE HISTORIES.

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The past few decades have witnessed many significant advances in our understanding of algal systematics, some of these advances coming from life-history studies. Discoveries of heteromorphic life histories involving phases that had previously been given different binomials and often assigned to different families or orders have serve to shake classification schemes to their very core. A personal perspective on this recent past history will recount some of the discoveries made by phycologists that I have known from meetings or from shared field trips, such as J. Feldmann, P. Kornmann, H.-A. von Stosch, and Y. Nakamura. Notable examples of heteromorphic life histories were those demonstrated in the Derbesiales and Bonnemaisoniales by Feldmann, the Codiolophyceae by Kornmann, Liagora by von Stosch, and the Scytosiphonales by Nakamura. These reminiscences will also recount the high regard for growing algal cultures and working out life histories afforded by my PhD mentor G. F. Papenfuss in Berkeley, my post-doc mentor R. E. Norris in Seattle, and another early influence, R. Starr at Woods Hole. Similarly, the distinctive life history in *Prasiola* was an unexpected discovery made by E. I. Friedmann, as was the detection of the sexual process in *Erythrotrichia* by F. Magne. The watch-glass cultures done by M. A. Pocock in the central courtyard of the Life Sci. Bldg. at Berkeley will be recalled, as she provided critical division stages for the cytological studies made by M. Cave. Some memories of working with my departmental colleagues H. C. Bold and W. R. Taylor and joining G. Prescott in the field will be offered. The teaching and research contributions of many of these persons had a profound impact on the development of contemporary phycology.

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SEASONAL VARIATION OF PIGMENTATION OF *PERIDINIUM GATUNENSE* (DINOPHYCEAE)

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Pigment composition was measured in natural phytoplankton samples from Lake Kinneret, Israel, dominated by the dinoflagellate *Peridinium gatunense*. Peak densities were found in April, close to the water surface, with >10^9 cells$\cdot$m^{-3}, and chlorophyll *a* concentration of 380 mg$\cdot$m^{-3}, and areal chlorophyll *a* density of >1300 mg$\cdot$m^{-2}. Cellular concentrations of chlorophyll *a* changed between 201 and 282 picogram/cell, but did not show a defined temporal fluctuation. The mass ratio of chlorophyll *c* to chlorophyll *a* changed from March to June between 0.16 and 0.22, and peridinin to chlorophyll *a* ratio changed from 0.25 to 0.41. Both of those ratios did not show a clear pattern of seasonal change. On the other hand there was a linear increase of diadinoxanthin and β-carotene ratios to chlorophyll *a* throughout the season, parallel to the increase in photon flux density impinging upon the lake surface. Diadinoxantin to chlorophyll *a* ratio changed from 0.11 to 0.28 and β-carotene to chlorophyll *a* ratio varied from 0.03 to 0.08 from March throughout June. Diatoxanthin was not found in natural samples, but could have been induced in controlled experiments, with *P. gatunense* cultures. Concentration of diatoxanthin increased rapidly, concurrent with a decrease in diadinoxanthin and β-carotene concentrations, while chlorophyll c and peridinin ratios to chlorophyll *a* were almost stable with photon flux density increase. The seasonal variation of cellular pigmentation of *P. gatunense* in Lake Kinneret suggests that accumulation of photoprotective pigments is essential for optimization of photosynthetic activity of that large dinoflagellate.

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ECOSYSTEM MODELING: A TOOL TO UNDERSTAND THE INTERACTIONS BETWEEN EXTRACTIVE AND FED AQUACULTURE

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One of the most difficult tasks resource managers face is understanding the carrying capacity of coastal waters for aquaculture. Aquaculture, like many other human activities, can threaten coastal waters. Aquaculture producing shrimp and finfish depends on supplemental feeding and can contribute to eutrophication. A second type, involving shellfish and macroalgae, extracts plankton and nutrients from surrounding waters, and can have a significant positive impact on moderately eutrophic waters. Ecosystem modeling offers a 3-dimensional physical, chemical and biological simulation that can help understand and predict the impacts of aquaculture on coastal embayments. Such a model is being explored for Xincun Bay (22 km^2), which is located in southeastern Hainan Province, China. Aquaculture in Xincun Bay includes 6500 fish pens, 100 ha of shrimp ponds, pearl oyster culture rafts and *Kappaphycus alvarezii* culture that produces 2,000 mt (Oct.-May). The surrounding area has ~ 15,000 people and Xincun City is a major offshore fishing and tourist center. The annual nitrogen and phosphorus removal capacity of *Kappaphycus* in Xincun Bay may have been in the order of 53.8 and 3.7 mt, respectively, during the 1999-2000 growing season. Lian Bay (~ 15 km^2), a nearby bay with only *Kappaphycus* and pearl oyster culture (and little anthropogenic input), has a macroalgal production of 1,500 mt annually. The annual nitrogen and phosphorus removal capacity of *Kappaphycus* here may have been in the order of 25 and 0.33 mt, respectively. Our prototype model may hold the promise for showing the importance of integrating seaweed culture activities in the maintenance and health of coastal embayments.

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PHYLOGENY OF ALARIACEAE, LAMINARIACEAE, AND LESSONIACEAE (PHAEOPHYCEAE) BASED ON PLASTID-ENCODED RUBISCO SPACER AND NUCLEAR-ENCODED ITS SEQUENCE COMPARISONS

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Concatenated sequences from the plastid-encoded RuBisCo spacer and nuclear-encoded rDNA ITS region of the Alariaceae, Laminariaceae, and Lessoniaceae as currently recognized were used to determine the phylogeny of the advanced kelps (Phaeophyceae). Our analyses indicate that all taxa currently assigned to the Alariaceae, Laminariaceae, and Lessoniaceae form a well-supported monophyletic lineage. The phylogenetic analyses showed that the kelps form eight independent clades (*Egregia, Laminaria, Hedophyllum, Macrocystis, Alaria, Agarum, Ecklonia*, and *Lessonia*) with strong bootstrap support. These clades conform to the tribes of the current morphological classification system of the advanced kelps. The interrelationships among the eight lineages were suggesting a rapid, possible simultaneous radiation in the history of kelps with *Egregia* likely diverging earliest and being the sister to the other genera. Our analyses also indicate that *Kjellmaniella* and *Laminaria* do not form a monophyletic group. We will discuss the evolutionary history of the advanced kelps and provide a new framework for establishing the systematics of these commercially important brown algae.

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NITROGEN ENRICHMENT OF *PORPHYRA PERFORATA* THROUGH HIGH DOSE PULSE FERTILIZATION.

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Porphyra perforata is a highly preferred seaweed used as fodder in abalone culturing due to its relatively high nutritional value. High growth rates of abalone, particularly in the early stages, are suspected to be due the high protein-aminoacid and low water content of the *Porphyra*. Also, high NO_3 content may be important to improve the bacterial flora in the animals, which in turn may favor more efficient digestion. Changes in the composition of Porphyra, however, can occur rapidly due to environmental conditions decreasing the nutritional value of the plant. Short term N pulse fertilization were performed on *P. perforata* in order to evaluate the feasibility to increase its nutritional value. Enrichment was performed under low light conditions (<5 µE m^{-2} s^{-1}) to inhibit growth and promote higher N enrichment per unit of biomass. Tissue N in the form of NO_3, NH_4 and total organic N were measured, after 3,6,12 and 24 hrs, in tissue exposed to 500 µM of N. Results indicated a rapid N tissue enrichment particularly in the form of NO_3. Nitrate accumulation occurs continuously, up to 24 hrs. Total organic N is maximum after 12 hrs and tends to decrease after that. Fertilization with NH_4 promotes NO_3 accumulation. These results suggest the feasibility to improve the nutritional value of *P. perforata* by short-term pulse fertilization. The capacity of this species to uptake NH_4 under low light conditions (similar to those use in abalone culturing) makes it also ideal for integrated aquaculture.

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MITOCHONDRIAL CYTOCHROME B IN *PFIESTERIA PISCICIDA* AND ITS POTENTIAL USE AS A SPECIES-SPECIFIC GENETIC MARKER

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The toxic dinoflagellate *Pfiesteria piscicida* may occur in nature as a complex consisting of morphologically similarspecies or other dinoflagellates. Accurate identification of this species is critical for studies on its population dynamics and impact on the estuarine ecosystem. Genetic markers with high resolving power are thus highly desirable for field studies ofthis species. Mitochondrial (mt) DNA has been widely used in many organisms as species/population- specific genetic markers and yet has not been exploited in dinoflagellates. In the present study, a 1.1 kb cDNA fragment containing nearly the full-length mt cytochrome b gene (cytb) cloned and sequenced from P. piscicida, The nucleotide (nt) and deduced amino acid sequences of *P. piscicida* cytb (Ppcytb) showed about 55% similarities to that of protozoa, and less than 50% to the counterparts in other organisms. nt sequences of cytb showed significantly higher variation than the 18S rRNA gene and allowed design of species-specific oligonucleotide primers. Polymerase chain reaction using Ppcytb-specific primers showed high species specificity and sensitivity and when combined with specific 18S rRNA primers, *P. piscicida* can be accurately distinguished from PLO. This is the first report on dinoflagellate mt cytb and the results presented here suggest that this gene may be a useful genetic marker for species identification of dinoflagellates.

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PHYLOGENY OF THE EUGLENOPHYTES INFERRED FROM SSU AND LSU rDNA

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The phylogeny of the Euglenophytes has previously been examined using the SSU rDNA. Results from these analyses indicated that the phototrophic genera are not monophyletic. To test this hypothesis, a second gene was sequenced, the LSU rDNA. The taxa used in this study were selected from clades represented in the SSU analyses so that comparisons could be made between gene phylogenies and a combined dataset could be created. Conserved areas of the aligned sequences for both the LSU and SSU were used to generate parsimony, maximum likelihood, and distance trees. Topology of the SSU and LSU trees was similar. The SSU and LSU data consistently generated the same four highly supported terminal clades and varied only in the placement of *Euglena stellata* and *Euglena viridis*. The internal nodes of the SSU trees were weakly supported, whereas the LSU provided higher support for these nodes. A combined LSU and SSU dataset was then created. Analysis of the combined dataset yielded trees with identical topologies to those found in the individual datasets and demonstrated strong support for the four terminal clades. These results show that phylogeny of the Euglenophytes as inferred previously from SSU data is confirmed by the LSU data and that the LSU rDNA gene may be useful in elucidating relationships among the major clades.