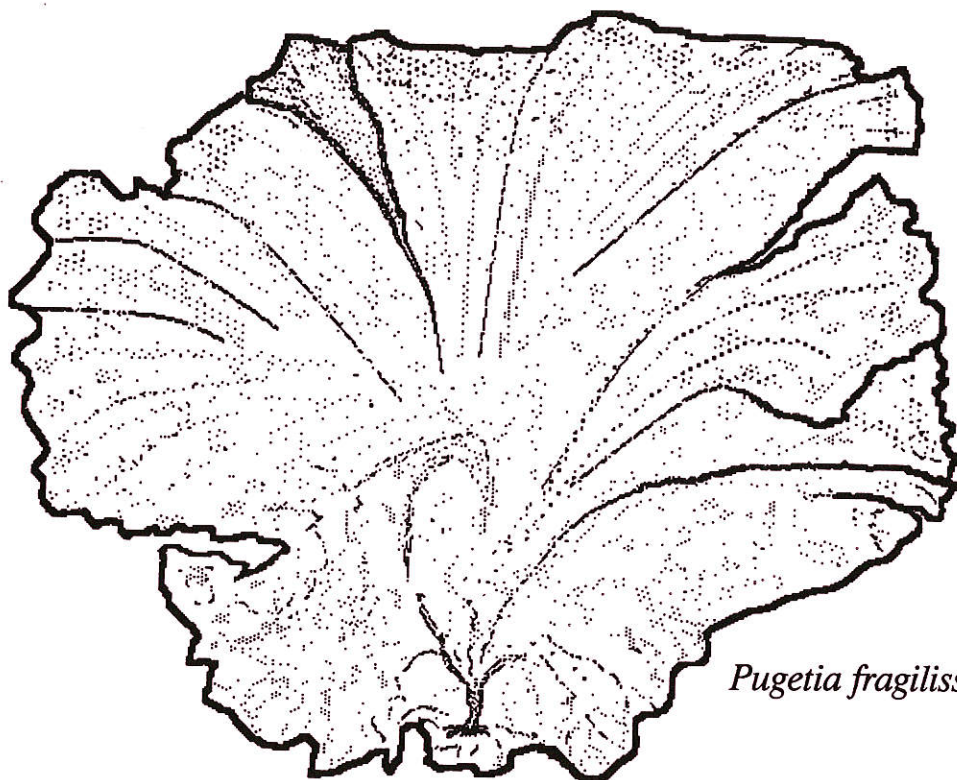


Seventh North West Algal Symposium



Pugetia fragilissima Kylin

PROGRAM AND ABSTRACTS

Fort Worden State Park
Port Townsend, Washington
April 9-11, 1993

SEVENTH NORTHWEST ALGAL SYMPOSIUM PROGRAM
Fort Worden State Park Conference Center, Port Townsend, Washington
HEADQUARTERS, PAPER & POSTER SESSIONS IN "USO" BUILDING

FRIDAY ** APRIL 9, 1993 *****

1237 LOW TIDE (-1.5 ft)

1300 ARRIVAL, REGISTRATION, HOUSING CHECK-IN, POSTER SET-UP AT USO BUILDING

1400 MARINE SCIENCE CENTER TOUR & PROGRAM (Phytoplankton and Eelgrass Communities)

1500 CHECK INTO HOUSING UNITS AFTER 1500

1600 POSTER SET-UP

1800 DINNER (DINING HALL)

2000 POSTER SESSION & SOCIAL

SATURDAY ** APRIL 10, 1993 *****

0730 BREAKFAST (DINING HALL)

0830 MINI-SYMPOSIUM: TOXIC ALGAE & TOXIC ALGAL BLOOMS. Session Chair: Rita Horner

0830 Rita A. Horner. Toxic diatoms in western Washington waters

0850 Jack Rensel. Nitrogen limitation and the spread of paralytic shellfish poisoning in Puget Sound, Washington

0910 Diane M. Altwein and Richard T. Newton. Detection and distribution of the marine toxin domoic acid on the Pacific Northwest coast.

0930 COFFEE/TEA BREAK

0950 J.N.C. (Ian) Whyte, Norma G. Ginther and Linda D. Townsend. Batch cultures of *Nitzschia pungens* forma *multiseries* for studies on domoic acid retention in commercial fisheries species

1030 John W. Chapman, Anja Robinson, Liu Xin and Peter Lawson. Could amnesic shellfish poison (ASP) in North America result from introductions of toxic marine diatoms?

1050 Janet M. Kelly. The role of cargo ship ballast discharge in harmful algal bloom events: Implications for science and policy

1110 Discussion

1145 LUNCH (DINING HALL)

1245 FIELD TRIP - Fort Worden vicinity (Low Tide, -1.27 ft @ 1326). Guides: Eric Henry & Gayle Hansen

1600 ALGAL CULTURE & DEVELOPMENT. Session Chair: Dina F. Mandoli

1600 Brenda E. Hunt and Dina F. Mandoli. A novel artificial seawater: Bringing *Acetabularia* into the 21st century

1615 Linda Runft and Dina F. Mandoli. Coordination of nucleocytoplasmic events leading to reproduction in *Acetabularia acetabulum*

1630 Ferran Garcia-Pichel, Margaret Mechling and Richard W. Castenholz. Optimal photosynthetic conditions for Oscillatorian cyanobacteria by vertical positioning within microbial mats

1645 Gregory L. Rorrer, Hans Qi and Jason Modrell. Development of liquid cell cultures from Pacific Northwest macroalgae

1900 BANQUET (DINING HALL)

2030 "Kelp on the Beach, in the Lab and at the FAR SIDE" share the experience with Louis D. Druehl

SEVENTH NORTHWEST ALGAL SYMPOSIUM PROGRAM
Fort Worden State Park Conference Center, Port Townsend, Washington

SUNDAY ** APRIL 11, 1993 *****

0700 EASTER EGG HUNT (BYOEE : Bring Your Own Easter Eggs!)

0730 BREAKFAST (DINING HALL)

0830 SYSTEMATICS & BIOGEOGRAPHY. Session Chair: Sandra C. Lindstrom

0830 Sandra C. Lindstrom. Biogeographic patterns and processes in the benthic marine algae in the northeast Pacific

0900 John W. Stiller and J. Robert Waaland. Molecular phylogenetic analysis of the red algal genus *Porphyra*

0915 Michael K. Liptack and Louis D. Druehl. Determination of parentage in apparent hybridizations among the Laminariales using the polymerase chain reaction (PCR)

0930 Ian H. Tan and Louis D. Druehl. Phylogenetic relationships among the northwest brown algal orders: A molecular approach

0945 Charlene Mayes and Louis D. Druehl. Molecular phylogeny of the genus *Laminaria* (Laminariaceae:Phaeophyta): Northeast Pacific and North Atlantic species affinities

1000 Eric C. Henry. Northwest Phaeophyceae - New records and puzzling rarities

1015 COFFEE/TEA BREAK NOTE: YOU MUST CHECK OUT OF HOUSING BY 1100!

1030 MARINE PLANT ECOLOGY. Session Chair: Annette M. Olson

1030 Jeong Ha Kim, K. Allcock, G. Eberle and R.E. DeWreede. Feeding preference of *Littorina scutulata* among three species of upper intertidal algae

1045 Darcy Lightle and Louis D. Druehl. Impact of *Egregia menziesii* (Phaeophyta, Laminariales) on the resident algal flora of Barkley Sound, B.C.

1100 Melissa McQuoid and Louis A. Hobson. Importance of the diatom resting stage in seasonal succession, past and present

1130 Annette M. Olson. Ecological models in eelgrass restoration and management: defining a research agenda

1145 BUSINESS MEETING! Agenda: 1. Date & place of 8th NWAS. 2. Affiliation with PSA 3. Other business

1200 LUNCH (DINING HALL)

1300 FIELD TRIP ("OPTIONAL") Low tide -0.73 ft @1417

SEVENTH NORTHWEST ALGAL SYMPOSIUM PROGRAM
Fort Worden State Park Conference Center, Port Townsend, Washington

POSTERS *****

TOXIC ALGAE AND BLOOMS-posters

- Kurt Buck, Francisco P. Chavez, Chris Sholin, Jon M. Krupp, Celia Villac and Greta Fryxell. *Pseudonitschia* species from Monterey Bay- Toxic and non-toxic taxa
- J. Roderick Forbes. Distribution and associated water properties of some harmful algal species on the British Columbia continental shelf
- Rita A. Horner. Toxic diatoms in western Washington waters
- Janet M. Kelly. Transport of non-native organisms in ballast sediments: An investigation of woodchip ships entering Washington State waters
- McCallum, Mary and Linda Hanson. Early warning marine biotoxin monitoring in Washington State: sentinel mussel cages and phytoplankton sampling
- P.M. Walz, D.L. Garrison, M.W. Silver, M. Cattey, W.M. Graham and R. Tjeerdema. A synopsis of phytoplankton monitoring in Monterey Bay: The *Pseudonitschia* species and domoic acid production.

ECOLOGY-posters

- Emily C. Bell and Mark W. Denny. Predicting productivity and persistence of an intertidal macroalga
- Susan A. Britting. High and dry in the intertidal: The influence of dehydration and thermal environment on protein synthesis in an alga
- Robert E. DeWreede, R.E. and Frank Shaughnessy. Biomechanical properties of life-history stages of *Iridaea*: Do they help explain stage ratios observed on the shore
- M. Wood, F. Garcia-Pichel, and S. Miller. Photosynthesis vs. irradiance relationships in ultraphytoplankton from the eastern Mediterranean
- Tanya M. Leatham. Phytoplankton community structure and seasonal succession in two central Oregon coastal lakes
- Ron Thom et al. Effects of petroleum products on bull kelp *Nereocystis luetkeana* Postels et Ruprecht

ALGAL CULTURE & DEVELOPMENT-posters

- Rene Fester, Carla Hopkins and Dina F. Mandoli. Wounds incurred in routine cell culture prolong the duration of the life cycle of *Acetabularia acetabulum* and require K^+ to heal
- W.H. Gerwick, D.G. Nagle, M.A. Roberts, J.L. Chen, P.J. Proteau, M.L. Wise, J.S. Todd and L. Meek. Oxylipins: Novel fatty acid metabolites from Pacific Northwest marine algae
- Brian R. Oates and Kathleen M. Cole. Do chloroplast division planes determine algal morphogenesis?
- Richard L. Steele and Glen B. Thursby. A toxicity test protocol for performing kelp sexual reproduction tests

SYSTEMATICS-posters

- James R. Postel. Computer taxonomy: The Linnaeus program
- Thomas B. Widdowson. A PC-based relational database for the UBC phycological herbarium
- Eric C. Henry, Russel H. Meints and Sharon Kueger. Studies of a genome of a brown algal virus

Detection and Distribution of the Marine Toxin Domolc Acid on the Pacific Northwest Coast

Diane M. Altwein, and Richard T. Newton. Federal Food and Drug Administration, Science Branch, Bothell Laboratory, Bothell, Washington.

The Seattle District Laboratory of the FDA began surveillance of seafood for the marine toxin Domolc acid in the Summer of 1991 by analyzing several oyster samples under our 1991 FY Domestic Fish and Fishery Products Inspection Assignment. Veterans Day weekend in 1991 we were asked by the Washington State Health Dept to analyze a sample of razor clams for domolc acid because they had noted a characteristic scratching syndrome in mouse bioassay for PSP toxins. The analysis revealed the presence of significant amounts domolc acid, which was confirmed by Michael Quilliam of the Canadian National Research Council Laboratory in Halifax, Nova Scotia, and prompted a wider sampling plan and an immediate alert of public officials in Washington, Oregon, Alaska, California, and British Columbia. Since that time we have continued and expanded the surveillance as well as conducted a limited comparison of the AOAC method and the Canadian NRC method and conducted a 'Round Robin Study' involving 23 laboratories across the United States and Canada to aid laboratories wanting to gear up to analyze for this toxin. So far, domolc acid has been found in significant quantities in anchovies, razor clams, and crab viscera. A number of other species have been checked for the toxin. Those species include mussels, other types of clams, oysters, scallops, shrimp, squid, and other finfish such as tuna, greenling, mackerel, sardines, and sole.

PREDICTING PRODUCTIVITY AND PERSISTENCE OF AN INTERTIDAL MACROALGA

Emily C. Bell and Mark W. Denny. Hopkins Marine Station, Pacific Grove, CA 93950 and Department of Zoology, University of British Columbia, Vancouver, B.C. V6T 1Z4 CANADA.

A computer simulation was developed that predicts the productivity of a macroalgal thallus exposed to alternating periods of emersion and submersion during the course of a lunar month. Meteorological data from a weather station was used to define the microhabitat for a Mastocarpus papillatus Kützinger thallus. An energy budget model predicted thallus temperature and relative water content during periods of emersion, while carbon fixation was based on previous physiological experiments. The simulations suggest that emersed carbon fixation is not an important component of the total carbon budget of a thallus, and that productivity is largely determined by the coincidence of daytime with periods of submersion in water. Changes in thallus morphology have only subtle effects on productivity, but dramatic effects on whether lethal temperatures are encountered during emersion. Aggregations have a similar effect on the productivity and persistence of individual thalli. Measurements of growth and demography of natural populations generally substantiate the predictions of the model.

HIGH AND DRY IN THE INTERTIDAL: THE INFLUENCE OF
DEHYDRATION AND THERMAL ENVIRONMENT ON PROTEIN SYNTHESIS IN
AN ALGA

Susan A. Britting. Department of Biology, University of
California, Los Angeles, CA 90024

Endocladia muricata (Cryptonemiales, Rhodophyta) occurs abundantly along the central coast of California. This species may be emerged for more than 10h in a 24h period in its upper intertidal habitat. Thallus hydration can range from 3-100% RWC and thallus temperatures vary between 9°C and >35°C. Presented are experiments that characterize the rate of protein synthesis and variety of proteins synthesized using radio-isotopes in emerged and resubmerged thalli of E. muricata. Characterization of the variety of proteins synthesized was derived from electrophoretic patterns (SDS-PAGE). Unique proteins were synthesized in response to elevated temperatures (35°C) during emergence. Synthesis of these proteins, however, was not apparent in resubmerged thalli recovering from elevated temperatures which were experienced during a prior emergence. A differential abundance of two proteins (30 and 33 kDa) was also apparent in emerged versus submerged incubations at all temperatures and hydration states.

E. muricata produces specific proteins in response to environmental change. Elevated thermal states and emergence produce distinct patterns of protein synthesis. Tissue dehydration, however, produces no detectable difference in the variety of proteins synthesized.

Pseudonitzschia spp. from Monterey Bay, CA: Toxic and non toxic species.

Kurt R. Buck, Francisco P. Chavez and Chris A. Scholin.. Monterey Bay Aquarium Research Institute, 160 Central Ave., Pacific Grove, CA 93950.

Jon M. Krupp. Electron Microscope Center, University California Santa Cruz, Santa Cruz CA 95064.

Celia Villac and Greta Fryxell. Dept. Oceanography, Texas A&M University, College Station, TX 77843-3126.

Monterey Bay was the site of the first reported occurrence of domoic acid in the United States during the fall of 1991. *Pseudonitzschia australis* was the diatom identified as producing the toxin, its ingestion by anchovies causing the pelican mortality that was the first symptom of this outbreak. In addition to *P. australis*, several other *Pseudonitzschia* species have been found in Monterey Bay. These include *P. subpacifica*, *P. pungens* f. *pungens*, *P. pungens* f. *multiseries*, *P. delicatissima* and *P. americana*. Identification to the species level is often dependent upon resolution obtainable only with electron microscopy of cleaned valves. We present scanning electron micrographs of the species of *Pseudonitzschia* found in Monterey Bay during the toxic bloom as well as during subsequent studies. As of yet unidentified species of this group will also be presented.

COULD AMNESIC SHELLFISH POISON (ASP) IN NORTH AMERICA RESULT FROM INTRODUCTIONS OF TOXIC MARINE DIATOMS?

John W. Chapman¹, Anja Robinson², Liu Xin¹, and Peter Lawson³.
1 Hatfield Marine Science Center, Oregon State University (OSU), Newport, Oregon 97365-5296, USA; 2 Fisheries & Wildlife, OSU Hatfield Marine Science Center, Newport, Oregon 97365-5296, USA; 3 Oregon Department of Fish and Game, Hatfield Marine Science Center, Newport, Oregon 97365, USA.

In the fall of 1991, a series of illnesses due to amnesic shellfish poisoning (ASP) in the Pacific Northwest were linked to domoic acid from razor clams. Dungeness crab, oyster, and razor clam fisheries were closed for varying periods in Oregon, California and Washington due to the persistence of the toxin and major economic losses resulted. The source of domoic acid in the 1991-1992 Oregon and Washington poisonings is assumed to be from a naturally occurring toxic diatom species of the genus Pseudonitzschia or a closely related species. The global distributions of both species most likely to be sources of ASP, Pseudonitzschia pungens multiseriata and Pseudonitzschia australis have disjunct global distributions that are associated with major shipping lanes. Ballast water from transoceanic shipping could be a mechanism for global distribution of either species and their recent toxic blooms in North America.

BIOMECHANICAL PROPERTIES OF LIFE-HISTORY STAGES OF IRIDAEA. DO THEY HELP EXPLAIN STAGE RATIOS OBSERVED ON THE SHORE?

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

Various individuals in our laboratory, working with Iridaea splendens in Vancouver and Bamfield Marine Station, have documented a predominance of haploid thalli in summer, and diploid ones in winter. Also, diploid thalli of this species often are more common on wave exposed sites than on adjacent wave sheltered sites. Habitats with very high wave impact lack Iridaea splendens and instead I. lineare is present. These distributions may, in part, be due to species and phase related biomechanical differences. We calculated force to break (FB), modulus of elasticity (ME), and per cent extension to break (EB), for all phases and the two species. In general, I. splendens stipes require a greater FB (per unit area), have a lower ME, and tolerate a higher EB, than do stipes of I. lineare. The reverse is true for the blades. Stipes of diploid thalli of I. splendens require less FB, have a higher ME, and a lower EB, than haploid thalli. When data from Carrington (1990) is combined with our data to calculate drag, diploid thalli create less drag, and I. lineare less than I. splendens. We conclude that the biomechanical properties, when considered together with an estimate of generated drag, help explain the observed distributions of the two species and their life-history phases.

WOUNDS INCURRED IN ROUTINE CELL CULTURE PROLONG THE DURATION OF THE LIFE CYCLE OF *ACETABULARIA ACETABULUM* AND REQUIRE K⁺ TO HEAL

René Fester, Carla Hopkins and Dina F. Mandoli, Department of Botany KB-15, University of Washington, Seattle, Washington 98195 USA

We have examined the pressure-wound healing response in *Acetabularia acetabulum* (L.) Silva (Chlorophyta). Commonly incurred in routine cell culture, these wounds induce disruption of the vacuolar membrane and translocation of the cytoplasm away from the wound site. Detailed analyses of the kinetics of wound healing suggest a physical connection between the vacuolar membrane and the cytoplasm. The accumulation of multiple wounds by individual cells retarded cytoplasmic healing, but had no effect on the rate of vacuolar healing. The position of the wound along the cell stalk also affected the ability of the cell to heal: cells wounded near the rhizoid healed more slowly and were less likely to achieve reproduction than were cells wounded either near the apex or at mid-stalk. The high mortality of cells wounded at the rhizoid suggests the existence of a physical structure near the primary nucleus which is essential to cell viability. The impact of wounding on reproductive potential and time to heal differed with the phase of cell development: juvenile and early adult cells healed more quickly but were less likely to complete reproduction than late adult or cells which had already begun reproduction when wounded. Growth in high population densities impaired the ability of the cells to heal. Growth of cells in seawater containing a range of potassium concentrations revealed that healing depends on potassium availability in a concentration dependent fashion.

DISTRIBUTION AND ASSOCIATED WATER PROPERTIES OF SOME HARMFUL ALGAL SPECIES ON THE BRITISH COLUMBIA CONTINENTAL SHELF

J. Roderick Forbes, Department of Fisheries and Oceans, Institute of Ocean Sciences, P.O. Box 6000, Sidney, B.C., Canada. V8L 4B2

During eleven years of sampling of phytoplankton species composition and abundance on the continental shelf of British Columbia, data has been collected on the occurrence of a number of harmful or potentially harmful species. Information is presented on seasonal occurrence and associated water properties for a variety of species, including *Heterosigma akashiwo*, several other Raphidophyceae, *Pseudonitzschia* spp, *Chaetoceros concavicornis/convolutum* and several dinoflagellates. Some preliminary analysis is undertaken to see if there is potential for predicting the occurrence on the basis of water properties measurable in the field.

OPTIMAL PHOTOSYNTHETIC CONDITIONS FOR OSCILLATORIAN CYANOBACTERIA
BY VERTICAL POSITIONING WITHIN MICROBIAL MATS.

Ferran Garcia-Pichel, Margaret Mechling, and Richard W. Castenholz.
Biology Department, University of Oregon, Eugene, OR 97403, USA.

Marine microbial mats are often perennial, lamellated structures in which cyanobacteria predominate as the accreting surface layer. The structure is commonly a fabric of microbial filaments and their remains as well as mucilaginous material of microbial origin. Although most of the microorganisms are non-motile or immobilized within the compact mat, some species "migrate" vertically over a distance of a few millimeters or less within the mat as a response to the steep physical and chemical gradients, often showing a diel periodicity. In the perennial mats of hypersaline ponds near Guerrero Negro, Baja California Sur, Mexico, a few prominent species of cyanobacteria and the non-photosynthetic sulfide/sulfur-oxidizing bacterium, Beggiatoa sp. migrate in this way. These movements have been quantified by repeated corings of the mats with glass capillary tubes. "Mini-cores" allowed the microscopic determination of the depths of specific layers of microorganisms, using color, fluorescence, and distributions of identifiable species over the course of a 24 h period. These samplings were accompanied by micro or semi-micro measurements of O₂, sulfide, and spectral irradiance within the mat. As a result of movements to the mat surface towards dusk and the downward movement after dawn, and the retention of high pigment content, Spirulina and Oscillatoria spp. were able to escape photoinhibitory light but also maintain photosynthetic saturation for most of the day. Possible cues for initiating these movements will be discussed.

OXYLIPINS: NOVEL FATTY ACID METABOLITES FROM PACIFIC NORTHWEST
MARINE ALGAE.

W.H. Gerwick, D.G. Nagle, M.A. Roberts, J.L. Chen, P.J. Proteau, M.L. Wise, J.S. Todd, and L. Meek, College of Pharmacy, Oregon State University, Corvallis, Oregon 97331.

Marine algae are recognized as a rich source of polyunsaturated fatty acids including a number of those associated with the beneficial dietary effects of fish oils. In man, fish oil derived fatty acids act as substrates in metabolic oxidation reactions producing key inflammatory mediators such as prostaglandins, leukotrienes, thromboxanes, and hydroxyeicosanoids. Our ongoing research has shown marine algae, particularly temperate species, to contain numerous unusual oxidized fatty acid metabolites, collectively known as oxylipins. In mammalian systems these metabolites are typically derived from the 20 carbon precursor arachidonic acid and generally referred to as eicosanoids. Eicosanoids and eicosanoid receptors have recently been shown to be found in the cells of nearly all human tissues and are of vital importance in maintaining normal mammalian physiology. Dietary substitution of unusual fatty acids is thought to alter physiological processes including those involved in heart disease by producing fatty acid oxidation products with significantly different biochemical and physiological properties. Some of these algal metabolites are simple hydroxy acids identical to those found in mammalian systems. Others are structurally unique chemical structures with biological and pharmacological properties which we have only recently begun to explore. We have also found that some algae possess previously undescribed metabolic pathways capable of transforming simple fatty acid precursors into complex novel compounds. An overview of our findings including chemotaxonomic and biosynthetic perspectives will be provided.

NORTHWEST PHAEOPHYCEAE- NEW RECORDS AND PUZZLING RARITIES

Eric C. Henry, Dept. of Botany & Plant Pathology, Oregon State University, Corvallis, OR 97331-2902.

Although considerably less species-rich than the Rhodophyte flora, the Phaeophyte flora of northwest North America continues to present puzzling aspects. In Oregon, *Sphacelaria plumosa*, *Saundersella simplex*, *Spongonema tomentosa* and *Dictyota binghamae* are known only from single or very rare collections, even though they are common species in other localities. Two exotics, *Acinetospora* sp. (European?) and *Scytothamnus* sp. (S. Hemisphere) have been found in the last year, the latter also known from British Columbia for 15 years. The Atlantic *Pilinia rimosa* (*Waerneilla lucifuga*) was also recently discovered in British Columbia. While it may not be surprising that small subtidal algae such as *Syringoderma* have only spotty known distributions, it is not clear why this is so for the kelp *Eisenia arborea*. The huge area of the Alaska coastline promises additional new discoveries; the presence of cold-water Atlantic forms such as *Omphalophyllum* suggests that *Tilopteridales*, the only brown algal order restricted to the N. Atlantic, may also exist in the Pacific. In fresh water, *Heribaudiella*, one of the very few freshwater brown algae, is widely distributed in Europe but remains known in N. America only from B.C. and northern Washington and at Yellowknife in the Canadian Northwest Territories.

STUDIES OF THE GENOME OF A BROWN ALGAL VIRUS

Eric C. Henry, Russel H. Meints and Sharon K. Krueger. Dept. of Botany & Plant Pathology, Oregon State University, Corvallis, OR 97331-2902.

A large (150 nm diameter) icosahedral virus causes a stable virus infection in sporophytes of an undescribed species of *Feldmannia* from New Zealand. The infection can be demonstrated directly by Pulsed-Field Gel Electrophoresis (PFGE) performed on intact algal filaments, without the necessity of first making algal protoplasts. Under optimal PFGE parameters the double-stranded DNA from the virus migrates as two bands of 165.5 and 180 kilobase pairs, which we interpret as two distinct viral genomes. Analysis by restriction digests of the co-occurring genomes together, and separated by PFGE, and probing of Southern blots of the two genomes with small cloned portions of the viral DNA, demonstrate that the genomes are highly similar but also exhibit distinct differences. At lower culture temperatures the smaller band is reduced in intensity, suggesting that it represents a temperature-sensitive deletion mutant. The behavior of the infection in culture suggests that the viral genomes integrate into the genome of the algal host, and this is confirmed by detection of viral DNA restriction digest patterns on Southern blots of restriction digested DNA from *Feldmannia* sp. gametophytes, which show no overt signs of virus infection.

TOXIC PHYTOPLANKTON ON THE PACIFIC COAST: A REVIEW

Rita A. Horner. School of Oceanography, WB-10, University of Washington, Seattle, WA 98195.

The earliest known outbreak of paralytic shellfish poisoning occurred in British Columbia in 1793, but the largest mass mortality occurred in 1799 when more than 100 people died after eating mussels collected in Peril Strait, near Sitka, Alaska. It wasn't until 1937 that toxicity was found to be associated with the dinoflagellate *Gonyaulax* (now *Alexandrium*) *catenella* and a mouse bioassay was developed to detect the toxin. Monitoring for PSP began in California in 1939, in Washington and British Columbia in 1942, and in Oregon in 1958. In Alaska, regular monitoring is done only at a few selected sites because of the extremely long coastline. Monitoring has led to annual and temporary closures of areas along the Pacific coast because of PSP levels greater than $80 \mu\text{g g}^{-1}$ of shellfish meat.

Although not known to be toxic to humans, the diatoms *Chaetoceros concavicornis* and *Ch. convolutus* and the raphidophyte flagellate *Heterosigma carterae* all kill fish, particularly Atlantic salmon, grown in net pens. *Heterosigma* occurs in British Columbia each summer and occasionally also in Washington.

Perhaps the greatest threat to human safety from harmful phytoplankton is the newly discovered neurotoxin, domoic acid. This toxin, originally isolated from *Chondria armata* in Japan, is also produced by diatoms in the genus *Pseudonitzschia*. The toxin occurred in Monterey Bay, California, in September 1991, and killed pelicans and cormorants. In October 1991, it was found in razor clams in Oregon and Washington, and in November 1991, in Dungeness crabs.

TOXIC DIATOMS IN WESTERN WASHINGTON WATERS

Rita A. Horner and James R. Postel. School of Oceanography, WB-10, University of Washington, Seattle, WA 98195.

Members of the planktonic diatom genus *Pseudonitzschia* H. Peragallo, *P. pungens* (Grunow.) Hasle f. *multiseries* (Hasle) Hasle and *P. australis* Frenguelli, that may produce the marine biotoxin, domoic acid, have been recognized in western Washington waters. Their distribution is not well-known, probably because they often have been misidentified. However, they appear to be relatively common and may be abundant, especially in late spring and summer. Domoic acid, a potent neurotoxin, was found at levels up to $154 \mu\text{g g}^{-1}$ wet weight in razor clams, *Siliqua patula* Dixon, and Dungeness crabs, *Cancer magister* Dana, on the pacific coast of Washington in late October 1991. It was also found in trace amounts in blue mussels, *Mytilus edulis* Linnaeus, and oysters, *Crassostrea gigas* Thunberg, in the inland waters of northern Puget Sound in summer 1992. The presence of these potentially toxic diatoms signals the need for regular phytoplankton monitoring and additional shellfish monitoring to ensure that seafood is safe for human consumption. Further, studies are needed on the physiological ecology of the diatoms to determine the causative factors leading to production of the toxin.

A NOVEL, ARTIFICIAL SEAWATER ALLOWS RAPID GROWTH AND COMPLETE DEVELOPMENT OF *ACETABULARIA ACETABULUM* WITHOUT A CHANGE OF MEDIUM.

Brenda E. Hunt and Dina F. Mandoli. Department of Botany KB-15, University of Washington, Seattle, Washington 98195, USA.

Few mutants of *Acetabularia acetabulum* (L.) Silva (Chlorophyta), a classic unicellular model for cell biology since the 1930's, have ever been selected and most of these have been lost due to difficult and labor intensive cell maintenance. To make genetics feasible in this giant alga, previously we have 1) improved mating efficiency from $\leq 1\%$ to $\geq 62-100\%$ (Mandoli & Larsen, submitted); 2) developed methods which facilitate handling and growth of millions of cells at a time (Zeller & Mandoli, March 1993, Phycologia); and 3) reduced the juvenile phase 3-10 fold by adjusting growth conditions for the zygotic phase of development (Mandoli et al., unpublished).

Now, we have developed a novel, artificial seawater for axenic culture of *A. acetabulum* by physiologically optimizing the concentrations for each element in the medium. In "Ace seawater", *A. acetabulum* grows from juvenile through gametangial formation in 6-9 weeks at high cell densities (≤ 1.5 mL/cell), without changing the medium. **Development of this novel seawater has reduced the labor of routine *A. acetabulum* cell culture by 80%, the space required for growing large numbers of cells by 30%, and the duration of the life cycle by $\geq 50\%$.** Data on additional changes in the seawater recipe and applicability to growth of other marine species will be discussed. **This substantial advance in cell culture makes feasible genetic selections and screens for mutants in *A. acetabulum* for the first time.**

THE ROLE OF CARGO SHIP BALLAST DISCHARGE IN HARMFUL ALGAL BLOOM EVENTS : IMPLICATIONS FOR SCIENCE AND POLICY.

Janet M. Kelly. School of Marine Affairs, University of Washington, Seattle, WA 98195.

Ballast water and sediments from bulk cargo carriers have been implicated in the transfer of a diverse assortment of non-native species to near-shore environments worldwide. Two major invasions attributed to ballast discharge have catalyzed policy efforts on sub-national, national and international levels. One of these events involves toxic microalgae.

Australian researchers identified dinoflagellate cysts present in discharged ballast sediments and believe they are responsible for the introduction of PSP-causing algal blooms and the subsequent disruption of the shellfish culture industry in Tasmania. Although 14 non-native species of fish, invertebrates and macroalgae have been identified in Australia as ballast introductions, current policy efforts are directed toward minimizing the introduction of toxic microalgae.

In Washington state, incubation of ballast sediment samples taken from Japanese woodchip carriers arriving at the ports of Tacoma and Port Angeles resulted in a proliferation of microalgae including diatoms and dinoflagellates. In 1988, Norwegian pollution control authorities convinced international shipping officials to alert vessels trading in Northern Europe to amend ballasting operations to avoid spread of toxic algae during a severe bloom event.

In light of recent evidence that harmful algal blooms appear to be increasing in frequency and magnitude on a global level, the need to assess the role of shipping as a mechanism of transfer is necessary. Recognition of this mechanism by phycologists forces re-thinking of conventional concepts relating to cosmopolitan species and origin of bloom events. Documentation and analysis of bloom events in relation to shipping activity is essential for further understanding of this issue.

TRANSPORT OF NON-NATIVE ORGANISMS IN BALLAST SEDIMENTS: AN INVESTIGATION OF WOODCHIP SHIPS ENTERING WASHINGTON STATE WATERS.

Janet M. Kelly. School of Marine Affairs, University of Washington, Seattle, WA 98195.

An investigation of cargo vessels engaged in the export of woodchips from Washington state was conducted to determine whether ballast discharge (water and sediments) can act as a mechanism for transfer of non-native organisms, particularly toxic microalgae.

Samples of ballast water and sediments from woodchip carriers entering the ports of Tacoma and Port Angeles were found to contain numerous viable organisms which survived the 10-13 day trans-oceanic voyage from Japan to Washington. Water samples contained live zooplankton and phytoplankton including larval bivalves, gastropods, polychaetes and fish as well as amphipods, isopods and copepods.

Ballast sediment samples contained few motile cells. However, incubation in nutrient-enriched seawater induced a proliferation of microalgae including various diatoms, dinoflagellates and phytoflagellates. These incubations trials suggest the presence of benthic spores and cysts which may remain viable for extended periods of time in the receiving port environment.

Interviews with ship personnel provided insight into the variety of operational procedures used to discharge and dispose of ballast. The presence of microalgae in ballast sediments is of particular note as recent policy efforts to have focussed on the discharge of ballast water. For bulk cargo carriers, the procedure for disposal of sediments is almost always conducted within port waters, whereas ballast water may be discharged offshore. This difference is critical to the creation and implementation of future control efforts.

Feeding preference of *Littorina scutulata* among three species of upper intertidal algae.

J.H.Kim, K.Allcock, G.Eberle and R.E.DeWreede. Department of Botany, University of British Columbia, Vancouver, B.C. V6T 1Z4 CANADA

Three dominant algae, *Iridaea cornucopiae*, *Pelvetiopsis limitata* and *Fucus distichus*, and the most abundant littorine, *Littorina scutulata*, from the upper intertidal on the west coast of Vancouver Island, B.C., Canada, were studied. Feeding preference tests with the above algae were undertaken in order to understand the littorine's role in affecting upper intertidal community structure. A series of laboratory experiments were conducted with different snail densities and different feeding times. Tides and daily irradiance were simulated. Portions of three algal species were presented as either single choice or multiple choice to *L.scutulata*. *I.cornucopiae* was heavily grazed in most single choice experiments and differences in weight loss between treatments (with snails) and controls (without snails) were significant. No significant grazing occurred on the two fucoids, except for *P.limitata* with a higher density of snails. In multiple choice experiments, *I.cornucopiae* was the most palatable species, followed by *P.limitata* and *F.distichus*. More grazing activity seemed to occur during "low tide" than "high tide". In this experiment, we propose a new statistical method for analyzing the data from feeding preference experiments.

Phytoplankton community structure and seasonal succession in two central Oregon coastal lakes.

Tanya M. Leatham. Department of Biology, University of Oregon, Eugene Oregon, 97403.

Lakes on the central Oregon coast have been the subjects of controversy over the past 20 years regarding the effects of increases of residential housing developments on water quality. Several studies have been conducted including a baseline data set for the evaluation of long term trends in the trophic status of 13 lakes (Larson 1974). Changes in phytoplankton community assemblages can reflect changes in the trophic status of a lake (Hutchinson 1967). However, naturally occurring seasonal succession patterns must be considered (Reynolds 1984). This study, currently in its second year, provides information on the current trophic status of two coastal lakes, Mercer and Tahkenitch, through monthly examinations of phytoplankton community structure and density, chlorophyll *a* concentration, and secchi depth, as well as oxygen, temperature, and light profiles. A long term approach to the study is being taken to determine if patterns of seasonal succession are consistent through time. Comparisons with previous studies will help assess directional patterns in trophic states for these recreationally important lakes.

Impact of Egregia menziesii (Phaeophyta, Laminariales) on the resident Algal Flora of Barkley Sound, B.C.

Darcy Lightle and Louis Druehl. Bamfield Marine Station, Bamfield, B.C. V0R 1B0.

Egregia menziesii (Turner) Areschoug is a common macrophyte on the west coast of Vancouver Island. In the Barkley sound area it is often one of the more dominant plants in the intertidal zone. This may not have always been the case. A survey of various sites in Barkley Sound in 1969 showed specific areas where Egregia was not present. In 1990 some of these sites were found to have Egregia present, one site now has a density of 2.5kg/m². A permanent transect was set up to examine the effects harvesting Egregia would have on the other seaweeds in the immediate area. Alternate control and treatment (Egregia removed) sites were measured for individual percent cover of nine other species at the start of the study in summer of 1991 and were remeasured in summer of 1992. An analysis of the data showed a significant difference in the total percent cover between the control and treatment of the first year and the control and treatment of the second year. A comparison of the individual species showed some of them increased their relative percent cover with the treatment plots while others remained approximately the same or decreased.

DETERMINATION OF PARENTAGE IN APPARENT HYBRIDIZATIONS AMONG THE LAMINARIALES USING THE POLYMERASE CHAIN REACTION (PCR).

Michael K. Liptack and Louis D. Druehl. Simon Fraser University, Burnaby, B.C. V5A 1S6 CANADA and Bamfield Marine Station, Bamfield, B.C. V0R 1B0 CANADA.

Establishing the parentage of apparent hybrids is complicated by the possible production of "sporophytes" by apogamy and the potential autodiploidization of these haploid "sporophytes". The Polymerase Chain Reaction (PCR) is an effective tool in determining the parents of potential hybrids because of the controlled specificity of the primer hybridizations and the small amount of template DNA necessary. For this study, we used genera specific primers from the ITS1 region of the ribosomal cistron and one primer complimentary to a sequence in the highly conserved 18S region of the ribosomal cistron in each PCR reaction. The primers were designed to produce a different sized band for each genus and the resulting bands were separated by gel electrophoresis. The sizes of the bands were used to determine the origins of the DNA present in crosses from gametophyte cultures. This technique required only a small number of cells and as a result could be performed on small samples or very young "sporophytes".

BIOGEOGRAPHIC PATTERNS AND PROCESSES IN THE BENTHIC MARINE ALGAE IN THE NORTHEAST PACIFIC

Sandra C. Lindstrom, Department of Botany, University of British Columbia, Vancouver, B. C., Canada V6T

The distribution of species of benthic marine algae along the coasts of Washington, British Columbia, and Alaska is examined from a biogeographical perspective. Several patterns of changes in species composition are recognized to occur as one goes from the outer coast to the inner coast. In one pattern, species drop out along a gradient of decreasing salinity, the factor most often cited as responsible for changes in the flora. In a second pattern, species are added to the flora as one approaches areas of strongly mixed waters. These additions to the flora represent range disjunctions, which require an explanation of how they came to be. Methods for testing the factors hypothesized to be responsible for these patterns will be discussed.

MOLECULAR PHYLOGENY OF THE GENUS *LAMINARIA*
(LAMINARIACEAE: PHAEOPHYTA): Northeast Pacific and North Atlantic
Species Affinities

Charlene Mayes & Louis D. Druehl.
Simon Fraser University, Burnaby, B.C. V5A 1S6
and Bamfield Marine Station, Bamfield, B.C. V0R 1B0,
CANADA

Phylogenetic relationships among eleven representative species of the kelp genus *Laminaria* Lamouroux, including *L. angustata*, *L. digitata*, *L. ephemera*, *L. groenlandica*, *L. japonica*, *L. longipes*, *L. saccharina*, *L. sinclairii*, *L. setchellii*, *L. solidungula*, and *L. yezoensis* were inferred from nuclear encoded ribosomal DNA sequence data encompassing the ITS1 (Internally Transcribed Spacer 1) and 5.8S gene. A second phylogeny was inferred for a subset of the study species including additional sequence data from the ITS2 region of the ribosomal cistron. The resulting phylogenies facilitate a re-examination of the genetic and evolutionary affinities between *Laminaria* species of the Northeast Pacific and North Atlantic oceans.

In addition, two Japanese species, *L. angustata* and *L. japonica* were included in this comparison as Northwest Pacific ocean representatives of the genus.

Importance of the diatom resting stage in seasonal succession, past and present.

McQuoid, Melissa and Louis A. Hobson. Department of Biology, P.O. Box 1700,
University of Victoria, Victoria, B.C. V8W 2Y2 CANADA.

Diatoms are a major component of the spring phytoplankton bloom that is often seen in near-shore waters. Diatom species appear in a predictable succession as the spring bloom progresses. However, little is known about the source of the bloom and the forces driving this succession. It is possible that a very low concentration of vegetative cells might be able to initiate a bloom or that there is some translocation of cells to the bloom area either by lateral advection or upwelling from the sediments. In the sediments diatoms can exist in a resting state. Resting spores can be formed in response to nutrient limitation and are able to withstand periods of unfavorable conditions, such as those found in the benthos. Resting spores may either become buried, forming a sedimentary record, or germinate, seeding the next year's diatom bloom. The role of environmental cues in the germination sequence of different species of diatoms is currently under investigation using laboratory cultures. Data suggest that the effects of temperature and photoperiod on resting spore germination is species specific, which could result in the observed patterns of seasonal succession. Those resting spores that do not germinate leave a historical record of that year's bloom in the sediments. These cells have thicker frustules than those of the vegetative life stage, making them more resistant to diagenesis in the sediments. The historical record of diatom vegetative cells and resting spores in Saanich Inlet sediments is being examined using light and scanning electron microscopy.

DO CHLOROPLAST DIVISION PLANES DETERMINE RED ALGAL MORPHOGENESIS?

Brian R. Oates and Kathleen M. Cole. The Department of Botany, University of British Columbia, Vancouver, B. C. V6T 1Z4 CANADA.

Chloroplast division planes have recently been shown to be markers for the planes of cytokinesis in Bangia vermicularis (Oates and Cole; Protoplasma, (1992) 169:155-167). Since algal cytokinetic planes and ensuing cell walls are fixed in place this finding suggests that chloroplast division planes play key roles in establishing thallus morphology. Cells of Bangia are filled by large, lobed plastids that divide through a previously undescribed process resulting in narrow cytoplasmic channels (CCs). The cylindrical morphology of Bangia is established early in the first periclinal division of the cylindrical cells at the base of the filamentous meristem as multiple plastokinesis commences, generating several radially-arranged wedge-shaped plastids separated by narrow CCs. The CCs between the daughter plastids serve as sites of new radially-arranged cell walls during cytokinesis, thus establishing the cylindrical morphology. Preliminary observations of the closely related Porphyra and Smithora have provided evidence suggesting that similar plastokinetic and cytokinetic phenomena take place during cell division in these algae. These similar division patterns may be related to the chloroplast size relative to the cell size. Explanations for the different morphogenetic patterns in these algae will be presented.

ECOLOGICAL MODELS IN EELGRASS RESTORATION AND MANAGEMENT: DEFINING A RESEARCH AGENDA

Annette M. Olson. University of Washington, Seattle, WA 98195 USA.

Ecological models are important for environmental decision-making, because they define the ecological information needed for decisions and they provide a framework for interpreting that information in a policy context. I propose to evaluate the role of ecological models in basic and applied research on eelgrass (Zostera) beds, a critical habitat for numerous important fish and invertebrates in the Pacific Northwest. I am cataloging the published ecological knowledge on eelgrass-dominated systems of the North Pacific and quantifying the types and frequency of models implicitly or explicitly tested. I can use this database to identify trends and gaps in research that may presently limit policy development for management and restoration of this critical habitat. I will then use this analysis to systematically formulate an agenda for new ecological research directions in the ecology of eelgrass habitats.

COMPUTER TAXONOMY: THE LINNAEUS PROGRAM

James R. Postel. School of Oceanography, WB-10, University of Washington, Seattle, WA 98195.

'Linnaeus Protist' software, for taxonomic identification, was developed at the Institute of Marine Research, Bergen, Norway, with support from the UNESCO program for the promotion of marine sciences (PROMAR). It contains information on over 350 species of marine protists (algae and protozoans) from northern European coastal waters. The program has taxonomic keys with the species presented by descriptions, line drawings, photomicrographs (LM and EM), and references. In addition, there are distribution maps of toxic and bloom-forming species, methods in protistology, glossary, and master reference list.

The program works on Macintosh computers with HyperCard version 2.0 or higher and System 6.0.5 or higher; the full version 1.0 occupies about 35 Mb. It is available for a nominal cost (\$33) on a CD-ROM that also has Linnaeus Zooplankton, the first taxonomic key made with HyperCard, and Linnaeus Toolkit, an empty version of the protist program that allows the user to enter new phyla into the program. Additional protist species can be added directly into Linnaeus Protist.

The program, demonstrated using a Macintosh Powerbook and NEC CD player, will be available for testing.

NITROGEN LIMITATION AND THE SPREAD OF PARALYTIC SHELLFISH POISONING IN PUGET SOUND, WASHINGTON.

JACK RENSEL School of Fisheries, HF-15
University of Washington, Seattle, Washington 98195.

Paralytic shellfish poisoning (PSP) has spread throughout much of Puget Sound, Washington since the mid 1970's. Now all but parts of southern Puget Sound (SPS) and all of central and southern Hood Canal (CHC and SHC) are affected by PSP. The initial spread of PSP has been traced to major physical events, but lack of PSP in most of SPS and all of CHC and SHC has not been investigated until this study. Monitoring and experimental data suggest the lack of surface and subsurface (10 m) dissolved inorganic nitrogen in the unaffected areas prevents the growth of *Alexandrium catenella* (Whedon and Kofoid) Balech.

In August of 1991 and 1992, filtered water from the surface and subsurface depths of CHC did not support any growth of *A. catenella* in the laboratory. However, growth of *A. catenella* occurred with water from the same depths in SHC, although PSP has never been reported in that area. Slow estuarine flow transport in CHC coupled with a seasonal lack of nitrogen in surface and subsurface waters forms a barrier to the passage of *A. catenella* cells to the more nutrient-rich SHC. Correlation analysis showed that PSP toxicity in mussels was related to elevated subsurface nitrogen concentrations and water temperature above 13° C. Increased nitrogen discharge from rapid urbanization and non-point sources could lead to annual PSP problems in areas presently unaffected by PSP, unless preventative measures are taken.

DEVELOPMENT OF LIQUID CELL CULTURES FROM PACIFIC NORTHWEST MACROALGAE.

Gregory L. Rorrer, Hans Qi, and Jason Modrell, Department of Chemical Engineering, and William H. Gerwick, College of Pharmacy, Oregon State Univ., Corvallis, OR 97331.

Liquid cell suspension cultures of marine macroalgal plants have potential to biosynthesize valuable natural products and biomedicinals in a controlled environment. Since cell culture systems for marine macroalgae are not well developed, we considered callus induction, protoplast isolation, and gametophyte isolation techniques for establishing liquid cell cultures from selected red and brown macroalgae found off Oregon coastal waters. Undifferentiated, filamentous callus tissue was induced from sterilized stipe explants of the brown alga *Laminaria setchellii* after six weeks of incubation in the dark at 8 °C on PES agar medium. However, continued cultivation on liquid medium did result in sustained growth of callus tissue. Protoplasts were obtained from the red algae *Porphyra* and *Gracilariopsis lemaneiformis* by hydrolytic enzymic digestion of sterilized explant blade tissue and subsequently cultivated in PES liquid medium at 15 °C under photosynthetic conditions, but sustained growth of the cell suspension was not observed. Female, filamentous gametophytes isolated from *Laminaria saccharina* could be cultivated as a relatively uniform liquid suspension in an illuminated bubble-column bioreactor, but not in agitated shake-flask culture.

COORDINATION OF NUCLEOCYTOPLASMIC EVENTS LEADING TO REPRODUCTION IN ACETABULARIA ACETABULUM

Linda Runft and Dina F. Mandoli, Department of Botany KB-15, University of Washington, Seattle, Washington 98195 USA

We have quantitated how the nucleus and apex coordinate the switch from vegetative to reproductive development in the single-celled alga *Acetabularia acetabulum* (L.) Silva (Chlorophyta) using differentiation in the absence of the nucleus, cell amputation, and DAPI staining. Nucleocytoplasmic coordination is significant for this organism both because the nucleus and the site of morphogenesis are separated by 4-6 cm and because all the cytoplasm of the parent is partitioned into its progeny. Vegetative growth in *Acetabularia*, characterized by growth of the cell stalk, terminates with initiation of the reproductive structure, a cup-shaped "cap", which grows perpendicular to the stalk. Reproductive **competence** requires that a cell attain a certain age and a minimum cytoplasmic volume: an apex needs 5/8 or 3/8 of the cytoplasm to make a cap in the absence of the nucleus when it is in the "early adult" or in the "late adult phase" of development, respectively. Morphologic **determination**, a 1 d event at the cell apex that is blue light-regulated, may occur at a specific circadian time. Cells initiate another cap either when a cap aborts or is amputated: a cell can initiate up to 23 caps in succession at a rate of 1cap every 11 days. The last **checkpoint**, when a cell can return to an earlier time in development and re-attempt to reproduce, occurs during cap expansion: aborted caps reach $61 \pm 5\%$ of their mature diameter about 13 days post cap initiation. **Commitment** to reproduction occurs 15 days after cap initiation and corresponds to 72% of the mature cap diameter: the ability of a cell to form a second cap declined dramatically if it was amputated ≥ 15 days after cap initiation. Use of DAPI staining to see how commitment corresponds to meiosis will also be discussed. Events which occur after commitment (mitosis, nuclear transport to the cap, cap-stalk partitioning, and gametangial formation) will be described.

A TOXICITY TEST PROTOCOL FOR PERFORMING KELP SEXUAL REPRODUCTION TESTS.

Richard L. Steele and Glen B. Thursby. Environmental Protection Agency, 2111 S.E. Marine Science Drive, Newport, Oregon 97365-5260 USA and Science Applications International Corp., Environmental Testing Center, 165 Dean Knause Drive, Narragansett, Rhode Island 02882-1154 USA.

A toxicity test has been devised using kelp gametophytes. Most research has been accomplished using Laminaria saccharina but other genera and species are being considered. Spores released from large diploid (2N) sporophytes can be isolated and develop into male or female gametophytes which are haploid (1N). Gametophytes are grown under conditions that insure vegetative growth and can be grown in quantities needed for testing. Cells are blended, dispersed, and grown under conditions inducing reproduction for four days. At the end of four days, the inducing medium is replaced. Females are put into testing vessels with males with testing medium containing toxicants for two days. At the end of two days the males are removed and the testing medium is replaced with recovery medium. The sporophytes that form are allowed to grow for two days and are counted at this time. Results of testing indicate that kelp sexual reproduction is especially sensitive to organic pollution, particularly petroleum products. Kelp reproduction does not appear to be as sensitive to some metals.

MOLECULAR PHYLOGENETIC ANALYSIS OF THE RED ALGAL GENUS *PORPHYRA*.

John W. Stiller and J. Robert Waaland. Dept. of Botany, University of Washington, Seattle, WA, 98195.

Despite a great deal of research focused on the taxonomy and systematics of *Porphyra* (Bangiaceae, Rhodophyta), species identification remains problematical and little is known about phylogenetic relations among species. The genus is large, has an ancient evolutionary history but offers few morphological characters for systematic analysis. This has probably lead to repeated evolutionary convergence of morphological characters, making traditional phylogenetic analysis impossible. Analysis of variability among selected *Porphyra* species in restriction sites from PCR amplified small subunit rDNA is used to assess the reliability of morphological characters as indicators of evolutionary relationships. The implications of the analysis are discussed for the current subgeneric divisions within *Porphyra*.

PHYLOGENETIC RELATIONSHIPS AMONG THE NORTHWEST BROWN ALGAL ORDERS: A MOLECULAR APPROACH.

Ian H. Tan and Louis D. Druehl¹. Department of Biological Sciences, Simon Fraser University, Burnaby, B.C. V5A 1S6 Canada and ¹Bamfield Marine Station, Bamfield, B.C. V0R 1B0 Canada.

The phylogenetic relationships among the Northwest brown algal orders were re-examined by comparison of molecular sequence data. Partial cytoplasmic small-subunit ribosomal RNA gene sequences from thirteen brown algae representing the following brown algal orders, Chordariales, Desmarestiales, Dictyosiphonales, Dictyotales, Ectocarpales, Fucales, Scytosiphonales, Sphacelariales and Syringodermatales, were determined by means of polymerase chain reaction (PCR) and direct sequencing. The neighbor-joining phylogeny inference method (PHYLIP) was used to infer a phylogeny among these sequences and published sequences from a kelp (Laminariales), a xanthophyte and a diatom. The inferred molecular phylogeny supported a recent report that the Phaeophyceae is closer related to the Xanthophyceae than the Bacillariophyceae. In addition, the inferred phylogeny also separated the ten brown algal orders into two distinct clades; the orders Chordariales, Ectocarpales, Dictyosiphonales and Scytosiphonales formed a clade, whereas the orders Desmarestiales, Dictyotales, Fucales, Laminariales, Sphacelariales and Syringodermatales formed the sister clade. Further bootstrap analysis suggested that members in the former clade formed a monophyletic assemblage. However, certain branching orders within the overall inferred ordinal phylogeny remain unresolved. Nonetheless, results from this study provided valuable insights to the perplexing brown algal phylogeny. In addition, this study contributed additional data to the phylogenetic relationship study among the chromophyte classes.

*Not necessarily
Separate*

EFFECTS OF PETROLEUM PRODUCTS ON BULL KELP (*NEREOCYSTIS LUETKEANA* P. & R.).

Ronald M. Thom, Liam D. Antrim, William W. Gardiner, Valerie I. Cullinan, David K. Shreffler and Ray W. Bienert. Battelle/Marine Sciences Laboratory, 439 W. Sequim Bay Road, Sequim, WA 98382 USA.

The present study tested whether petroleum products, like those spilled in coastal waters of Washington State from the fishing vessel *Tenyo Maru*, could result in the loss of coloration and death of bull kelp as was observed in the field. The study investigated the effects of three petroleum products including: intermediate fuel oil (IFO), diesel fuel and Prudoe Bay crude oil; each product was tested unweathered and weathered. Following 4 and 24-hr exposures to the products, the plants were reintroduced to field conditions. Observations on the plants were made daily for seven days. In addition, controlled bioassays were performed to measure the effects of petroleum exposure on net photosynthetic rate (NP) and respiration rate (R), using light and dark bottle techniques. The experiments verified the susceptibility of *N. luetkeana* tissue to the damaging effects of direct exposure to several oil types. Four-hour exposures to weathered diesel and unweathered IFO, and 24-hour exposures to weathered and unweathered diesel and IFO resulted in moderate to severe damage (i.e., a clearly delineated bleached line accompanied by decay of the tissue) to the stipe, bulb and blade tissue. Petroleum significantly affected blade and stipe NP, R and NP:R ratios. Diesel treatments had a greater negative effect on NP than IFO. Based upon the studies, the relative ranking of petroleum-treatment effects to kelp are: weathered diesel>unweathered IFO>unweathered diesel>weathered IFO>unweathered crude>weathered crude. The present study provides the first evidence that can guide damage assessment of oil spills on bull kelp forest habitat.

A SYNOPSIS OF PHYTOPLANKTON MONITORING IN MONTEREY BAY:
THE *PSEUDONITZSCHIA* SPECIES AND DOMOIC ACID PRODUCTION

P.M. Walz, D.L. Garrison, M.W. Silver, M. Cattey,
W.M. Graham, and R. Tjeerdema.
Institute of Marine Sciences
University of California, Santa Cruz, Santa Cruz, CA 95064

Since first documented in September 1991, domoic acid and its production by species of the pennate diatom genus *Pseudonitzschia* have been the focus of regular sampling in northern Monterey Bay. The goal of these efforts has been to monitor phytoplankton populations and concurrent hydrographic conditions, to obtain field samples for domoic acid analysis by HPLC, and to confirm species identifications using Scanning Electron Microscopy. Plankton samples for species and toxin analysis have been collected monthly at four permanent stations. Physical (CTD), chemical (nutrients), and biological (Chl *a*) data have also been obtained during these cruises. Additional samples were collected more frequently from the Santa Cruz Municipal Pier and a nearshore station. Preliminary analysis indicates *Pseudonitzschia* blooms are found relatively close to shore. During conspicuous blooms, the three stations closest to shore exhibited highest cell counts. Additionally, it appears that *Pseudonitzschia* species can be prolific throughout the Autumn and Winter months in this coastal band along northern Monterey Bay. Complete analysis for domoic acid in samples collected thus far is now underway, as is SEM for species identifications. The information collected during this monitoring project may provide an early indication of conditions associated with *Pseudonitzschia* blooms and toxin production.

These studies have been funded by a grant from the National Sea Grant College Program, NOAA, U.S. Dept. of Commerce under grants 447610-22574-3 and 447610-22576-3 through the California Sea Grant College.

BATCH CULTURE OF *NITZSCHIA PUNGENS* FORMA *MULTISERIES* FOR STUDIES ON DOMOIC ACID RETENTION IN COMMERCIAL FISHERIES SPECIES.

J.N.C. (Ian) Whyte, Norma G. Ginther and Linda D. Townsend. Fisheries and Oceans Canada, Pacific Biological Station, Nanaimo, British Columbia, Canada, V9R 5K6.

Batch cultures of *N. pungens* f. *multiseries* (250-500 litres) have been grown, using continuous light, from concentrations of 1,500 cells/ml at log phase to 0.42M cells/ml at stationary phase of growth. Cell densities varied significantly with silicate concentrations in the media. Dry weight per cell ranged from 200 to 490 pg during the growth of the alga. Domoic acid production varied from 0.01 to 0.07 pg/cell but no major increase at the stationary phase of growth was observed; this may be a function of continuous irradiance. The fatty acid profiles of this alga contained high levels of 16:2n6 and 16:4n1 acids which are not common in the tissue of many commercial species and may prove useful indicators of active ingestion by the target species during toxin uptake studies.

A PC-BASED RELATIONAL DATABASE FOR THE UBC PHYCOLOGICAL HERBARIUM

Thomas B. Widdowson, Department of Botany, UBC
4635 West Saanich Road, Victoria, B.C. V8Z 3G7

The UBC Phycological Herbarium has been stored for some time on the local mainframe. With the help of Steve Lower of SFU, I have organized the approximately 50,000 records from Alaska, BC, Washington, Oregon, and California into lines. Fixed length fields have added at the left for doubt, taxon sorting code, locality sorting code, and month of collection. The accession number, the genus name, specific epithet, and subspecific epithet have been moved from the text to the next positions left. The rest of the original text follows. These lines are then sorted by accession number. In another file, the fixed length fields are taken and sorted by locality code, with another field added to define every collecting locality, no matter how close geographically. Reproductive, phenological, and ecological data are missing from most records, are of variable length, and in no systematic format. In a third file, the fixed length fields are taken and sorted by taxonomic code, and fixed length fields added for reproductive state/phenology, vertical zone, height in intertidal zone, depth in subtidal zone, upper limit of a range in the subtidal zone, lower limit of a range in the subtidal zone, wave exposure, abundance, and substrate/associated species. There is a great deal of redundancy in the system to aid browsing and proof reading. I can examine most relationships using a line editor such as "Qedit". I am developing 'user-friendly' software using "Knowledgeman".

EARLY WARNING MARINE BIOTOXIN MONITORING IN WASHINGTON STATE: SENTINEL MUSSEL CAGES AND PHYTOPLANKTON SAMPLING

Mary McCallum and Linda Hanson. Washington State Department of Health,
Office of Shellfish Programs, Airdustrial Park, Building 4, P. O. Box
47824, Olympia, Washington 98504-7824, USA

In Washington State, the State Department of Health, Office of Shellfish Programs is responsible for monitoring commercial and recreational shellfish for marine biotoxins potentially harmful to human health. In recent years, several costly recalls of commercial shellfish due to paralytic shellfish poisoning (PSP), episodes of PSP in previously unaffected areas and the detection of domoic acid in shellfish have initiated the development of an early warning component to the basic PSP monitoring program. Wire mesh cages are stocked with mussels (*Mytilus edulis* or *Mytilus californianus*) and suspended from docks or other shore-based structures to provide a readily accessible early indicator species for PSP and domoic acid. The cages are sampled biweekly from April through October or year around by state and local health department staff and citizen volunteers. Toxin levels between caged mussels and other molluscan shellfish species such as clams and oysters are compared when toxicity occurs and shellfish samples can be obtained. In addition, an 18-month pilot phytoplankton monitoring program is underway in 1993. Plankton net tows and microscopic examinations of collected material are currently conducted weekly in several shellfish growing areas.

