

Global Ecology and Oceanography of Harmful Algal Blooms



HABs and Eutrophication







7-10 March 2005 Baltimore, Maryland USA

Programme and Abstracts



EXEMPTING COMMITTEE ON OCCUME AND A

# **ORGANIZATION OF THE OPEN SCIENCE MEETING**

# **Convener**

Patricia Glibert, USA

# **Conference Steering Committee**

Donald Anderson, USA Edna Graneli, Sweden Mingjiang Zhou, China J. Icarus Allen, UK Michele Burford, Australia

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# Global Ecology and Oceanography of Harmful Algal Blooms

# OPEN SCIENCE MEETING ON HABS AND EUTROPHICATION BALTIMORE, MARYLAND, USA 7-10 MARCH 2005

# **PROGRAMME AND ABSTRACTS**

# THIS MEETING IS DEDICATED TO DR. JOHN HEISLER U.S. EPA OCEANS AND COASTAL PROTECTION DIVISION,

WHO LONG ENVISIONED AN INTERNATIONAL MEETING ON HABS AND EUTROPHICATION.

John's death in September 2004 is a loss to this community, scientifically and personally.

The GEOHAB Scientific Steering and the Conference Steering Committees are grateful for the support for this meeting provided by the Scientific Committee on Oceanic Research (SCOR) and the Intergovernmental Oceanographic Commission (IOC) of UNESCO. Financial support was provided by the National Oceanic and Atmospheric Administration - National Ocean Service, the U.S. National Science Foundation -Division of Ocean Sciences, University of Maryland Center for Environmental Science, U.S. National Office for Marine Biotoxins and Harmful Algal Blooms, Maryland Department of Natural Resources, Chesapeake Research Consortium, Gallaudet University, YSI Environmental and the Fordham Brewery.



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# AN INTRODUCTION TO THE MEETING

Welcome to the GEOHAB Open Science Meeting on HABs and Eutrophication.

GEOHAB, the Global Ecology and Oceanography of Harmful Algal Blooms Programme, sponsored by the Scientific Committee on Oceanic Research (SCOR) and the Intergovernmental Oceanographic Commission (IOC) of UNESCO, is an international program aimed at fostering and promoting cooperative research directed toward improving the prediction of harmful algal bloom (HAB) events.

HABs have been associated with fish and shellfish kills, human health impacts, and ecosystem damage throughout the world. Concurrent with escalating influences of human activities on coastal ecosystems, the environmental and economic impacts of HABs and consequent challenges for coastal zone management have increased in recent years. The relationship between HABs and the increasing nutrient enrichment of many of the world's coastal and estuarine environments is of particular concern. Increasing nutrient loading to coastal and enclosed or estuarine environments is a result of agricultural, aquacultural, industrial, and sewage effluents. The relationship between nutrient loading and alteration in nutrient supply ratios and many HABs is now recognized, but much remains to be understood. The overarching questions to be addressed re: to what extent has increased eutrophication influenced, and continues to influence, the occurrence of HABs and their harmful effects, and what are the mechanisms underlying this process?

This meeting, the third in a series of Open Science Meetings convened by GEOHAB, is designed to bring experts together from around the world to review the state of knowledge with respect to our understanding of the role of eutrophication in the proliferation of worldwide HABs, and take the initial steps in designing the next phase of research on comparative, eutrophic ecosystems and species that will be necessary to address this critical, global issue. **HABs in Eutrophied Systems** is designated as one of the **Core Research Projects** in the GEOHAB Science and Implementation Plans, which can be found at: www.geohab.info.

The GEOHAB Core Research Project on HABs in Eutrophied Systems must be comparative, interdisciplinary, and international. It will directly address the goal of GEOHAB of improved prediction of HABs by determining the ecological and oceanographic mechanisms underlying their population dynamics, integrating biological, chemical, and physical studies supported by enhanced observation and modelling techniques. The overall objective is to understand and quantify the critical processes underlying HAB population and community dynamics in eutrophied systems.

The purpose of this meeting is to obtain community input for the development of a detailed research plan for the GEOHAB Core Research Project on HABs in Eutrophied Systems. This plan will be drafted by the Coordinating Committee based on the full input of those assembled for this meeting, as well as the broader community unable to attend. As the plan develops, comments will be sought from meeting participants and will be augmented as additional planning occurs.

The GEOHAB Scientific Steering Committee and the Conference Steering Committee thank you for your participation in developing and implementing GEOHAB research.

We also hope you will find this book to be helpful, both as a reference during the Open Science Meeting and afterward. The list of participants includes all those who completed registration for the meeting before this book went to print on February 16, 2005. Similarly, the abstracts, both for speakers and the poster sessions, reflect the status of the program for the conference on that date. Changes to the program will be announced and posted at the conference and you are advised to look for these.

Enjoy your stay in Baltimore!

Grant Pitcher GEOHAB SSC Chair

Patricia Glibert Convener

# **Transportation**

Air travel—The closest airport to the meeting location is the Baltimore-Washington International (BWI) airport. Upon arrival at the airport, SuperShuttle service can be arranged (reservations are not required). Go to the lower level and follow signs to the SuperShuttle desk located between bag claims 6 and 7. The ticket counter is open between the hours of 6:00 AM and 2:00 AM. During other times please call 888-826-2700 to arrange service. The cost of the shuttle should be about \$16 one way and will take 30-60 minutes. Taxis can also be arranged at BWI Airport, for a higher fee. Return to the airport from the hotel can be arranged through the hotel front desk.

Driving—The Radisson Hotel is located at 5100 Falls Road in the Cross Keys development on the east side of I-83, between the exits for Northern Parkway and Cold Spring Lane. Please see www.radisson.com/baltimoremd/ for directions.

# <u>Hotel</u>

A block of rooms has been reserved at the Radisson at Cross Keys Hotel. Participants should reserve a room through a special Web site set up by the hotel: <u>http://www.radisson.com/geohab</u>.

# <u>Meals</u>

Coffee breaks, receptions, and a group dinner will be provided as part of the registration fee. Other meals will be the responsibility of participants. The Crossroads Restaurant in the Radisson Hotel is available for all meals. Donna's Restaurant in the same complex is available for lunches. Several restaurants are located within a few miles of the hotel and can be reached by private car or taxi. These are listed on the following page.

#### **Ambassador Dining Room**

3811 Canterbury Rd, Baltimore 21218 • 410-366-1484 Distance: ~2.01 mi \*

**Description:** CHARLES VILLAGE. This is Indian cuisine fit for a king or a queen. Solicitous service, a fabulously ornate dining room and a lovely outdoor patio supplement a perfectly executed menu of flavorful Indian favorites. Enjoy excellent chicken tikka masala with a rich tomato-based sauce, spicy grilled shrimp in limejuice and chili, or the exotic Bengali swordfish in a tangy-sweet mango sauce.

#### Cafe Hon

1002 W 36th St, Baltimore 21211-2415 • 410-243-1230 Distance: ~1.85 mi \*

**Description:** HAMPDEN. A casual cafe with a downhome feel, this restaurant was once a hardware store. It no longer sells hammers and socket sets, instead serving all-American comfort food in appealingly nostalgic atmosphere.

#### **Gertrude's**

10 Art Museum Dr Baltimore Museum Of Art, Baltimore 21218 • 410-889-3399 Distance: ~2.55 mi \*

**Description:** NORTH BALTIMORE. Gertrude's is a lovely cafe with a prime location inside the Baltimore Museum of Art. The name of the restaurant pays homage to both the chef's grandmother, who introduced the chef to the wonderful world of cooking, and Gertrude Stein, who introduced several important artists to the museum.

#### **Holy Frijoles!**

908 W 36th St, Baltimore 21211-2483 • 410-235-2326 Distance: ~1.87 mi \*

**Description:** HAMPDEN. Frijoles are beans in Spanish, and this charming little eatery has plenty of them, served in a number of tasty incarnations. The artsy, colorful dining room only has a dozen tables, but they're always full of folks enjoying great tacos, enchiladas and burritos. No alcohol is served; feel free to bring your own.

#### Jeannier's

105 West 39th St, Baltimore 21210 • 410-889-3303 Distance: ~1.89 mi \*

**Description:** NORTH BALTIMORE. Fine French cuisine in the Provençal style draws elegant crowds to this longstanding North Baltimore mainstay. The tranquil, romantic dining room, complete with classical art and gauzy lace curtains, makes this a great destination for special occasions. More casual groups may prefer the brasserie-style bar area. Reservations required.

#### Loco Hombre

413 W Cold Spring Ln, Baltimore 21210 • 410-889-2233 Distance: ~5.05 mi \*

**Description:** The name of the place means "Crazy Man," and if you pass it by, you're crazy, man. This fantastic local place bursts with color, from decor to cuisine. You'll find a host of classic dishes (including basic tacos and burritos), along with the eatery's own take on Mexican cuisine.

Many other restaurant options are available at the Baltimore Inner Harbor, Little Italy, and other areas of the city. A map of Baltimore will be provided with meeting materials.

# **PROGRAMME SUMMARY**

Sunday 6 March	Monday 7 March	Tuesday 8 March	Wednesday 9 March	Thursday 10 March	Friday 11 March
	Registration & Poster Set-up 8:00–9:00 (Mezzanine)	Registration 8:00-9:00 (Mezzanine)	Registration & Poster Set-up 8:00–8:30 (Mezzanine)	Registration 8:00-9:00 (Mezzanine)	
	9:00 Opening Remarks 9:45 SESSION 1 (General Trends) (White Oak Room)	8:30 SESSION 3 (GEOHAB Programme) (White Oak Room)	8:30 SESSION 5 (Nutrients and interacting factors) (White Oak Room)	8:30 SESSION 7 ( <b>Modeling</b> ) (White Oak Room)	
	10:25 COFFEE	9:55 COFFEE	9:50 COFFEE	9:50 COFFEE	
	11:00 SESSION 1 (cont.)	10:20 SESSION 4 (International Programmes)	10:20 SESSION 5 (cont.) 11:20 SESSION 6 (New challenges, methodologies)	10:10 SESSION 7 (cont.) 11:30 SESSION 8 – (Implementation)	Conference Steering
	12:00 LUNCH	12:20 LUNCH	12:30 LUNCH	12:45 LUNCH	Committee Meets in
	13:30 SESSION 2 (Physiology, Ecology)	14:00 SESSION 4 (cont.)	14:00 SESSION 6 (cont.)	14:15 SESSION 8 (cont.)	Closed Session
	14:50 COFFEE	15:20 COFFEE	15:00 COFFEE	16:00 COFFEE	
15:00-17:00 Registration ( <i>Mezzanine</i> )	15:15 SESSION 2 (cont.)	15:45 SESSION 4 (cont.)	15:40 SESSION 6 (cont.)	16:15 SESSION 8 (cont.)	
17:30-18:30 Welcome Drinks ( <i>Lobby Bar</i> )	17:30-19:30 Posters Session One (Mezzanine)	18:00 Buses depart 18:30 Group Dinner	16:40-18:30 Poster Session Two (Mezzanine)	17:00 Adjourn	

# **Oral Presentations (White Oak Room)**

- Speakers will be expected to adhere to the allocated times for their talks. The chairs of the plenary sessions will be strict about the timing, in order to keep the conference running smoothly. Please allow time for questions in your allocated time.
- All oral presentations should be loaded onto the meeting computer (PC only) before the session starts. Please see Judy Kleindinst for help with this. Please bring your presentation on a Memory stick or CD.

# **Poster sessions (Mezzanine)**

- Posters are to be 3' x 4' (1 m x 1.3 m) and may be hung vertically or horizontally.
- Posters for Poster Session One are to be hung by Monday coffee break, and removed by Tuesday evening
- Posters for Poster Session Two are to be hung by Wednesday coffee break and removed by Thursday evening
- All poster authors are expected to stand by their posters during their designated poster session. Posters may also be viewed during all coffee and lunch breaks. Supplies will be provided for mounting the posters.

# **Internet Service**

- High speed internet service is available in the hotel for a small charge. Wireless hot spot areas are also available.
- No internet service is available in the meeting rooms.

# Sunday, 6 March 2005

15:00-17:00	Registration desk open (Mezzanine)
17:30-18:30	Welcome drinks ( <i>Lobby</i> )

# Monday, 7 March 2005

8:00-9:00	Registration desk open (Mezzanine)
9:00-9:15	WELCOME AND OPENING LECTURE Welcome and opening remarks - Patricia Glibert (USA) (White Oak Room)
9:15-9:45	Harmful algal blooms in the context of major ocean initiatives - Richard Spinrad (USA)
SESSION 1	<b>TRENDS in EUTROPHICATION and HABS</b> (Session Chair: <i>Patricia Glibert</i> )
9:45	Nitrogen pollution: Sources, trends, and effects globally and regionally – <b>Robert Howarth (USA)</b>
10:25-11:00	COFFEE BREAK
11:00-11:30	National and global trends in HABs – Donald Anderson (USA)
11:30-12:00	Multidecadal changes in the diatom: flagellate ratio and Si:N and Si:P ratios in Narragansett Bay, and influence of SI:N supply ratios on diatom species competition – <b>Theodore Smayda (USA)</b> and D. Borkman
12:00-13:30	LUNCH
SESSION 2	PHYSIOLOGY and ECOLOGY of HABS with RESPECT to NUTRIENTS (Session Chairs: <i>Edna Granéli and Cindy Heil</i> )
13:30-14:10	The role of nutrient conditions on toxicity, allelopathy and mixotrophy in HABs – Edna Granéli (Sweden)
14:10-14:30	Nitrogen uptake by the toxigenic diatom <i>Pseudo-nitzschia australis</i> – William Cochlan (USA), J. Herndon, N.C. Ladizinsky and R.M. Kudela

- 14:30-14:50 Urea-ammoninium-nitrate interactions in Thau lagoon (Southern France): relationships with *Alexandrium catenella* blooms – **Yves Collos (France)**, A. Vaquer, M. Laabir, E. Abadie, T. Laugier, A. Pastoureaud
- 14:50-15:15 COFFEE BREAK
- 15:15-15:35 A role for anthropogenically derived nitrogen in the formation of harmful algal blooms along the U.S. west coast **Raphael Kudela (USA)**, M. Armstrong, W. Cochlan, and J. Herndon
- 15:35-15:55 Nitrogen preference of the fish-killing flagellate *Chattonella cf. verruculosa* Carmelo Tomas (USA)
- 15:55-16:15 Intraspecific variability in the nutritional ecology of harmful algae JoAnn Burkholder (USA)
- 16:15-16:35 The effects of macro- and micro-nutrient limitation on karlotoxin production by *Karlodinium micrum* strains **Jason Adolf (USA)**, T. Bachvaroff, G.F. Reidel, A. R. Place
- 16:35-17:05 Open discussion
- 17:15-19:15 Poster Session One: Long- term Trends and Physiology (Mezzanine) (Hosted by the University of Maryland Center for Environmental Science, and the Maryland Department of Natural Resources)

# Tuesday, 8 March 2005

8:00-9:00	Registration desk open (Mezzanine)
SESSION 3	THE GEOHABPROGRAMMEandOTHERINITIATIVES (Session Chair: Donald Anderson)
8:30-8:45	Introduction to the GEOHAB Programme – Grant Pitcher (S. Africa)
8:45-9:05	Introduction to IOC and SCOR – Henrik Enevoldsen (Denmark) and Ed Urban (USA)
9:05-9:35	NOAA extramural HAB research: Present and future – <b>Quay Dortch (USA)</b> , S. Banahan, M. Suddleson, Juli Trtanj, Leon M. Cammen, and Teri Rowles
9:35-9:55	Implications of the U.S. Ocean Commission reports – Donald Boesch (USA)
9:55-10:20	COFFEE BREAK

### SESSION 4 COMPARATIVE STUDIES and INTERNATIONAL PROGRAMMES on HABS in EUTROPHIC AREAS (Session Chairs: Mingilang They and Lars Edlar)

(Session Chairs: Mingjiang Zhou and Lars Edler)

- 10:20-11:00Which is the trigger factor to the outbreak of large scaleProrocentrum blooms in the East China Sea? -Mingjiang Zhou (China)
- 11:00-11:40 Do the coastal eutrophication and warming cause widespread and persistent *Cochlodinium polykroides* blooms in Korean waters? **HakGyoon Kim (Korea)**, C-K. Lee, W-A. Lim, S-Y. Kim, and H-G. Jin
- 11:40-12:20 Influence of monsoons and oceanographic processes on red tides in Hong Kong waters **Kedong Yin (Hong Kong China)**
- 12:20-14:00 LUNCH
- 14:00-14:40 Harmful algae and eutrophication in the Baltic Sea area Lars Edler (Sweden)
- 14:40-15:00 Oceanographic and environmental assessment of Kuwait's waters in relevance to algal blooms Faiza Al-Yamani, W.A. Ismail, **Kholood .S. Al-Rifaie (Kuwait)**, and A. Lennox
- 15:00-15:20 HABs in Western Australia: Expressions of eutrophication in a southern climate Malcolm Robb (Australia), T. Reitsema, W. Hosja, and A. Begum
- 15:20-15:45 COFFEE BREAK
- 15:45-16:05 An integrated approach to predicting harmful algal blooms: phytoplankton physiology, nutrient dynamics and their application in an ecosystem model **Paul Armstrong (Australia)**, P. A. Thompson, C. J. S. Bolch, S. I. Blackburn, J. P. Parslow, M. Herzfeld and K. Wild-Allen
- 16:05-16:25 Comparative analysis of the relationships between nutrient cycling and phytoplankton community composition in two eutrophied subtropical estuaries: Florida Bay, USA, and Moreton Bay, Australia **Cynthia Heil (USA),** P.M. Glibert, J. O'Neil, W.C. Dennison, D. Hollander, J. Greenwood, M. O'Donohue, S. Costanzo, M. Revilla, J. Alexander, A. Hoare and S. Murasko
- 16:25-16:45 Linkages between land-based nutrient discharges and harmful macroalgal blooms: comparative studies on coral reefs of southeast Florida and Jamaica **Brian Lapointe (USA)**, B. Bedford, P.J. Barile, C. Hanson, and L. Getten
- 16:45-17:15 Open discussion
- 18:00 Buses leave hotel
- 18:30-20:30 GROUP DINNER Phillips Restaurant, Baltimore waterfront
- 21:00 Buses return to hotel

# Wednesday, 9 March 2005

8:00-9:00	Registration desk open (Mezzanine)
SESSION 5	MACRONUTRIENTINTERACTIONSwithOTHERFACTORSCONTROLLINGHABs(Session Chair: Ted Smayda)
8:30-8:50	How does eutrophication affect the role of grazers in harmful algal bloom dynamics? – Edward Buskey (USA)
8:50-9:10	Freshwater flow and nutrients: Effects on top-down control of bloom-forming dinoflagellates? – <b>Diane Stoecker (USA)</b> , M.L. Reaugh, A. E. Thessen, D. E. Gustafson, M. R. Roman and W. C. Boicourt
9:10-9:30	A conceptual model for ecosystem disruptive algal blooms: The interactive roles of eutrophication, algal toxicity, and limitation by nutrients and light – <b>William Sunda (USA)</b> and R. Hardison
9:30-9:50	Abiotic and biotic factors controlling a nutrient driven dinoflagelllate bloom and likely responses to increased eutrophication – <b>R. Wayne Litaker</b> (USA) and P.A. Tester
9:50-10:20	COFFEE BREAK
10:20-10:40	Brackish stormwater detention ponds as promoters of HABs and eutrophication along the South Carolina coast – Alan Lewitus (USA), M.K. Burke, L. J. Mason, K. N. Bunker, S. R. Drescher, and W. H. J. Strosnider
10:40-11:00	Iron induced development pathway of HABs community and its consequence on mitigation of eutrophication – <b>Jun Sun (China)</b> , Y. Feng, and P. Sun
11:00-11:20	The synergy of iron, copper and the toxicity of diatoms – M. Wells, Charles G. Trick (USA), W. P. Cochlan, P. Hughes, and N. C. Ladizinsky
SESSION 6	<b>NEW CHALLENGES and METHODOLOGIES</b> (Session Chair: <i>Marc Suddleson</i> )
11:20-11:50	Implementing the coastal module of the Global Ocean Observing System (GOOS): Toward rapid detection and timely predictions of harmful algal blooms – <b>Thomas Malone (USA)</b>
11:50-12:30	New approaches and technologies for observing harmful algal blooms – Marcel Babin (France)
12:30-14:00	LUNCH

## **SESSION 6 CONT'D** (Session Chair: *Marcel Babin*)

- 14:00-14:20 Application of the environmental sample processor (ESP) for remote detection of harmful algae and toxins they produce Chris Scholin and **Greg Doucette (USA)**
- 14:20-14:40 Autonomous nutrient monitoring and water sampling as tools for studying HABs: Progress and prospects – Lou Codispoti (USA), V. Kelly, P. Glibert, and J. Alexander
- 14:40-15:00 New technologies for monitoring and assessing harmful algal blooms and water quality in Chesapeake Bay, Maryland Christopher Heyer (USA), T. M.Trice, P. J. Tango, B.Michael, L.Codispoti, V. Kelly
- 15:00-15:30 COFFEE BREAK
- 15:30-15:50 Diagnostic indicators of HAB nutritional physiology Sonya Dyhrman (USA)
- 15:50-16:10 New approaches to understanding the role of dissolved organic matter in HAB dynamics **Sybil Seitzinger (USA)**, P. M. Glibert, J.P. Simjouw, and R. Sipler
- 16:10-16:40 Open discussion
- 16:45-18:45 Poster Session Two: Interaction of Other Factors, New methods, Monitoring, and Modeling (Mezzanine) (Hosted by the Chesapeake Research Consortium, YSI Environmental and the Fordham Brewing Company)

# Thursday, 10 March 2005

8:00- 9:00	Registration desk open (Mezzanine)
SESSION 7	<b>MODELING of NUTRIENTS and HABS</b> (Session Chairs: <i>Icarus Allen and Kevin Flynn</i> )
8:30-9:10	Eutrophication and HAB models for the NW European continental shelf – J. Icarus Allen (UK), F. Gilbert, J. Holt, M. Holt, R. Proctor, J. Siddorn
9:10- 9:50	Garbage in, Garbage out? – Problems in experimental design and modeling of HAB ecology – Kevin Flynn (Wales)
9:50-10:10	COFFEE BREAK

10:10-10:30	Assessing the validation of a preliminary <i>Karlodinium micrum</i> nowcast model system in Chesapeake Bay and its tributaries: A framework for HAB nowcasts and forecasts – <b>Peter Tango (USA)</b> , C. W. Brown, T.F. Gross, D. L. Ramers, R. R. Hood and B. D. Michael
10:30- 10:50	Modeling <i>Pfiesteria</i> life cycle attributes and population dynamics – <b>Raleigh Hood (USA),</b> X. Zhang and J. T. Anderson
10:50-11:15	Open discussion
<b>SESSION 8</b>	GEOHAB IMPLEMENTATION (Session Chair: Grant Pitcher)
11:15-11:30	Charge to Working Groups – Patricia Glibert and Grant Pitcher
11:30-12:45	First break-out groups meet (Rooms to be announced)
12:45-14:15	LUNCH
14:15-14:45	Reports of first break-out groups (White Oak Room)
14:45-16:00	Second break-out groups meet (Rooms to be announced)
16:00-16:15	COFFEE BREAK
16:15-16:45	Reports of second break-out groups (White Oak Room)
16:45	Final wrap-up
17:00	Adjourn

# Friday, 11 March 2005

8:30	Steering Committee meets to outline draft meeting report for GEOHAB
	(Room to be announced)

#### Poster Session One – Monday, 7 March 2005

- Eutrophication and harmful algal blooms in the Swan River estuary, Western Australia - <u>J. John</u>
- 2. The research of the eutrophication status of East China Sea X.-L. Wang, <u>X.-Y. Shi</u> and C.-S. Zhang
- Coastal nutrification following the passage of Hurricane Charley and its relation to a subsequent *Karenia brevis* bloom on the West Florida Shelf – <u>M. B. Neely</u>, C. A. Heil and G. A. Vargo
- The effect of nutrient concentration at different growth stages on hemolytic ability of three clones of the ichthyotoxic flagellate *Prymensium parvum* from blooms in the United States –<u>M. Clouse</u> and C. Tomas
- 5. Strain variation in *Karlodinium micrum* toxin production <u>T. Bachvaroff</u>, J. E. Adolf and A. R. Place
- 6. Fatty acids and growth in the heterotrophic dinoflagellates *Pfiesteria* spp. and PLO's

   <u>L. W. Haas</u>, V. Foster, L. Ott, W.K. Vogelbein, K. S. Reece, J. D. Shields and P. Mason
- The influence of dissolved copper on the production of domoic acid by toxigenic species of *Pseudo-nitzschia* in Monterey Bay, California – N. C. Ladizinsky, G. J. Smith, K. H. Coale and <u>W. P. Cochlan</u>
- Toxin levels in the benthic cyanobacterium Lyngbya majuscula in relation to tissue nutrient content and bloom intensity – <u>J.</u> <u>M. O'Neil</u>, J. M., S. Albert, N. Osborne and G. Shaw
- The potential role of increased nutrient inputs to higher incidences of ciguatera in Hawaii – <u>M. L. Parsons</u>
- Nutrient regulation of toxin production: Comparison of hemolytic activity of Amphidinium carterae and Amphidinium klebsii – <u>L. A. Zimmermann</u> and C. R. Tomas

- Nitrate uptake kinetics of the toxic dinoflagellate Alexandrium tamarense in response to nitrate supply mode – <u>S. C. Y.</u> <u>Leong</u>, M. Maekawa and S. Taguchi
- Bioavailability of dissolved organic phosphorus compounds to typical harmful dinoflagellate *Prorocentrum donghaiense* Lu – <u>B. Huang</u>, L. Ou, H. Hong, H. Luo and D. Wang
- Dissolved organic matter concentration and characteristics during Aureococcus anophagefferens blooms in 2002 and 2003: A comparison – <u>J. -P.Simjouw</u>, E. C. Minor and M. R. Mulholland
- 14. The role of natural DOM sources in Prorocentrum minimum growth dynamics
   <u>R. Sipler</u>, S. P. Seitzinger and P. M. Glibert
- Urease kinetics of several harmful algal species from the Chesapeake Bay, USA – <u>C.M. Solomon</u> and P. M. Glibert
- A comparison of nutrient effects on the growth of *Chattonella subsalsa* and *Heterosigma akashiwo* (Raphodophyceae) isolated from the Inland Bays, Delaware (U.S.A.) *Y. Zhang and D. A. Hutchins*
- 17. The assessment of brown tide blooms caused by the alga, Aureococcus anophagefferens and related environmental factors in coastal waters of New Jersey (2000-2002) <u>M.</u> <u>Downes Gastrich</u>, R. Lathrop, S. Haag, M. P. Weinstein, M. Danko, D. A. Caron and R. Schaffner
- Harmful algae in Suffolk County (N.Y., USA) Estuaries – a 30 year History – <u>R. Nuzzi</u>
- Interannual variability of brown tide, *Aureococcus anophagefferens*, blooms in the Maryland Coastal Bays – <u>C. Wazniak</u>, *M. Hall, P. Tango, and B. Sturgis*
- Effect of N:P supply ratio on biochemical composition and toxicity in dinoflagellate Alexandrium tamarense <u>A. Murata</u>, S. C. Y. Leong, Y. Nagashima and S. Taguchi

#### Poster Session Two – Wednesday, 9 March 2005

- 1. Top down control and demise of a nutrient driven dinoflagellate bloom <u>P. A. Tester</u> and R. W. Litaker
- Modelling the contribution of prey deselection in the formation of harmful algal blooms <u>A. Mitra</u> and K. J. Flynn
- Raphidophyte systematics and rapid identification: Sequence analyses and real time PCR assays – <u>H. A. Bowers</u>, C. Tomas, J. W. Kempton, S. Goto, A. J. Lewitus and D. W. Oldach.
- 4. Geographic distribution of *Pfiesteria* spp. and environmental factors *H. Zhang and* <u>*S. Lin*</u>
- Improved accuracy of quantitative real-time PCR of HAB species in environmental water samples using an exogenous DNA internal standard – <u>K. J. Coyne</u>, S. M. Handy, E. Demir, K. J. Portune, Y. Zhang, M. A. Doblin, D. A. Hutchins and S. C. Cary
- 6. Harmful phytoplankton indicator species applied to eutrophication assessments of Scottish coastal waters supporting aquaculture <u>M. J. Gubbins</u>, P. J. Sammes and I. M. Davies
- Monitoring toxic phytoplankton and shellfish in support of eutrophication assessments for Scottish coastal waters – <u>M. J. Gubbins</u>, E. A. Smith, M. Grieve and E. Bresnan
- History of HAB monitoring in Maryland tidewaters: Monitoring, response, nowcasting and forecasting – <u>P. J. Tango</u>, B. Michael, D. Goshorn, R. Magnien, C. Heyer, T. M. Trice, W. Butler, C. Wazniak, R. Karrh, S. Bowen, R. Lacouture, H. Bowers, D. Oldach, C. Luckett, C. Poukish, D. Matuzsak, J. Ryan, H. Lynch, C. Brown, R. Hood, T. Gross and D. Ramers
- 9. An autonomous urea monitor for studying HABs – <u>V. Kelly</u>, L. A. Codispoti, P. Glibert and J. Alexander

- Applications of an *in situ* water quality monitoring platform (MARVIN) for HAB research: A comparison of data collected in the St. Johns and Caloosahatchee River systems in Florida – <u>J. Rueter</u>, M. B. Neely, B. Bendis, R. Pigg, K. Steidinger and C. Heil
- Relationships between nitrogen loading and concentrations of nitrogen and chlorophyll in coastal embayments – <u>E. H. Dettmann</u>, L. B. Mason, A. Erhunse and K. M. Henry
- Modeled Karenia brevis bloom initiation and subsequent accumulation in the vicinity of a coastal nutrient front – <u>G. S. Janowitz</u> and D. Kamykowski
- Ecosystem modelling of the NW European shelf seas towards the forecasting of harmful algal blooms – <u>J. R. Siddorn</u>, J. I. Allen and M. Holt
- Environmental and behavioral influences on Karenia brevis' nitrate uptake: A bloom initiation scenario – <u>G. Sinclair</u>, D. Kamykowski, E. Milligan and B. Schaeffer
- A behaving drifter for simulating transport of mobile HAB organisms in nature – T. G. <u>Wolcott</u>, D. Kamykowski and G. Janowitz
- 16. Potential roles of Prorocentrum minimum to Chesapeake Bay dissolved oxygen and oyster dynamics – E. Brownlee, S. Sellner and <u>K. G. Sellner</u>
- 17. Volunteer HAB monitoring provides a "First Watch" for resource managers and researchers in the Delaware Inland Bays, U.S.A. <u>E. Whereat</u> and M. Farestad
- Problems with ballast water exchange as a means of controlling movement of harmful algal species throughout the world. <u>C. E.</u> <u>Orano-Dawson</u>, R. Dawson and D. A. Wright
- 19. Ballast water management and climate change in the coastline of Nigeria [Paranomic View] - L. E. Akeh, E. Udoeka, <u>A. O. Ediang</u> and A. A. Ediang

# **ABSTRACTS**

## (ALPHABETICAL BY FIRST AUTHOR)

# The effects of macro- and micro-nutrient limitation on karlotoxin production by *Karlodinium micrum* strains

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Karlodinium micrum has been associated with fish mortalities in coastal environments where along the eastern U.S. seaboard and other parts of the world. Our observations of field and laboratory samples of K. micrum show a nearly 10-fold range in cellular karlotoxin production, as well as variability in structural forms of karlotoxin produced by strains from different geographical regions. This raises the question, 'What causes variability of cellular toxicity?'. We are addressing this question by examining both genotypic (*i.e.* strains, see Bachvaroff abstract) and phenotypic (*i.e.* nutrients, light, prey availability) effects on toxin production. Here, we present the effects of nutrient limitation on karlotoxin production. A 3 week experiment with K. *micrum* strain 1974, isolated from Chesapeake Bay, showed dependence on > 1 nM selenite (SeO<sub>3</sub>) for growth. SeO<sub>3</sub>-deprived cells remained in cultures at low concentrations ( $10^3$  ml<sup>-1</sup>) and had cellular levels of toxicity > 10-fold that of SeO<sub>3</sub>-replete cells that grew to concentrations of 10<sup>5</sup> ml<sup>-1</sup>. This result is ecologically significant considering that total Se in Chesapeake Bay averages 1 - 2 nM, only 10% of which is typically found as SeO<sub>3</sub>. Further experiments are underway to examine the effects of Se, N, and P limitation on karlotoxin production by various strains of K. micrum. We hypothesize that conditions that limit cell division will result in increased cellular toxicity. With  $\geq 20$  strains of *Karlodinium micrum* in culture, we have the opportunity to test this hypothesis on both inter- and intra-strain basis.

### ORAL

### Ballast water management and climate change in the coastline of Nigeria [Paranomic View]

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One of the four greatest threats to the world ocean currently the most pressing marine environmental issue is the introduction of harmful aquatic organisms and pathogens into new marine environment via ships ballast water. At least one foreign marine species is introduced into a new environment every nine weeks. Ballast water transfer of harmful organisms is an international problem needing international solution. In Nigeria, especially the Niger Delta area [coastline] which include Delta, Edo, Lagos, Akwa Ibom, Cross River, Rivers, Bayelsa States. The impacts of climate change events vary, the most devastating being the loss of life. Many of the impacts are long-lasting and complex especially when there is a rapid changes in climate will most likely alter the composition of ecosystem with some species benefiting while others unable to migrate or adapt fast enough may become extinct. The paper discusses the human health ecological and economic impacts can be severe, economic losses due to ballast water introduction in Nigeria coastline is currently running into billions of Nairas also the numbers of ships entering into Nigeria from 1980 to present is mention.

The paper conclude by the role ballast water can play especially in influencing pollution, introduction of alien marine life are threathen and have added to the list of the endangered species, specified proposal and available information and know how to provided through modern climate services. Also to provide forum policy makers in maritime, shipping association and water treatment specialists.

## **POSTER - Session 2, Poster 19**

# Eutrophication and HAB models for the NW European continental shelf

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The state of the art POLCOMS-ERSEM model system has been applied to the North West European Continental Shelf on a 6km grid. Two sets of simulations have been made. Firstly a hindcast of the southern North Sea for 1988/89 was made because this is a particularly data rich period to evaluate the performance of the ecosystem model. Secondly a pre-operational hindcast with coupling to ocean (FOAM) and meteorological forecast models was made to assess the forecast potential of such systems.

We will discuss model validation and assessment of model performance. In doing so we shall demonstrate how the relative influence of hydrodynamic and biological processes changes spatially and seasonally.

Additionally we will demonstrate our capability to provide information products of the eutrophication and HAB status in European waters. Typical indicators which are tackled by model based monitoring systems include: bottom oxygen concentrations, zoobenthos, oxygen consumption, nutrient concentrations and ratios, nutrient transports from different sources to targeted areas, plankton concentration and total or new primary production, and peak production of different algal groups. From these Indicators more simple management information in terms of maps can be produced to indicate, for instance, the environmental condition in different areas

(being in very good, good, fair or bad) with respect to eutrophication, and eventually to see possible improvements due to various management actions.

# **ORAL-INVITED**

# Oceanographic and environmental assessment of Kuwait's waters in relevance to algal blooms

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Globally frequency of the incidence of red tides has been on the increase. Some of the algal bloom incidences are attributed to urban pollution and eutrophication. This is true in the waters off Kuwait where 21 potentially harmful algal species exist. Because of the economic impact, i.e., loss of revenue, and societal impact, a study was conducted to investigate the red tides in Kuwait's waters. Objectives of this project include quantitative studies of phytoplankton in these waters including areas in close proximity to sewage outfalls and testing the hypothesis that eutrophication is initiating red tides in Kuwait Bay. Phytoplankton biomass at the stations off the sewage outfalls was higher than the biomass at the station away from the outfall. The presence of a variety of phytoplankton in bloom densities, particularly the dinoflagellates implicated in toxigenic episodes elsewhere is a matter of concern and demonstrated the existence of a potential for development of toxigenic algal blooms off this coast due to eutrophication. Inventory of red tide incidences in the Arabian Gulf, Gulf of Oman and Arabian Sea is included.

## ORAL

### National and global trends in harmful algal blooms

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Harmful algal blooms (HABs) are common along the coasts of countries throughout the world. The impacts from these aquatic disasters are substantial, affecting public health, fisheries resources (both wild and farmed), local economies, tourism, ecosystem health, and coastal aesthetics. HAB problems include five shellfish poisoning syndromes termed paralytic, diarrhetic, neurotoxic, azaspiracid, and amnesic shellfish poisoning (PSP, DSP, NSP, AZP, and ASP), ciguatera fish poisoning (CFP), farmed and wild fish kills, destructive brown tides, macroalgal (seaweed) overgrowth, and a poorly defined syndrome of human health problems and fish mortality caused by dinoflagellates in the genus *Pfiesteria*. Many believe that HAB phenomena have been increasing in frequency, areal coverage, and diversity over the last several

decades, but this is a difficult assertion to prove given the diversity of organisms, impacts, and habitats. Few would argue, however, that there are now more algal toxins, more toxic algal species, more fisheries resources affected, larger areas affected, and higher economic losses. Reasons for this expansion are many, and include natural species dispersal via storms or currents, human assisted dispersal (e.g., via ballast water discharge), better detection as a result of increased monitoring and better analytical techniques for toxins, and enhancement of the bloom populations due to nutrients supplied by sewage, agricultural runoff, and other pollution sources. This latter mechanism applies to some HABs, but certainly not to all. This talk will review the nature of the HAB problems in the US and the world, the trends in their occurrence in different locations, and the lines of evidence linking eutrophication to blooms in certain locations.

# ORAL-INVITED

# An integrated approach to predicting harmful algal blooms: Phytoplankton physiology, nutrient dynamics and their application in an ecosystem model

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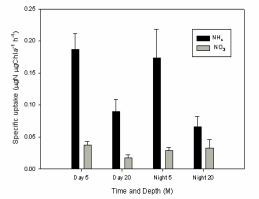
<sup>1</sup> CSIRO Marine Research and Aquafin CRC, PO Box 1538, Hobart Tasmania, Australia.

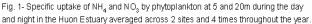
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Salmon farming is a growing industry in south-east Tasmania presently worth \$170M (AUD) per year. Both Industry and Government regulators are alert to the potential for overloading the systems with nutrients, and in one farming region, the Huon Estuary, further increases of salmon production have been capped to ensure nutrient release does not cause unacceptable environmental outcomes. Major research investigations include: the Huon Estuary Study (HES) from 1996-1999, which showed that phytoplankton growth was primarily limited by nitrogen derived from all sources and currently the Aquafin Co-operative Research Centre (Aquafin CRC) (2002-2007) which has extended the research into the D'Entrecasteaux Channel. The Aquafin CRC is addressing the link between phytoplankton blooms and nutrients from all sources in the

estuary. In particular we are interested in understanding the bloom dynamics of *Gymnodinium catenatum* a toxic dinoflagellate that causes periodic closures of shellfish farms in south-east Tasmania.

A multidisciplinary approach has been adopted which includes experiments in both the field and laboratory. Results from these activities are then used to parameterize an ecosystem-level biogeochemical model as an aid to environmental management of the Huon Estuary and D'Entrecasteaux Channel.





Uptake of NO<sub>3</sub>, NH<sub>4</sub> and urea were measured at two depths during the day and night in both the Huon Estuary and at a site in the D'Entrecasteaux Channel where *G. catenatum* was blooming. In the Huon Estuary, NH<sub>4</sub> was taken up preferentially over NO<sub>3</sub> at both depths, during the day and night (Fig. 1). Preliminary data indicate that urea is also an important nitrogen source in the Huon.

In the laboratory the effects of 72 combinations of temperature and irradiance were investigated, which showed that the greatest  $\mu_{max}$  (0.295 d<sup>-1</sup>) for *G. catenatum* occurred at a temperature of 23.7°C and no growth was observed at 13.1°C regardless of irradiance. Also growth on NH<sub>4</sub> (0.123 d<sup>-1</sup>) as a sole nitrogen source was measured and found not to be significantly different (P = 0.161) from growth solely on NO<sub>3</sub>.

ORAL

# New approaches and technologies for observing harmful algal blooms

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New technologies for autonomous observation of phytoplankton and their environment have emerged during the last decade from optical imaging, molecular biology, chemistry, acoustics and marine optics. Optical and chemical sensors are, for instance, increasingly deployed from various platforms (e.g., *in situ* profilers, autonomous underwater vehicles, moorings) to derive quantitative and qualitative information about the pelagic environment (e.g., the light field, concentrations of colored dissolved organic matter and nutrients), and about phytoplankton themselves (spatial distribution, species composition, cell size, optical and photosynthetic properties).

Populations of HAB species develop in different environments (e.g., bays, fjords, open ocean), and at various temporal and spatial scales (e.g., patches, large blooms, thin layers). Some form high-biomass blooms while others do not. Observations can be used for different goals, for instance early-warning and the study of bloom dynamics. Because many sensors operate only in specific conditions (e.g., cloudless for ocean color remote sensing) and at specific spatial and temporal resolution, a combination of platforms and sensors is necessary. In other words, an observation strategy must be developed for each HAB species that makes the best use of the available platforms (e.g., satellites, mooring, profiling systems) and sensors to best serve identified scientific and management goals (e.g., early-warning, description of the environment, time-series). In this talk, new technologies for autonomous observation of phytoplankton and their environment, and strategies for observation of specific HABs are discussed.

# **ORAL-INVITED**

#### Strain variation in Karlodinium micrum toxin production

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The bloom-forming dinoflagellate *Karlodinium micrum* has been associated with fish kill events along the Atlantic coast of the United States. *Karlodinium* appears to have a worldwide distribution in estuarine and marine environments yet does not always cause fishkills. To better understand the causes and consequences of variable cell toxicity in *Karlodinium micrum* we have undertaken a survey of strain to strain variation in phenotype and genotype. Here we report on phenotypic differences in toxin type and abundance using *Karlodinium* strains from Chesapeake Bay, the U.S. Atlantic coast, as well as from New Zealand using liquid chromatography/mass spectroscopy. To better understand genetic differences between strains we have used variation at microsatellite loci to genetically characterize strains and are attempting to correlate these genetic differences with toxin type and abundance in Chesapeake Bay waters will be combined with environmental parameters to predict potential harmful events.

### **POSTER – Session 1, Poster 5**

### Implications of the U.S. Ocean Commission reports

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Two separate commissions have recently made recommendations concerning United States ocean policy that will undoubtedly have implications for the conduct and use of marine science in this country. The Pew Oceans Commission (POC) was sponsored by a private foundation and the U.S. Commission on Ocean Policy (USCOP) was mandated by Congress and appointed by the President. Despite the different sponsorship, orientation, and membership of the two commissions, they came to strikingly similar conclusions. Namely, both commissions concluded that ocean and coastal environments and resources of the U.S. are in peril and that a new approach is needed to sustainably manage them in the future, one that is ecosystem-based and founded on the best science. Very significantly, both commissions address the importance of land-based sources of pollution, particularly by nutrients, and the growing problems of eutrophication and harmful algal blooms in coastal waters. The President and Congress are already taking steps to respond to the recommendations of the USCOP. These responses will likely include governmental reorganization and coordination, new legislation, and increased scientific investment. Implication of the commission recommendations that should help inform our agenda as scientists include: requirement for greater interdisciplinary integration to support

ecosystem-based management; improvements in ecological observations and forecasting; linkage with society, particularly human health; and coupling of phenomena, such as harmful algal blooms, with management solutions.

# **ORAL - INVITED**

# Raphidophyte systematics and rapid identification: Sequence analyses and real time PCR assays

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Species within the class Raphidophyceae have been associated with fish kill events in Japanese, European and U.S. coastal waters. Fish killing activity is attributable to toxin production, and gill damage via production of superoxide radicals, physical clogging, and hemolytic substances. Morphological identification of these organisms in environmental water samples can be difficult, particularly when fixatives are used. Because of this difficulty, and because these toxinproducing species have been found in estuarine waters of the mid-Atlantic, we initiated the development and validation of a suite of real-time PCR assays. Through sequencing we generated a complete data set (18S, ITS, partial LSU, 16S) for a panel of characterized raphidophyte cultures from various geographic locations and developed rapid and specific assays for several species (Chattonella verruculosa, C. cf. verruculosa, C. marina, C. subsalsa, Heterosigma akashiwo and Fibrocapsa japonica) using appropriate loci. Utilizing this comprehensive data set, we were able to perform phylogenetic analyses that allowed us to determine the genetic relationship between these species, and in particular determine the relationship of C. verruculosa and the newly described C. cf. verruculosa to members of the family Raphidophyceae.

## **POSTER – Session 2, Poster 3**

# Potential roles of *Prorocentrum minimum* to Chesapeake Bay dissolved oxygen and oyster dynamics

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As in other nutrient-enriched estuaries, the dinoflagellate Prorocentrum minimum is a recurrent bloom forming species in the Chesapeake Bay and its tributaries. Laboratory studies were conducted to assess potential bloom impacts on diel oxygen concentrations in shallow littoral zones as well as settlement success and post-set growth of the eastern oyster Crassostrea virginica. Using light-dark and dark cultures and periodic diel subsampling, bloom levels of P. *minimum* produced supersaturated oxygen levels at the end of each day while darkened cultures were typified by rapid decreases in DO  $(1.1-1.3 \text{ mgL}^{-1}\text{h}^{-1})$  to hypoxic and anoxic levels within 4 days. These data suggest shallow, poorly flushed systems and the biota in them will experience rapid and large diel variations in oxygen, implying recurrent P. minimum blooms need be considered as short-term oxygen stressors for Bay oyster spat and other living resources. Direct effects of P. minimum impacts on oysters were not as expected or previously reported. In one experiment, settlement of larval oysters on PVC plates pre-exposed to pre-bloom P. minimum isolates was the same as recorded for plates exposed to filtered seawater, Instant Ocean®, or Isochrysis. In a second oyster experiment, spat on PVC plates were exposed to field collected P. minimum blooms and a commercial mixture of several other food types including Isochrysis. Oyster growth was significantly higher in *P. minimum* exposures than noted in the commercial mix. These results, compared to results with other isolates from the same region, indicate substantial positive impact from some of the P. minimum blooms of the area while others separated in space, time, or nutrient status could severely curtail oyster success through toxin production induced by nutrient limitation.

### **POSTER – Session 2, Poster 16**

### Intraspecific variability in the nutritional ecology of harmful algae

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Most generalizations about the nutritional ecology and physiology of harmful algae are based upon consideration of one to a few strains per species. Yet, when multiple strains have been experimentally examined, significant intraspecific variability commonly has been documented. Ecological benefit would be expected from the ability of strains within a species to show a range of responses to changing nutrient availability. We present examples of high intraspecific variability for diverse harmful algae in response to macronutrient (N, P, C) limitation, micronutrient limitation, pulsed versus continuous nutrient enrichment, and nutrient form. These nutritional differences, in turn, have influenced the bulk chemical composition (lipids, proteins, carbohydrates), cell production, life history expression, susceptibility to grazing, and toxin production of a given harmful algal species. In fact, opposite interpretations, and opposite generalizations, would result from consideration of individual strains. The data collectively underscore the importance of considering multiple strains in research to advance understanding about the nutritional ecology of harmful algal species, toward reliably predicting their response to changing nutrient regimes.

# ORAL

## How does eutrophication affect the role of grazers in harmful algal bloom dynamics?

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Population dynamics of harmful algal bloom species are regulated both from the "bottom up" by factors that affect their growth and from the "top down" by factors that affect their loss rates. While it might seem obvious that eutrophication would have the greatest effect on factors affecting growth rates of phytoplankton (nutrient supply, light availability) the roles of top-down controls, including grazers and pathogens, cannot be ignored in studies of harmful bloom dynamics. Grazers that avoid feeding on harmful species and actively graze on competing species may play important roles in bloom initiation. Grazers that are not affected by phytoplankton toxins and have growth rates comparable to phytoplankton (protozoan grazers) may have the potential to control the initiation of blooms. There is growing evidence that the inhibition of grazers may vary with cell concentration for blooms of toxic phytoplankton. The potential effects of eutrophication on grazer-bloom interactions will be examined using examples from both red tide (*Karenia brevis*) and brown tide (*Aureoumbra lagunensis*) blooms along the coast of Texas.

# ORAL

# The effect of nutrient concentration at different growth stages on hemolytic ability of three clones of the ichthyotoxic flagellate *Prymensium parvum* from blooms in the United States

#### Clouse, Melissa, and Carmelo Tomas

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Since the mid-1980s, blooms of the ichthyotoxic flagellate *Prymnesium parvum* have been recurrent in Texas. More recently, South Carolina experienced a bloom on May 22, 2001 at a brackish golf course pond lacking fish in Kiawah Island. Artesian Aquafarms in Elizabeth City, North Carolina experienced *P. parvum* blooms in 2002, which started in March and persisted through October. When the water source changed from fresh to brackish to optimize growth of hybrid striped bass, *Prymnesium* bloomed, causing large fish kills. Low phosphate concentrations were found in association with these *P. parvum* blooms. Based on the natural conditions associated with the blooms, clones were isolated and used under laboratory settings to better understand the relationship between nutrients, bloom dynamics, and toxin production. Cultures were grown and counted daily in nutrient-replete, nitrogen-limited, and phosphorus-limited conditions. Samples were taken at various stages in growth (lag, log, and stationary phases) to assess their hemolytic ability. Preliminary studies show that both nutrient concentrations and growth phase affect hemolytic ability. Hemolytic abilities also vary among the three clones.

### **POSTER – Session 1, Poster 4**

### Nitrogen uptake by the toxigenic diatom Pseudo-nitzschia australis

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The nitrogen uptake capabilities of the toxigenic diatom *Pseudo-nitzschia australis*, freshly isolated from Monterey Bay, California, were examined in uni-algal laboratory cultures at saturating photosynthetic photon flux densities (100  $\mu$ E m<sup>-2</sup> s<sup>-1</sup>) and 15°C. The kinetics of nitrogen (nitrate, ammonium, urea and glutamine) uptake as a function of substrate concentration were estimated from short (10-min) incubations using the <sup>15</sup>N-tracer technique, and are compared to the long-term exponential growth rates of *P. australis* determined in semicontinuous, batch cultures grown on the various nitrogen substrates. Based on the estimated maximum specific uptakes rates, nitrate is the preferred nitrogen substrate, but rates of urea uptake by *P. australis* did not appear to saturate even at concentrations as high as 36 µg-at N L<sup>-1</sup>. The growth rate (determined using *in vivo* fluorometry) of *P. australis* was slower for cells grown on urea (0.15 d<sup>-1</sup>) compared to the cells grown on nitrate and ammonium, which both

maintained significantly greater growth rates (*ca.*  $0.6 \text{ d}^{-1}$ ). However, initial results suggest that the particulate domoic acid content of the urea-grown cells was significantly greater. These laboratory results demonstrate the capability of this diatom to grow equally well on both oxidized and reduced forms of nitrogen, supporting our field observations that *P. australis* blooms during both upwelling and non-upwelling conditions off the west coast of North America. During these times, substantial differences in the nitrogenous nutrition of *P. australis* can be expected, and anthropogenic inputs of reduced N substrates could contribute significantly to its growth.

# ORAL

# Autonomous nutrient monitoring and water sampling as tools for studying HABs: Progress and prospects

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Frequently, sampling of HAB events provides inadequate definition of the nutrient conditions that precede and accompany these outbreaks. Reasons for this include, weather events that make traditional sampling difficult, and the time demands on personnel involved in a several-week or longer high frequency sampling experiment. Because of these difficulties, we have deployed autonomous nutrient monitors in the Choptank and Pocomoke Rivers (Chesapeake Bay tributaries) to test whether high frequency nutrient data will provide a better understanding of HABs. Our deployments include devices that can determine and telemeter ammonium, nitrate, nitrite and reactive phosphorus, and we are developing a urea monitor. We are also working on interfacing a telemetered data stream (including nutrients) with automated water samplers, so that samples for phyotplankton speciation, etc. can be taken autonomously, before and during the development of an HAB. We believe that these automated approaches provide improved opportunities for advancing our understanding of relationships between nutrients and HAB outbreaks.

So far, our data reveal relationships between nutrients and rainfall events that would be difficult to observe using conventional methodology. For example, while both nitrate and phosphate increased after rainfall events in the Pocomoke River as expected, both records revealed high frequency variability that would be missed by most traditional sampling, the character of the inputs was also different. One difference was that the phosphate data show strong but extremely short duration spikes that are not evident in the nitrate record. In this presentation, we will review our suite of autonomously collected nutrient data and discuss our future plans for integrating these samplers with automated adaptive water samplers.

## ORAL

# Urea-ammonium-nitrate interactions in Thau lagoon (Southern France): relationships with *Alexandrium catenella* blooms

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Monitoring programs in Thau lagoon (Southern France) indicate a threshold effect of urea (0.5 to  $1.5 \mu mol N/liter$ ) on the triggering of *A. catenella* blooms with a time lag of about a week. This confirms results by Glibert & Terlizzi (1999) obtained for dinoflagellate blooms in aquaculture ponds. However the effect of urea may not be a direct one as positive correlations with ammonium are also sometimes observed. Time series over a 6 month time scale at a fixed point also show correlations between urea and dissolved organic nitrogen (other than urea) indicating that urea represents between 3 and 25% of DON in that environment. Concentrations and rate measurements are used to describe interactions between urea, ammonium and nitrate. In the field, part of urea is probably degraded to ammonium before being taken up by the cells. In laboratory cultures, ammonium is also excreted by *A. catenella* during urea assimilation and leads to inhibition of nitrate uptake. Thus there are complex interactions between those three nitrogen sources in field situations.

### ORAL

# Improved accuracy of quantitative real-time PCR of HAB species in environmental water samples using an exogenous DNA internal standard

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Quantitative real-time PCR (QRT-PCR) is an extremely powerful and sensitive method for quantitative detection of microorganisms. The potential of this methodology for enumeration of harmful algae has been demonstrated for several species, and its application to environmental investigations may help identify correlations between nutrient concentrations and cell abundances at pre-bloom levels. While the process is relatively straightforward, existing QRT-PCR methods often fail to incorporate controls to assess the accuracy of the results. Co-precipitation of inhibitory compounds with DNA extracted from environmental samples, in particular, can affect the outcome of quantitative PCR by several fold. Here, we describe the development and rigorous analysis of QRT-PCR assays for the Raphidophyte species *Chattonella subsalsa* and *Heterosigma akashiwo* in which we introduce a known concentration of exogenous DNA into the extraction buffer as an internal standard. Since the target DNA is

extracted in the presence of the internal standard, inherent variability in extraction efficiencies and the presence of inhibitors equally affect both the target and standard. The introduction of an exogenous internal standard also reduces variability due to human error such as differences in laboratory technique, day-to-day proficiency and pipetting or dilution errors. We rigorously tested QRT-PCR assays for *H. akashiwo* and *C. subsalsa*. Using primers and probes targeting the 18S rDNA gene, this method has a detection range of over 8 orders of magnitude and is sensitive enough to detect as few as 4 copies of the target gene. In addition, we evaluated intra-samples variability and accuracy of this assay using environmental bloom samples as calibrators. The results of this investigation demonstrate that QRT-PCR is a sensitive and accurate method for quantitative analysis of HAB species in environmental water samples and is amenable to the rigors of routine monitoring efforts.

## **POSTER – Session 2, Poster 5**

# Relationships between nitrogen loading and concentrations of nitrogen and chlorophyll in coastal embayments

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We describe results obtained with a simple model that uses loading rates of total nitrogen (TN), defined as dissolved inorganic nitrogen plus dissolved and particulate organic nitrogen, to calculate annually and spatially averaged concentrations of TN in coastal embayments. We also describe ongoing research to extend this model to estimate planktonic chlorophyll a from nitrogen loading.

The Embayment Nitrogen Model (ENM) is a simple mass balance model that predicts annual average concentrations of TN in coastal embayments from TN input rates from watersheds, atmospheric loading, and point sources. The ENM calculates internal loss rates of nitrogen in the embayment to processes such as denitrification and burial in sediments as first-order in nitrogen, net export across the seaward boundary from the embayment flushing time, and input across the seaward boundary from salinity data and the nitrogen concentration at this boundary. The model demonstrates the importance of flushing time in determining the magnitudes of nitrogen losses to flushing and internal sinks. It has been used to calculate annually and spatially averaged concentrations of TN in numerous embayments, and appears to be useful for estimating sensitivity of embayments to nitrogen loading.

Research in Long Island Sound has revealed power-law relationships between chlorophyll *a* and TN concentrations and has provided insights into seasonal and interannual variations in these

correlations, as well as their causes. These relationships are being evaluated for possible addition to the nitrogen model to permit prediction of chlorophyll *a* concentrations from TN loading. Data from Boston Harbor and Massachusetts Bay have shown similar relationships between TN and chlorophyll *a*, and data from other embayments are now being analyzed to determine their generality, including the degree of interannual and system-to-system variability.

# **POSTER – Session 2, Poster 11**

## NOAA extramural HAB research: Present and future

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In the US virtually every coastal state has reported major algal blooms, resulting in threats to human and animal health and coastal economies. In response NOAA has developed a variety of extramural programs that address causes, impacts, and possible remedies. Two programs focus entirely on understanding, predicting, and monitoring Harmful Algal Blooms, Ecology and Oceanography of Harmful Algal Blooms (ECOHAB) and Monitoring and Event Response (MERHAB). Three other NOAA programs address specific HAB problems. The Oceans and Human Health Initiative (OHHI) is developing partnerships within NOAA and with the external ocean and human health community to address a broad array of interrelated research topics, including human health aspects of marine toxins. Sea Grant addresses HAB problems in the context of environmental stewardship and mitigation of economic impact through research, detection technology, outreach and education. The Marine Mammal Health and Stranding Response Program is assessing the role of biotoxins in marine mammal morbidity and mortality.

Recent reports and legislation will guide the future expansion of NOAA HAB research. The importance of HAB research, and especially the ecosystems approach taken by NOAA programs, has been emphasized by the report of the US Commission on Ocean Policy. The reauthorized Harmful Algal Bloom and Hypoxia Research and Control Act requires reports on Prediction and Response and National Scientific Research, Development, Demonstration, and Technology Transfer within one year and Scientific Assessments of Freshwater and Marine Harmful Algal Blooms within two years. Passage of the Oceans and Human Health Act and relocation of the OHHI program within NOAA will insure its continuation and increase coordination with other HAB research. Title IV of the Marine Mammal Protection Act requires that NOAA evaluate the causes of morbidity and mortality in marine mammals.

ORAL

# The assessment of brown tide blooms caused by the alga, Aureococcus anophagefferens and related environmental factors in coastal waters of New Jersey (2000-2002)

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This study was conducted in Barnegat Bay-Little Egg Harbor (BB/LEH), New Jersey (USA) (2000-2002) by the New Jersey Department of Environmental Protection, Division of Science Research and Technology (DSRT), in cooperation with several partners, to assess the spatial and temporal extent of A. anophagefferens blooms and associated environmental factors and analyze the potential risk of these algal blooms to submerged aquatic vegetation (SAV) communities. A. anophagefferens were enumerated and associated environmental factors were measured from 2000-2002. A. anophagefferens abundances were classified using the Brown Tide Bloom Index and mapped, along with sampled parameters, to their geo-referenced location using the ArcView GIS. To determine the possible risk that brown tide blooms pose to the BB/LEH submerged aquatic vegetation (SAV) communities, the GIS was used to determine the spatial coincidence between locations of high A. anophagefferens abundances or duration and the mapped location of seagrass habitat. The results indicated that the highest A. anophagefferens abundances (>10<sup>6</sup> cells ml<sup>-1</sup>), including Category 3 blooms ( $\geq$  200,000 cells ml<sup>-1</sup>) and Category 2 blooms ( $\geq$  35,000 to  $\leq$  200,000 cells ml<sup>-1</sup>), recurred during each of the three years of sampling and covered significant geographic areas of the estuary, especially in Little Egg Harbor. While Category 3 blooms were generally associated with warmer water temperatures (> 16°C) and higher salinity (> 25-26 ppt), these factors were not sufficient alone to explain the timing or distribution of A. anophagefferens blooms. There was no significant relationship between brown tide abundances and dissolved organic nitrogen measured in 2002 but this was consistent with other studies. Extended drought conditions, with corresponding low freshwater inputs and elevated bay water salinities, occurring during this time were conducive to blooms. A. anophagefferens abundances were well above the reported threshold levels that have been reported for negative impacts on shellfish. For the first time, it was shown that 50% of the seagrass habitat located in Barnegat Bay/Little Egg Harbor was categorized as having a high frequency of Category 2 or 3 blooms for all three years.

**POSTER – Session 1, Poster 17** 

### Diagnostic indicators of HAB nutritional physiology

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Nutrient availability can influence important aspects of toxic algae biology and ecology, such as growth, toxin production, and life cycle, as well as bloom initiation, persistence and decline. One way of studying nutritional physiology, and potentially better understanding the cues which allow harmful algal blooms (HAB) to form, is to directly monitor cell physiology in a nutrientspecific manner. There are a number of recent advances in our ability to monitor the nutritional physiology of HAB in complex communities. These advances include the application of immuno-separation techniques, immunoassays for nutrient-regulated proteins, and enzyme labeled fluorescence detection of different hydrolytic activities. For example, we have developed and applied assays for phosphorus bioavailability in Prorocentrum minimum from Narragansett Bay. We have also used an immuno-separation approach to examine urease activity in Alexandrium fundvense populations from the Gulf of Maine. These cell-specific approaches avoid many of the complications of traditional bioassays or nutrient addition experiments (e.g. grazing) and other diagnostic methods, because they are specific to particular harmful taxa and they can directly indicate nutrient bioavailability to a particular cell. Such diagnostic indicators can provide near real-time information about the physiological status of different species from mixed communities, and they represent a powerful approach to studying nutritional physiology and ultimately examining links between HAB and eutrophication.

## ORAL

### Harmful algae and eutrophication in the Baltic Sea area

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Despite the long tradition of phytoplankton studies in the Baltic Sea area it is only during the last 25 years that toxic, or in other ways harmful, phytoplankton have been given well-deserved attention. The reason for this may partly be found in the lack of realizing connections between the presence of certain phytoplankton species and the injury and mortality of marine, as well as terrestial animals. Whereas some of the harmful phytoplankton species thrive and develop blooms near the coast, in river mouths, harbour basins and close to big cities, there are also considerable blooms in the open Baltic Sea, mainly manifested by the annual surface accumulations of the toxic cyanobacterium *Nodularia spumigena*. Harmful phytoplankton not only harm man and animals. They also have a considerable economic impact on fishery and recreation. The problem of eutrophication in the coastal area of the Baltic Sea goes back to the early 20<sup>th</sup> century, but it is only about 35 years since it was shown that the open Baltic Sea might suffer from eutrophication. The connection between the increase in presence and blooms of harmful phytoplankton and the increased eutrophication is discussed using examples from

different parts of the Baltic Sea. It includes blooms of *Nodularia spumigena*, *Prorocentrum minimum*, *Dinophysis* spp., *Prymnesium parvum* and *Chrysochromulina polylepis*. While eutrophication in the Baltic Sea is expected to decrease in future, mainly because of the reduction of local nutrient discharge, there is a potential risk that the introduction of harmful phytoplankton species will increase, due to the increasing ship traffic into the Baltic.

# **ORAL-Invited**

### Garbage in, Garbage out ? - Problems in experiment design and modelling of HAB ecology

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Consideration is given to the problems faced by modellers in obtaining data for the design of models and subsequent verification of their output, and in the understanding of these problems by biologists. Too many experimental studies do not yield data of sufficient breadth and frequency to make them useful for modelling work. There are also significant holes in the coverage of experiments on HAB species with respect to the interactions of nutrients at different concentrations, the use and production of DOM, and for mixotrophy. Studies of zooplankton and bacteria are similarly poor with respect to design to support modelling efforts. In part as a consequence, models of organisms important in HAB events may be incomplete or contain flawed components. Both biologists and modellers need to spend more time explaining their interests to each other. In particular, modellers should not simplify biological systems without biological justification.

## ORAL-INVITED

### The role of nutrient conditions on toxicity, allelopathy and mixotrophy in HABs

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The role of nutrients for toxin production has been investigated in many phytoplankton groups especially species belonging to the genera *Alexandrium* (dinoflagellates) and *Pseudonitszchia* (diatoms). For these two genera, phosphorus limitation induces a higher production of toxins compared to that of cells grown under nitrogen limitation or under nutrient sufficient conditions. This has been suggested as a mechanism enabling the algae to store the excess nitrogen in the growth medium, since the toxins have a high nitrogen content. Under phosphorus limitation nitrogen accumulates in the cells and part of it is used in more production of the toxins. The same situation applies for the cyanobacterium *Nodularia spumigena* (even though this species is able to obtain their own nitrogen in case of deficiency through  $N_2$  fixation). For the

dinoflagellate Dinophysis acuminata on the other hand the opposite occurs, i.e., under Nlimitation is when okadaic acid production increases. The cells of these above mentioned HABs under nitrogen deficiency have the lowest toxin concentrations per cell, but toxin will be produced nevertheless. However, for the species whose toxins do not contain either N or P, toxin production increases when the algae are growing under nutrient limiting conditions, independent if phosphorus or nitrogen is the limiting nutrient. Ichthyotoxic species are among this phytoplankton group. Thus, there is a group of phytoplankton species that can increase toxicity in the cells when they are growing under both N- and P-deficiency. It seems that it is not only toxicity that increases under nutrient limitation but for some of these algae but also their grazer deterrence, their production of allelochemicals and their mixotrophic capabilitity. Killing the nutrient-competing phytoplankton species enables these HABs to freely utilize limiting resources and by adding mixotrophy, i.e., the capability to ingest bacteria, other algae and even potential grazers, will contribute even further to the bloom-forming ability of these species. We can thus assume that a consequence of the increased input of N and P to aquatic ecosystems is provoking an unbalanced nutrient situation for "normal" phytoplankton species to growth but favoring blooms of species able to increase their toxin production, allelopathy and mixotrophy under such nutrient unbalanced conditions. In conclusion it seems that the increased discharge of nutrients to coastal waters might not only be behind an increase in algal blooms but also that some of these blooms are more toxic, and the species involved have other kind of advantages to form bloom in relation to the "so-called" normal photosynthetic phytoplankton species.

### **ORAL-INVITED**

# Harmful phytoplankton indicator species applied to eutrophication assessments of Scottish coastal waters supporting aquaculture

#### Gubbins, M.J., P.J. Sammes, and I.M. Davies

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The intergovernmental Oslo and Paris Commission (OSPAR) 'Comprehensive Procedure' for assessing marine eutrophication status involves the application of harmonised assessment criteria covering nutrient enrichment (category I), direct (category II), indirect (category III) and other (category IV) effects of eutrophication. Elevated concentrations of toxic/nuisance phytoplankton indicator species are an important (category II) assessment parameter within this procedure. Presented here are the results of a monitoring programme to assess the eutrophication status of Scottish sea lochs supporting high levels of finfish aquaculture. Sea lochs subject to high levels of nutrient enrichment from aquaculture sources were selected for monitoring and compared to less intensively farmed areas. During 2002 and 2004, 14 and 27 lochs respectively were sampled for phytoplankton (integrated 10 m depth) and the species present identified and quantified by microscopy. Over the course of the survey several toxic/nuisance species were observed including notable blooms of *Gymnodinium* and *Chaetoceros*. There was no apparent relationship between phytoplankton diversity or the presence of toxic/nuisance species and nutrient enhancement from aquaculture. However, assessment of this parameter under the OSPAR

toxic/nuisance phytoplankton is elevated in areas supporting aquaculture. Therefore this programme of monitoring and assessment has been expanded and extended to 2006.

**POSTER** – Session 2, Poster 6

# Monitoring toxic phytoplankton and shellfish in support of eutrophication assessments for Scottish coastal waters

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Under the Oslo and Paris Commission (OSPAR) 'Comprehensive Procedure' for assessing the eutrophication status of marine waters, the incidence of potentially toxic phytoplankton species and algal toxins in shellfish are considered as relevant parameters for assessment, forming an important component of the 'Harmonised Assessment Criteria' (HAC). Current phytoplankton monitoring programmes in Scotland are based on time-consuming identification of potentially toxic species by microscopy, and do not measure toxicity. In this study, we have determined the feasibility of applying the Jellet Rapid Test (JRT) to concentrated phytoplankton net tow samples in the field to monitor the presence of paralytic shellfish poisoning (PSP) and amnesic shellfish poisoning (ASP) toxins. During eutrophication assessment surveys of 16 sea lochs and voes around Scotland the JRT detected the presence of PSP toxins in six phytoplankton net tows and ASP toxins in 13 net tows taken from a research vessel during April 2004. Using speciesspecific molecular probes, known toxin producing phytoplankton species were detected at these sites. HPLC techniques also detected PSP and ASP toxins in mussel (Mytilus edulis) samples at concentrations below the current regulatory limits. These data, when integrated with monitoring results from other parameters of the OSPAR HAC, help to provide a more holistic assessment of the eutrophication status of Scottish coastal waters.

#### **POSTER – Session 2, Poster 7**

#### Fatty acids and growth in the heterotrophic dinoflagellates *Pfiesteria* spp. and PLO's

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*Pfiesteria* spp. are heterotrophic dinoflagellates implicated in human health and fish mortality events on the eastern seaboard, USA. Typically cultured on algae, with cryptophytes the preferred prey, *Pfiesteria* spp. also feed on a wide range of animal prey including the epidermis of larval and juvenile fishes, and a variety of larval and adult invertebrates. Prey type has been

shown to affect life stage transformations and division cyst development. The growth dependence on live prey complicates the task of defining required nutrients for growth. We recently reported the capability for sustained, exponential growth ( $0.35 d^{-1}$ ) by *P. shumwayae* on a prey-free, lipid-rich organic media. In the present study, we compare the capability of *Pfiesteria* strains and species for growth on this dissolved media. In addition, growth on the dissolved media provided the opportunity to investigate the biochemical nutrient requirements for growth for *Pfiesteria* spp. and *Pfiesteria*-like organisms (PLO's). We compared the role of specific lipids/fatty acids to support growth in these species. We hypothesize a relationship between chemosensory attraction to specific fatty acids and their role as required nutritional components for growth. We propose a role for the peduncle in the uptake of dissolved substrate by *Pfiesteria shumwayae*. The results of this study are discussed in the context of prey selection in the natural environment. Supported in part by ECOHAB Grant NA-160P1487.

# **POSTER – Session 1, Poster 6**

#### Comparative analysis of the relationships between nutrient cycling and phytoplankton community composition in two eutrophied subtropical estuaries: Florida Bay, USA, and Moreton Bay, Australia

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Florida Bay, USA and Moreton Bay, Australia are both shallow, subtropical estuaries at equivalent latitudes which lie adjacent to major metropolitan areas (i.e. Miami and Brisbane), are subjected to increasing eutrophication, and are characterized by frequent harmful phytoplankton blooms. We compared nutrient (C, N, P and Si pools, ratios, fluxes) and phytoplankton community composition over an annual cycle in both estuaries. Similarities observed between the estuaries include: a temperature effect upon primary productivity and a significant relationship between the form of nitrogen dominating phytoplankton uptake and phytoplankton community composition. In both systems, increases in the contribution of DON to total phytoplankton nitrogen uptake were positively correlated with the percentage of dinoflagellates in the community while increases in the NO<sub>3</sub> contribution favored diatom dominance. Fundamental differences were observed in the mode of suspended sediment inputs to each estuary and the nutrient limiting primary production. Within Moreton Bay, seasonal riverine inputs of suspended sediments impacted productivity via phosphorus delivery and light attenuation, while in Florida Bay, episodic wind related sediment resuspension events contributed to significant benthic pelagic coupling. Bioassay responses and nutrient uptake data support nitrogen limitation in Moreton Bay and both nitrogen and phosphorus limitation in Florida Bay. A comparative analysis of these systems supports the importance of both local hydrology and the prevalent dissolved nitrogen form on algal bloom development in subtropical estuaries.

# ORAL

# New technologies for monitoring and assessing harmful algal blooms and water quality in Chesapeake Bay, Maryland

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The Maryland Department of Natural Resources' (DNR) shallow water monitoring program employs state-of-the-art temporally and spatially intensive monitoring systems for monitoring ambient water quality, nutrient concentrations and harmful algal blooms.

The continuous monitoring network provides real- and near-time temporarily intensive water quality data from 34 fixed stations in Maryland's shallow water habitats. Real-time chlorophyll, dissolved oxygen and pH data, telemetered to the internet, provide early warnings of developing harmful algal blooms, and monitor the intensity of these blooms through time. An automated email alert system notifies staff of bloom conditions and allows additional sample collection to be coordinated in a timely fashion. Current efforts are underway, in coordination with the University of Maryland Center for Environmental Science, to trigger autosamplers with continuous and/or *in situ* nutrient monitors to better understand real-time relationships between water quality and nutrient dynamics with HAB events. The continuous monitoring network can provide even greater future utility by incorporating emerging technologies such as toxin detecting sensors.

DNR's Water Quality Mapping Program uses rapid continuous monitoring measurements aboard a moving small boat to characterize the spatial distribution of HAB events throughout Bay tributaries. A new real-time onboard mapping interface is being developed for this system to provide an adaptive sampling tool that will aid in the delineation of blooms and provide greater certainty in spatial water quality data collection.

Real and near-time results from continuous monitoring, water quality mapping, and HAB events are made available through a map-based interface on DNR's Eyes on the Bay website (www.eyesonthebay.net). The website has proved to be a valuable tool for dissemination of

alerts and stories on human and habitat impacts from HABs and a useful repository of HAB data from previous years.

ORAL

# Modeling Pfiesteria life cycle attributes and population dynamics

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As part of a multi-disciplinary ECOHAB study we developed a suit of mechanistic models which provide insight into some of the life cycle attributes and population dynamics of *Pfiesteria spp*. In the first phase of this effort we developed a numerical model to extract *Pfiesteria* growth and encystment parameters from data generated in laboratory experiments. By applying the numerical model we were able to isolate several key parameters for a "non-inducible" strain of Pfiesteria, including grazing rates, gross growth and assimilation efficiencies, as well as the retention time of chloroplasts. We also developed a more generalized model based upon literature data and laboratory observations for studying non-inducible strains. With this model we followed the time dependency of both individual body size and abundance of *Pfiesteria* zoospores, which provides a means to represent some of the unique aspects of Pfiesteria population dynamics, such as transformations among zoospores, and subtle relationships between food consumption, body size, mortality and division rate. The model successfully captured the major features of *Pfiesteria* population dynamics in a laboratory culture experiment, and suggests that *Pfiesteria* populations can be regulated by both bottom-up and top-down control, as suggested by recent field and laboratory studies. In the final phase of our modeling efforts we developed a second generation Pfiesteria population model that is capable of representing both non-inducible and toxic strains of *Pfiesteria*. We carried out a series of experiments with this model where we attempted to simulate the effects of turbulence, nutrient concentration and composition, and grazing on Pfiesteria population dynamics. Among other things, these experiments suggest that toxic blooms are more likely to occur in calm, organicnutrient rich conditions, which are often found in shallow, protected tributaries that are subject to anthropogenic effects. These results are therefore generally consistent with observed patterns of toxic blooms in Chesapeake Bay and the Neuse River of North Carolina.

ORAL

#### Nitrogen pollution: Sources, trends, and effects globally and regionally

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Human activity has roughly doubled the creation of reactive, biologically available nitrogen on the land masses of the Earth. Regional variation in this increase is great, and some regions of the Earth have seen little change, while in other areas, nitrogen fluxes through the atmosphere and through rivers have increased by 10- to 15-fold or more. Much of this increase has occurred over the past few decades. Increased use of synthetic nitrogen fertilizer and increased intensity of meat production has led the change globally and in many regions, but atmospheric deposition of nitrogen from fossil-fuel combustion also contributes globally and is the largest single source of nitrogen pollution in some regions. The consequences to coastal systems are many, and include degradation and alteration of habitat and food-web structure and loss of biotic diversity in addition to hypoxia and anoxia. Nitrogen pollution also has major effects on human health and on the ecological functioning of terrestrial ecosystems.

### **ORAL- INVITED**

Bioavailability of dissolved organic phosphorus compounds to typical harmful dinoflagellate *Prorocentrum donghaiense* Lu

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*Prorocentrum donghaiense* Lu is responsible for large scale bloom events (ca. 10000 km<sup>2</sup>) in Yangtzs River estuary and the adjacent East China Sea during spring in recent years. Some studies suggest that the East China Sea, especially the Yangtzs River estuary is P-limited. However, in the bloom season (spring time), higher dissolved organic phosphorus (DOP) waters coincide with higher Chl. a areas and DOP in this area contributes about 40% of total dissolved phosphorus. The potential ecological significance of DOP for the bloom needs to be investigated.

The bioavailability of DOP compounds to harmful alga *Prorocentrum donghaiense* Lu and its alkaline phosphatase activity (APA) were studied. Results showed that *P. donghaiense* could utilize the test DOP compounds such as glucose-6-phosphate (G-6-P), adenosine triphosphate (ATP) and ribonucleic acid (RNA) to sustain its growth. Nutrition between the test DOP compounds and orthophosphate was comparable, although *P. donghaiense* grew better in ATP and RNA media than that in orthophosphate media. *P. donghaiense* could utilize intracellular phosphorus to sustain growth under depletion of dissolved phosphorus. Variation of APA in

different test DOP compounds was addressed, the controlling mechanism of APA in different DOP media are discussed.

### **POSTER – Session 1, Poster 12**

# Modeled *Karenia brevis* bloom initiation and subsequent accumulation in the vicinity of a coastal nutrient front

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The two dimensional, alongshore independent, distribution of K brevis cells in a shelf environment forced by moderate upwelling favorable winds in the presence of a nearshore surface nutrient plume and an offshore bottom nutrient source over a sloping bottom. The current field, due to a moderate upwelling favorable wind with a magnitude of  $0.4 \times 10^{-4} \text{m}^2 \text{s}^{-2}$  over a bottom topography which decreases linearly with onshore distance from a depth of 40m to a depth of 10m with a slope of  $5 \times 10^{-4}$ , is first calculated analytically using a constant eddy viscosity assumption. The nutrient field is postulated to be composed of two components. The first is a surface plume with a maximum surface value of 1.25 µM simulating outwelling that extends from the coast offshore to approximately 27 m water depth and decays downwards from the surface and in the offshore direction. The second is a bottom source simulating a sediment flux (maximum bottom value of 5 µM) that extends from deep water onshore to a water depth of approximately 23m water depth and decays upwards and in the onshore direction. A light field with a maximum noon value of 1500  $\mu$ M quanta m<sup>-2</sup> s<sup>-1</sup> is applied to the system. The current, light, and nutrient fields are utilized in a previously developed extended Eulerian model which treats the concentration of the K. brevis cells as a function of space, time, the internal pools of nitrogen and carbon, and long term light exposure. At the start of the calculation no cells are present in waters shallower than 40m. At the 40m boundary a concentration of cells at 10<sup>6</sup> cells  $m^{-3}$  is specified in the onshore flowing region from the bottom to mid-depth and this concentration at the boundary is held fixed in time. The distribution of cells over a 37 day simulation is then examined.

# **POSTER – Session 2, Poster 12**

#### Eutrophication and harmful Algal Blooms in the Swan River estuary, Western Australia

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The Swan River estuary with its tributaries formed the focus point around which Perth the capital of western Australia was established when European settlers colonised Western Australia 175 years ago. From the early period the Swan River experienced filamentous algal blooms. With urbanisation and intense clearing and cultivation of the watershed signs of microalgal blooms began to emerge. It was only 25 years ago the basic pattern and succession of phytoplankton blooms was established in relation to the trophic status of the river. Diatoms, dinoflagellates and green algae initiate major blooms in the estuary seasonally regulated by rainfall in winter and marine water incursion in the summer–autumn period. Unusual weather pattern appear to be associated with many unpredicted blooms. Within the last 15 years supplementing the above pattern of algal blooms. These include toxic dinoflagellates and cyanobacteria. The species involved are *Dinophysis acuminata*, *Karlodinium micrum*, *Microcystis flose-aquae*, *Anabaena circinalis*, and *Anabaena spiroides*. This paper analyses the dynamics and environmental conditions associated with these blooms.

### **POSTER – Session 1, Poster 1**

#### An autonomous urea monitor for studying HABs

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Urea is increasingly being employed as a fertilizer on a global basis and in the watersheds of Maryland and the rest of the United States. Samples collected by hand indicate that concentrations can rival and at times exceed ammonium concentrations in Chesapeake Bay tributaries and in the Coastal Bays of Maryland. In addition, there are suggestions that urea concentrations may be a source of nitrogen favored by some HAB species. Because our experience with autonomous monitors for phosphate, nitrate and ammonium suggested variability that would be exceedingly difficult to resolve with conventional sampling, we decided to add a urea monitor to our suite of monitors.

The approach that we have taken is to implement the acetyl-monoxime urea chemistry on a modified monitor that we normally use to determine nitrate and nitrite concentrations. Preliminary study and tests suggested that the major requirement needed to accomplish this would be the construction of a high temperature (90°C) heater not currently available for the Envirotech NAS-2E nutrient monitor that we are modifying. While an colorimeter with

optimized wavelength sensitivity and optical path-length is desirable, we have demonstrated that "off the shelf" NAS nitrate+ nitrite colorimeters can also produce useable results, and we have experimented with both types of colorimeter. Our plans are to complete laboratory testing of the urea monitor in February, and to start deploying the instrument in March 2005.

# **POSTER – Session 2, Poster 9**

# Do the coastal eutrophication and warming cause widespread and persistent *Cochlodinium polykrikoides* blooms in Korean waters?

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Since 1995, a fishkilling dinoflagellate *Cochlodinium polykrikoides* have made widespread blooms and have recurred every summer for one or two months. However, the sporadic and temporary diatom blooms had been prevailed till early 1980s. So far, there has been no clear identification on the outbreaks mechanisms and the widespread and persistency of this dinoflagelate bloom. It is essential to clarify what is the relationship between coastal eutrophication and widespread *C. polykrikoides* blooms.

Based on the compiled nutrients data collected from the regular coastal environmental monitoring in Korean waters, a gradual nutrient increase has been observed for the last three decades. And it was found that their major sources were direct and indirect terrestrial loads, and the other minor part was the water-borne pollutants such as intensive aquaculture and exudation from bottom sediments especially in summer HABs season. In additions, geographical and oceanographical characteristics such as southwestward river runoffs and shallow marginal seas of no big exchange with ocean trap the nutrients inside the coastal areas. Accordingly, recent eutrophic level in the coastal waters bordering Yellow Sea and South Sea was more or less two fold as that of 1970s, and accompanied changes in the ratio of CNP in coastal waters. Seawater temperature along the coast, one of important parameters playing key role on the initiation and species succession of HABs, has been increased as of 0.7°C for the last three decades.

From three decadal observations of coastal water quality and the annual events of HABs, we can find some evidences that the widespread and persistent HABs were attributable to the eutrophication of coastal seawater, and the prevalence of *C. polykrikoides* blooms was owing to temperature increase.

# ORAL-INVITED

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# A role for anthropogenically derived nitrogen in the formation of harmful algal blooms along the U.S. west coast

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The U.S. west coast is generally considered to be dominated by coastal upwelling. However, recent evidence suggests that there is a substantial, and important, role for anthropogenically derived nutrients, particularly reduced nitrogen compounds such as ammonium and urea. Here we present results from field experiments in southern and central California in which the nitrogenous preference was assessed for potential HAB species. From Southern California, we examine the uptake response kinetics in the red tide forming organism Lingulodinium polyedrum, recently confirmed to produce yessotoxin. In central California, a series of nutrientamended grow-out experiments in the Gulf of the Farrallones (San Francisco Bay outflow) demonstrated that the toxigenic diatom Pseudo-nitzschia is likely present year round, supported by either nitrate, ammonium, or urea. Toxin measurements also suggest that urea-grown natural assemblages have higher toxin cell per cell than assemblages utilizing either nitrate or ammonium, and that toxicity occurs earlier in the growth phase (mid-exponential versus late exponential). In San Francisco Bay, all three N substrates were utilized by mono-specific blooms of the raphidophyte Heterosigma akashiwo, with a preference for nitrate or ammonium over urea. To assess the relative importance of anthropogenically derived nitrogen sources, we also present results from the most recent El Niño event (1998) suggestive of substantial nitrogenous input to coastal California waters during peak riverflow events. Taken together, these results demonstrate that urea is an important nitrogenous source for potential HAB species in California. and that episodic input of anthropogenic N, is a common feature of this upwelling dominated system.

# ORAL

The influence of dissolved copper on the production of domoic acid by toxigenic species of *Pseudo-nitzschia* in Monterey Bay, California

Ladizinsky, Nicolas C.<sup>1,2</sup>, G. Jason Smith<sup>1</sup>, Kenneth H. Coale<sup>1</sup> and William P. Cochlan<sup>2</sup> <sup>1</sup>Moss Landing Marine Laboratories, 8272 Moss Landing Road, Moss Landing, CA 95039 USA <sup>2</sup>Romberg Tiburon Center for Environmental Studies, San Francisco State University, 3152 Paradise Drive, Tiburon, CA 94920-1205 USA E-mail: nicolas@sfsu.edu Results from recent trace-metal experiments suggest that the production of domoic acid (DA) in *Pseudo-nitzschia* spp. may be linked to either Fe limitation, or excess Cu and Li. Due to increases in both the magnitude and frequency of DA-based harmful algal blooms along the California coast and concomitant rises in dissolved [Cu] since 1977, a field sampling effort was designed to discern the relationship between in situ concentrations of labile copper (Cu') and dissolved and particulate [DA]. Surface plankton tows and water samples were collected twice weekly from three sites along a 3-kilometer transect in Monterey Bay, CA from Mar 28 – July 3, 2001. Scanning electron micrographs of toxic and non-toxic communities revealed that the sampling period encompassed two temporally distinct *Pseudo-nitzschia* assemblages. The toxic community consisted primarily of *Pseudo-nitzschia australis* and the non-toxic community was dominated by *P. fraudulenta* and *P. heimii*. Correlation analysis between [Cu'] and DA<sub>total</sub>, yield highly significant relationships for *P. australis* at all three sites, while no correlation was exhibited for *P. fraudulenta* or *P. heimii*. While eutrophication often contributes to the initiation of monospecific blooms of some toxigenic phytoplankton, our results suggest that in the case of *P. australis* a significant relationship existed between the micronutrient [Cu'] and toxin [DA<sub>total</sub>].

#### **POSTER – Session 1, Poster 7**

#### Linkages between land-based nutrient discharges and harmful macroalgal blooms: Comparative studies on coral reefs of southeast Florida and Jamaica

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During the past two decades coral reefs in southeast Florida and Jamaica have been impacted by harmful blooms of macroalgae with a parallel reduction in hermatypic (reef-forming) corals. Many coral reef biologists attributed this "phase-shift" to overfishing of herbivorous fish stocks and die-off of the long-spined sea urchin *Diadema antillarum* (top-down control) with little consideration for the role of land-based nutrient enrichment (bottom-up control). Coastal waters in southeast Florida now receive ~  $4 \times 10^{9}$  L/d day of sewage where blooms of the green seaweeds *Codium isthmocladum*, *Caulerpa verticillata, Caulerpa racemosa, and Caulerpa brachypus* (a Pacific native) have developed since 1990. A comparison of tissue C:N:P contents of *C. isthmocladum* indicated that Floridian populations had significantly higher % P, similar %N, and lower % C, N:P, C:N, and C:P ratios compared to Caribbean populations. Near-bottom concentrations of NH4<sup>+</sup>, NO3<sup>-</sup> and SRP on Florida's reefs were high for coral reef ecosystems

and  $\delta^{15}$ N values of macroalgal tissue suggested sewage as a primary nutrient source. In the Negril Marine Park (NMP), Jamaica, increased macroalgal HABs have also correlated with increased sewage discharges and use of fertilizers in agricultural areas. Shallow reefs in the NMP are now dominated by the brown alga *Sargassum polyceratium* in the fall and winter whereas the green alga *Chaetomorpha linum* forms massive blooms in summer when SRP concentrations become elevated. Deep reefs are dominated by the calcifying green alga

*Halimeda copiosa*, which is favored in these habitats with lower irradiance and nutrient concentrations. The  $\delta^{15}$ N values of macroalgae in the NMP are higher on shallow compared to deep reefs and reflect spatial variability in agricultural versus urban sewage N discharges. These case studies provide compelling evidence that macroalgal HABs are linked to land-based nutrient discharges and that bottom-up as well as top-down forces need to be considered to fully understand these phenomena.

ORAL

# Nitrate uptake kinetics of the toxic dinoflagellate *Alexandrium tamarense* in response to nitrate supply mode

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Nitrogen (N) is an essential variable controlling the phytoplankton production in marine environments. Toxic blooms are often associated with high nutrient concentration particularly nitrogen. N sources are not always supplied to the coastal ecosystem continuously, but also in pulses. The nature of the supply mode could affect the physiology and the development of harmful taxa. In the present study, dinoflagellate A. tamarense were supplied with nitrate continuously and at pulses at a range of concentrations, and examined the cellular nutrient uptake kinetic parameters (<sup>13</sup>C and <sup>15</sup>N) and physiology of cells in response to nitrate supply mode. Increasing the nitrate concentration induced an increase in growth rates. The availability of nitrate was observed to cause variability in the cellular nutrient status (C:N ratios) and pigments. Low uptake C:N ratios were observed when nitrate supply was limited, suggesting that the uptake C:N ratio was dependent on the supplied nitrate concentration. The cellular nutritional status might also cause variability in the uptake C:N ratio. Therefore, the nitrogen supply mode in coastal environments could play an important role in controlling the nutrient uptake of toxic dinoflagellate. The information on the N requirement of dinoflagellate due to the effect of supply mode could provide an estimate of the N required to sustain a massive bloom. This kind of information may also assist in coastal management in controlling bloom of toxic dinoflagellate and thus preventing a bloom from being fully developed.

# **POSTER – Session 1, Poster 11**

# Brackish stormwater detention ponds as promoters of HABs and eutrophication along the South Carolina coast

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In the rapidly urbanizing South Carolina coastal zone, intensive landscape maintenance and turf management are significant sources of nonpoint source pollutant loadings. The stormwater best management practice of choice in this region is wet detention ponds, the majority of which are brackish lagoons. Typically, stormwater is piped directly into the ponds, and their capacity for processing pollutants is limited. These highly eutrophic brackish ponds are "hot spots" for harmful algal blooms – over 200 blooms from 23 different species were documented over the last four years, many associated with measured toxins, fish kills or shellfish health effects. Because these ponds exchange with tidal creeks, they are sources for harmful algal bloom dispersion into adjacent estuaries. Furthermore, flux measurements indicate that groundwater acts as both a source of nutrients to the ponds and a mechanism for transporting nutrients from the ponds. These findings imply that manmade ponds as presently designed along the South Carolina coast may contribute to estuarine eutrophication and harmful algal bloom prevalence. A project is described that tests the use of constructed wetlands as a supplementary best management practice to process stormwater and groundwater nutrients prior to entering wet detention ponds.

# ORAL

Abiotic and biotic factors controlling a nutrient driven dinoflagellate bloom and likely responses to increased eutrophication

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The dinoflagellate *H. triquetra* forms dense (>10<sup>6</sup> L<sup>-1</sup>), nearly monospecific blooms in estuaries and coastal regions throughout the world. This species provides an ideal model for understanding the dynamics of nutrient driven wintertime estuarine phytoplankton blooms which are likely to increase in response eutrophication. Like toxic *Karenia brevis*, *H. triquetra* thrives when nutrient supplies are high (Steidinger et al. 1998). *Heterocapsa* is capable of taking up both inorganic

and organic nutrients, and can switch to mixotrophic nutrition if nutrient inputs decline. Like K. brevis, H. triquetra also adapts effectively to low light conditions, so that neither low ambient light conditions nor self-shading during a bloom significantly affects its growth. Heterocapsa's is a relatively large cell (30 µm) and in theory would be a poor nutrient competitor relative to small cells. What allows *H. triquetra* to compete is its strong swimming ability which allows it seek out regions of higher nutrient concentration and the fact that it blooms in the winter when grazing rates are low and small-sized competitors relatively rare. Because Heterocapsa is a strong swimmer and positively phototatic it concentrates in runoff driven frontal zones that form in winter. Similar circulation patterns are associated with the development of other HAB blooms including *Pseudo-nitzschia* spp. and *Alexandrium tamaranse*. It is only by understanding the common conditions conducive to algal bloom formation, and how those conditions are likely to effect the growth of specific species, that eutrophication effects on individual phytoplankton populations can be understood. If bloom-forming species analogous to Heterocapsa can sufficiently disrupt grazing, then they are likely to thrive in warmer months when hydrodynamic and nutrient conditions are premissive. Ultimately, the only difference as to whether the blooms are composed of toxic or nontoxic species may depend on stochastic events surrounding the availability of seed populations and how those the individual species respond to the entire suite of abiotic and biotic factors associated with eutrophication.

### ORAL

# Implementing the coastal module of the Global Ocean Observing System (GOOS): Toward rapid detection and timely predictions of harmful algal blooms

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The coastal module of GOOS is intended to develop an integrated and holistic approach to addressing six goals for the public good: (1) improve the capacity to detect and predict the effects of global climate change on coastal ecosystems; (2) improve the safety and efficiency of marine operations; (3) control and mitigate the effects of natural hazards more effectively; (4) reduce public health risks; (5) protect and restore healthy ecosystems more effectively; and (6) restore and sustain living marine resources more effectively. Although each goal has unique requirements for data and information, there are also many shared data and information needs. Likewise, many of the requirements for data management and communications are similar across all seven societal goals. Thus, an integrated approach to developing a multi-use, multi-disciplinary observing system is feasible, sensible, and cost-effective.

Through routine and continuous provision of reliable data and information, coastal GOOS will address these goals by enabling rapid and repeated assessments of the condition of coastal marine and estuarine systems and the development ecosystem-based management practices.

Ecosystem-based management is especially important in coastal marine and estuarine environments where the combined effects of habitat alterations, land-based sources of pollution, basin-scale changes in current patterns and hydrography, and global warming and sea level rise are most severe.

Harmful algal blooms affect public health and the capacity of marine ecosystems to support living resources and other goods and services. The frequency and magnitude of HABs can be enhanced by a variety of global scale human activities including nutrient pollution, overfishing, introductions of non-native HAB species, and the combustion of fossil fuels. However, the hypothesis that there has been a global increase in HABs is difficult to test given the mismatch between the time-space scales of observations and model requirements. Although there are specific locations where monitoring and modelling have detected trends, only continuous plankton recorder surveys and global climate models operate on the time-space scales required to test the "global spreading" hypothesis. Development of a global coastal ocean observing system will address this problem and provide data and information needed to control and mitigate the effects of HABs. The implementation strategy for such a system, recently completed by the Coastal Ocean Observations Panel of the IOC, will be described and progress toward achieving its recommendations assessed.

# ORAL

Modelling the contribution of prey deselection in the formation of harmful algal blooms

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A critical factor affecting the development of HABs is predation. Most obviously this functions at the level of ineffective predation on the HAB species. In addition, continuing predation on non-HAB species increases the availability of nutrients for the HAB species. The latter operates via two routes; the increased availability of 'new' nutrients (often supplied through eutrophication) and also through the regeneration of nutrients. The availability of nutrients and other environmental factors (e.g. light) affect growth rates and the nutrient status of phytoplankton. Because factors, such as toxicity and other anti-predator factors, often also develop in response to changes in environmental factors, so prey-selectivity must be considered as a variable depending on prey quality as well as quantity. As a consequence there is potential for the development of a positive feedback loop promoting the growth of HAB species. We explore this avenue using multinutrient (C:N:P) predator-prey models. Prey-selectivity functions typically employ preference indices that do not allow a true deselection of prey. We have developed a new prey selectivity function as a front end to zooplankton models that takes into account both prey quality and quantity, enabling the simulation of deselection of prey. The model also includes a link between the quality of ingested prey and assimilation efficiency, thus effecting nutrient regeneration and growth rates. Such a structure provides a flexible and

adaptable predator model. This predator model operates in conjunction with multinutrient phytoplankton models to demonstrate the potential consequences of variable prey selectivity and nutrient recycling for the development of HABs. Implications for the conduct of experiments and design of future models are also discussed.

### **POSTER – Session 2, Poster 2**

# Effect of N:P supply ratios on biochemical compositions and toxicity in dinoflagellate *Alexandrium tamarense*

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The influence of N : P supply ratios on cell abundance, cellular carbon (C), nitrogen (N) and phosphorus (P) contents, cellular protein, carbohydrate and lipid, and toxicity of dinoflagellate Alexandrium tamarense were studied. A. tamarense was grown in four different N : P supply ratios (N:P = 8, 16, 24 and 48) by keeping N concentration at 880  $\mu$ M with variable P concentrations without P limitation under 12 h light and 12 h dark cycle of 350 µmol m<sup>-2</sup> s<sup>-1</sup> irradiance at 17 °C. As N:P supply ratios increased, cellular C and N contents decreased but cellular P content increased. The C:N ratio which represents nutritional condition of cells did not showed any significant responses to N:P supply ratios, however cellular N : P ratio showed an inverse relationship with N:P supply ratios. The variability in protein, carbohydrate and lipid were considered to cause changes in cellular C, N and P contents. Cellular protein increased, however cellular carbohydrate decreased with increasing N:P supply ratios. Cellular lipid showed independent variation compared with those of protein and carbohydrate. The toxins (C2, GTX1, GTX3 and GTX4) also increased with increasing N:P supply ratios and these responses had strong relationships with cellular P and protein contents. The present study may suggest that the increases in toxicity of A. tamarense with increasing N:P supply ratio were influenced by the variability in biochemical compositions, particularly cellular P and protein contents without P limitation.

**POSTER** – Session 1, Poster 20

# Coastal nutrification following the passage of Hurricane Charley and it's relation to a subsequent *Karenia brevis* bloom on the West Florida Shelf

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Nutrient and chlorophyll a concentrations increased dramatically over ambient conditions following the passage of category 4 Hurricane Charley over the west Florida Shelf (WFS) and Charlotte Harbor in mid-August, 2004. Lack of power to operate lift stations and record rainfall sustained delivery of untreated storm and sanitary sewer water to the southwest Florida coastal waters, as rivers exceeded floodstage until the end of September. Chlorophyll a and inorganic nutrient concentrations following the hurricane passage were an order of magnitude greater than had been typically recorded on the WFS during the ECOHAB:Florida project for this period. A post-hurricane phytoplankton bloom consisting of a mixture of diatom species, primarily Rhizosolenia and Pseudo-nitzschia spp, was apparent one week after the passage of Charley, and persisted in the WFS region until mid October, when a small bloom of the Florida red tide dinoflagellate, Karenia brevis, was detected 20 km southwest of Charlotte Harbor. By early December the K. brevis bloom had moved south, off the Shark River, and intensified. Mortalities were first reported December 5<sup>th</sup> by lay observers and commercial fishermen and included fish, dolphins and stone crabs. The immediate effect of the delivery of nutrients to the West Florida Shelf from Hurricane Charley was thus the development of a large diatom bloom, with significant K. brevis detected only after the decline of this bloom. These data suggest that increased inorganic nutrient inputs to coastal waters of the WFS Florida are more likely to sustain diatom blooms in the short-term, and that hurricane derived nutrients were available to K. brevis only after regeneration.

# **POSTER – Session 1, Poster 3**

Harmful algae in Suffolk County (N.Y., USA) estuaries - a 30 year history

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A historical perspective can often reveal fundamental changes in ecosystem dynamics associated with population increase, and/or cultural transformations. This is certainly true in Suffolk County where small form blooms in the mid 20<sup>th</sup> century were associated with the presence of duck farms, and the onset of "brown tide" blooms (*Aureococcus anophagefferens*) in the latter part of the century was suggested to be related to historical agricultural practices, and meteorologically forced interannual variations in groundwater flow. While the small form

blooms fit our classical understanding of eutrophication, the appearance of brown tide initiated a paradigm shift in our conception of HABs and their mechanistic relationship to eutrophication.

Although the only evidence to date suggesting that Suffolk County estuaries have experienced public health effects, or massive fish kills related to harmful algal blooms, is circumstantial, the presence of several species, some occasionally in bloom proportion, imply at least the potential for such harmful blooms to occur.

HAB species encountered over the last thirty years, their effects on public health and ecosystem dynamics, and, as far as possible, the conditions associated with their presence or bloom formation, are discussed.

**POSTER – Session 1, Poster 18** 

# Toxin levels in the benthic cyanobacterium *Lyngbya majuscula* in relation to tissue nutrient content and bloom intensity

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The benthic nitrogen fixing cyanobacterium *Lyngbya majuscula* causes major blooms covering seagrasses, macroalgae, corals and other benthic substrates causing health, economic and ecosystem effects during large bloom events in many tropical and subtropical regions. *L. majuscula* contains a suite of toxins that have tumor promoters and can cause severe dermatitis and asthma like symptoms e.g. lyngbyatoxin a (LTA) and debromoaplysiatoxin (DAT). Studies were undertaken to ascertain the nitrogen dynamics of *L. majuscula* sampled in Moreton Bay, Australia. Levels of the toxin LTA measured in Moreton Bay were highest at peak biomass of blooms, and highest tissue C, N and P content, suggesting peak toxin production at maximum metabolic rate and 'health' of the bloom. *L. majuscula* is capable of very high rates of nitrogen fixation (as determined by acetylene reduction and <sup>15</sup>N<sub>2</sub> incorporation), with highest rates in initial phases of blooms. Tissue del-N values suggest this is a major source of N. *L. majuscula* is also capable of high uptake rates of both inorganic (NH<sub>4</sub>, NO<sub>x</sub>) and organic N (urea) compounds, indicating that this very successful bloom species is adept at utilizing transient nutrient supplies of various forms, in the variable estuarine and coastal environments where it exists.

**POSTER – Session 1, Poster 8** 

# Problems with ballast water exchange as a means of controlling movement of harmful algal species throughout the world.

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It is generally accepted that ballast water is a significant vector for potentially harmful species of phytoplankton and protists throughout the world, and has become an important driving force behind national and international legislation. Transoceanic transport of non-indigenous species was the focus of a February, 2004, United Nations (IMO) Convention. In 1996, the US Congress authorized the National Invasive Species Act, calling for ballast water management in the form of mid-oceanic exchange or retention of ballast water on board. In 2004 the U.S. Coast Guard recommended mandatory ballast water exchange or retention pending the development of effective ballast treatment technologies.

Shipboard tests of ballast water treatment systems employing UV disinfection and filtration aboard the Coral Princess on a September 2004 cruise from Victoria, BC, Canada to Fort Lauderdale, Florida via the Panama Canal, provided a unique opportunity to compare and evaluate the effectiveness of ballast water exchange. A 200 ton ballast tank was emptied in the Pacific Ocean prior to entering the Panama Canal. The tank was filled and flushed three times with freshwater from Lake Gaston in the Canal. The salinity of the filled ballast tank was measured and a detailed taxonomic inventory of the freshwater phytoplankton species was recorded. Later, on route between Cristobal, Panama and Aruba, the freshwater was discharged and the tank flushed and filled three times with Caribbean water. Measurement of the salinity in the ballast tank suggested an exchange efficiency of 99% based on end member salt concentrations.

The biological efficacy, however, as measured by an examination of the abundance of the phytoplankton taxa and species was much lower and ranged from 48-80%. Freshwater species were clearly evident among the species identified in the Caribbean water samples as well as species unique to and originating from the Pacific.

# **POSTER – Session 2, Poster 18**

#### The potential role of increased nutrient inputs to higher incidences of ciguatera in Hawaii

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Coral reef ecosystems are adapted to oligotrophic conditions and are therefore sensitive to nutrient loading. Ciguatera, a reef fish-borne food poisoning caused by bioaccumulated and

biomagnified toxins produced by the benthic dinoflagellate, *Gambierdiscus toxicus*, appears to be more prominent following disturbances to reef ecosystems, possibly including nutrient loading impacts. Ciguatera was not endemic in Hawaii prior to 1975, after which local reef fish were implicated in increasing numbers of ciguatera cases. Residents and legislatures of Hawaii suspected that accelerated coastal development in the 1970s and 1980s was somehow to blame. A comparison of historical and recent nutrient data from the island of Hawaii suggests that nutrient inputs have increased at (some) reef sites over the past 20 years, on the order of 100-200%. Analysis of data from monthly sample collections of nutrients and benthic dinoflagellates at five sites around the island of Hawaii indicates that *Gambierdiscus* abundance is significantly and positively correlated with nitrate concentrations of the overlying water ( $r^2 = 0.86$ , p = 0.02). These results suggest that 1) nutrient loading could be a factor in the increased incidences of ciguatera reported in Hawaii since 1975, and 2) oligotrophic ecosystems like tropical coral reefs may be susceptible to increased harmful algae impacts due to (relatively) large increases in nutrient loading, although low (i.e., not eutrophic) by impacted, temperate coastal standards.

# **POSTER – Session 1, Poster 9**

### HABs in western Australia: Expressions of eutrophication in a southern climate

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Over 110 waterbodies in WA experience some level of algal nuisance on a regular basis. Much of the agricultural clearing and urban development has occurred in the greater southwest where eutrophication is expressed as excessive macroalgal or microalgal accumulations. Management responses have mainly been focussed on estuaries due to the prevalence of massive bloom and in some cases complete ecosystem collapse. These estuaries are subject to extremely seasonal flow and irregular openings to the ocean. With a drying climate and less winter flow combined with increased demands for water abstraction there appears to be an increase in harmful species in both estuaries and streams. Fish kills appear to be increasing in frequency and extent. Cyanobacteria are a common occurrence in Australia and in WA have been the dominant problem species. Most spectacular has been the collapse of the Peel Harvey system eventually due to massive growths of Nodularia spumigena. Microcytis, Anabaena and Anabeaenopsis are common in fresh to brackish water bodies in the State. More recently Karlodinium micrum in the Swan and 2 other estuaries has received considerable attention associated with fish kills. It appears that the drying climate is leading to period of extended stratification favouring dinoflagellates. Pfistieria shumwavae and Lyngbya majuscula have also recently made appearances along with a range of other species (such as *Heterosigma* sp, *Procrocentrum* sp, Karenia sp and Dinophysis.

The Aquatic Science Branch manages a range of long term environmental monitoring and phytoplankton surveillance programs over a number of years employing 5 taxonomists among

other specialists and generating a wealth of data. Toxicity testing, toxin identification and identification beyond microscopic techniques is accomplished through collaboration with other Australian, New Zealand and US laboratories on an ad hoc basis. We are keen to establish more coherent and better focussed collaborations in answering many of the fundamental questions we have about the behaviour of these species.

ORAL

### Applications of an *in situ* water quality monitoring platform (MARVIN) for HAB research: A comparison of data collected in the St. Johns and Caloosahatchee River systems in Florida

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High frequency sampling is crucial to understanding the links between HAB development and nutrient inputs related to episodic rainfall events in major tributaries influenced by development and cultural eutrophication. MARVIN, (MERHAB autonomous research vessel for in situ sampling) is a monitoring platform developed by the Fish & Wildlife Research Institute in collaboration with AMJ Equipment to study HAB events in the St. Johns River, Florida. Near real-time transmission of physical (e.g. temperature, salinity, currents), biological (e.g. relative fluorescence), chemical (e.g. nutrients (NO<sub>2</sub>+NO<sub>3</sub>, PO<sub>4</sub>)) and meteorological data from the monitoring station to a public access web site is accomplished via a GOES satellite link. Discrete preserved phytoplankton samples are stored onboard for weekly collection. MARVIN has been successfully deployed for two years in the St. Johns River, an urbanized estuarine system in northeast Florida, to monitor conditions leading to cyanobacterial blooms. It's recent deployment in the Caloosahatchee River, a gated southwest Florida River heavily impacted by both development and periodic water releases from Lake Okeechobee, is permitting the continuous monitoring of water quality parameters in a river system with a suggested link K. brevis bloom development and maintenance in adjacent waters. The data from both deployments will be compared to examine the role of episodic nutrient delivery within these systems to HAB development and evaluate the utility of such monitoring platforms for HAB research.

# **POSTER – Session 2, Poster 10**

# Application of the Environmental Sample Processor (ESP) for remote detection of harmful algae and toxins they produce

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Molecular diagnostic procedures for identifying harmful algal bloom (HAB) species, their genes, gene products, and toxins play a central role in many research and resource management activities, but such methods generally require the return of discrete samples to a laboratory for As a step towards overcoming this impediment we are exploring use of the analysis. Environmental Sample Processor (ESP), a novel instrument that can detect remotely, subsurface, and in near real-time, a wide range of waterborne microorganisms and substances they produce (http://www.mbari.org/microbial/ESP). A key feature of the ESP is an ability to automate application of custom ribosomal RNA-targeted DNA probe arrays to detect particular species and transmit assay results via radio modem to a remote location for processing and interpretation. Two-way communication with the instrument allows the user to receive data and instrument status reports, as well as alter the ESP's sampling schedule, modify analytical protocols, etc. In addition to supporting real-time analyses, the ESP is also suitable for archiving discrete samples for microscopy, nucleic acid, and phycotoxin analyses after the instrument is recovered and returned to a laboratory. To date, the ESP has successfully automated application of several different classes of DNA probe arrays in single deployments lasting over 20 days that have targeted a variety of bacterioplankton, HAB species, and invertebrate larvae. We are presently developing a competitive ELISA technique to detect and quantify domoic acid for deployment aboard the ESP and use in concert with probe arrays and sample archive capabilities, thereby enabling the remote, integrated assessment of algal cell abundance and associated toxin levels. The ESP is scheduled to operate autonomously, in situ, in Monterey Bay, CA, for approximately 135 days between March-August '05. In this presentation we will review our progress to date and future plans.

#### ORAL

#### New approaches to understanding the role of dissolved organic matter in HAB dynamics

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Eutrophication is recognized as one of the important factors contributing to the geographical and temporal expansion of HABs. The amount, form, and timing of nutrient inputs can be important in determining the response of particular HABs to nutrient enrichment. While many HAB

species are strict autotrophs and use inorganic nutrients, other species have the ability to acquire nutrients via uptake of dissolved organic compounds (DOM). A considerable amount of the N and P inputs to coastal systems from watersheds is in the form of dissolved organic matter. However, DOM from land-based sources are comprised of complex mixtures of compounds that are largely uncharacterized due to past analytical limitations. This has hampered previous attempts to understand the role of watershed sources of DOM in HAB nutrition. In the current study, the response of Prorocentrum minimum to DOM from urban and agricultural watershed sources was compared to their response to inorganic nutrients. The biomass of P. minimum was more than twice as great when grown on DOM from either of the watershed sources compared to growth on inorganic nutrients (see poster by Sipler et al.).

Electrospray ionization mass spectrometry (ESI-MS) is a relatively new analytical approach to chemically characterize DOM compounds in the environment. ESI-MS provides compound level information on the large suite of compounds present, including molecular weight (unit mass resolution), functional group characteristics, and changes in concentration over time. ESI-MS was used to follow which compounds in the complex mixture of DOM from the land-based sources were used (or not used) by P. minimum, the rates of utilization of those compounds, and their chemical characteristics. Initial ESI-MS results from these experiments will be presented.

# ORAL

Ecosystem modelling of the NW European shelf seas towards the forecasting of harmful algal blooms

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The POLCOMS-ERSEM model combines the physics of POLCOMS, a 3D baroclinic circulation model developed by the Proudman Oceanographic Laboratory (POL), and the European Regional Seas Ecosystem Model (ERSEM) provided by Plymouth Marine Laboratory (PML).

The Medium Resolution Continental Shelf model is a 6 km resolution implementation of POLCOMS-ERSEM for the NW European Shelf. The model is driven at the surface by hourly winds and pressures and 3-hourly heat, light and moisture fluxes from the Met Office mesoscale weather prediction model. At the open ocean boundaries the model is forced with tidal elevations and velocities and temperature and salinity from the 12km POLCOMS Atlantic Margin Model. Riverine sources of physical and biochemical parameters are also included. Forecast runs from the weather prediction and Atlantic Margin models allow the running of the MRCS on a weekly basis in hindcast and 5-day forecast mode.

ERSEM, which has a proven pedigree in European coastal areas, uses a 'functional group' approach to describe the biota, which are classified according to trophic levels and subdivided on the basis of trophic links and/or size. Physiological processes and population dynamics are described by fluxes of carbon or nutrients between functional groups, and constrained by nutrient, oxygen and carbon biochemistry, giving a state-of-the-art description of ecosystem processes. This lower trophic level 'functional group' modelling is used, in combination with model output of physical and biogeochemical parameters, to provide maps of the risk of occurrence of harmful algae.

Information from the system is freely available on the Met Office website.

# **POSTER – Session 2, Poster 13**

# Dissolved organic matter concentration and characteristics during *Aureococcus anophagefferens* blooms in 2002 and 2003: A comparison

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Since their first reported occurrence in 1985, brown tide blooms caused by the pelagophyte Aureococcus anophagefferens have been observed in coastal bays in the northeastern and mid-Atlantic states of the USA. To explore potential links between the quantity and quality of watercolumn dissolved organic matter (DOM) and the initiation, development, and decline of brown tide blooms, we collected a two-year time-series of surface water samples from Chincoteague Bay, where A. anophagefferens blooms have been reported since 1999. Dissolved organic carbon (DOC) concentrations and UV/Visible light absorbance characteristics were measured on bulk (<0.2 µm) DOM samples. High molecular weight (>1kDa) DOM was characterized using isotope ratio mass spectrometry (IRMS) and direct temperature-resolved mass spectrometry (DT-MS). Our results show that springtime DOM pools in 2002, a drought year, and 2003, a wet year, differed significantly. During 2003, there were higher concentrations of >1kDa DOC and this fraction represented a larger proportion of overall DOC. In addition, the high-molecularweight DOC in 2003 appeared to be considerably more allochthonous, as indicated by  $\delta^{13}C$ values and DT-MS analyses. Despite the springtime differences in DOC concentration and composition, brown tide blooms developed during both years. During the three brown tide blooms sampled as part of this study, the composition of the initially dissimilar DOM became more similar as the population of Aureococcus anophagefferens increased. These results suggest that Aureococcus anophagefferens populations can develop under a range of DOM conditions and that DOM pools are altered as a result of blooms.

**POSTER – Session 1, Poster 13** 

# Environmental and behavioral influences on *Karenia brevis'* nitrate uptake: A bloom initiation scenario

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A near-bottom population of Karenia brevis, observed in a 22 m water column on the West Florida Shelf, may use sediments as a nutrient source in oligotrophic environments. We examined how prior environmental exposure influences K. brevis' physiology and if K. brevis may use diel vertical migration (DVM) as a nutrient retrieval mechanism. Experiments isolating environmental effects support the importance of light and prior nutrient exposure to nitrate assimilation. Cells exposed to nitrate depleted conditions for 12 hours enhance nocturnal uptake relative to those cells exposed to higher diurnal nitrate conditions. We examined nitrate uptake over K. brevis' DVM at three depths corresponding to light levels 350, 125 and 60 umol quanta  $m^{-2}$  sec<sup>-1</sup> using a vertically stratified mesocosm (the upper 2/3 contained < 0.5  $\mu$ M NO<sub>3</sub> and the lower 1/3 contained > 10  $\mu$ M NO<sub>3</sub>). Cell movement and biochemical state support that vertical migration is a mechanism for nutrient retrieval. Nocturnal uptake rates in the mesocom were intermediate between cells exposed to nitrate depleted conditions and nitrate replete conditions. By coupling adjustments in nocturnal nitrate uptake physiology with a nutrient retrieval behavior, K. brevis may optimize exposure to light during the day and use sediments as a nutrient source at night. The nocturnal uptake rates of nitrate are sufficient to maintain near bottom populations at growth rates of 0.3 div day<sup>-1</sup>. Undetected near-bottom K. brevis populations advected near shore may account for population aggregations that suddenly appear at the surface.

#### **POSTER – Session 2, Poster 14**

#### The role of natural DOM sources in *Prorcentrum minimum* growth dynamics

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Nutrients are one of the many factors considered to be important in Harmful Algal Bloom (HAB) growth dynamics. While some HABs depend on inorganic nutrients, others, including dinoflagellates and cyanophytes, can use dissolved organic matter (DOM) for growth. Knowledge of the nutritional requirements of HABs is essential to understanding the local and global distribution of HABs.

Several studies have explored the effects of specific dissolved organic nitrogen (DON) compounds on HAB growth. However, the role of natural DOM sources, which are comprised of

complex mixtures of uncharacterized compounds, have generally not been addressed. In this study, we examined the effects of land derived DOM sources from urban and agricultural areas on *Prorocentrum minimum* growth.

Preliminary data yielded positive results for the use of land-based DOM sources in bloom formation. *P. minimum* cultures had a higher biomass when grown on land-based DOM source waters than on inorganic nitrogen (controls). *P. minimum* grew better on urban DOM than agricultural DOM, per mole DOM supplied. Fast Repetition Rate Fluorometer (FRRF) measurements were taken to determine nutrient limitation. These findings suggest that DOM from land-based sources may contribute to the growth of *P.minimum* in coastal waters and that not all DOM sources are equally bioavailable to *P. minimum*.

# **POSTER – Session 1, Poster 14**

### Multidecadal changes in the diatom:flagellate ratio and Si:N and Si:P ratios in Narragansett Bay, and influence of Si:N supply ratios on diatom species competition

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The ratio of the mean annual diatom to flagellate abundance (numerical) in Narragansett Bay based on weekly sampling between 1960 and 1996 exhibited a pronounced multidecadal, recurrent oscillation. This functional group ratio progressively decreased five-fold, from ca. 5:1 to 1:1, then progressively recovered to ca. 5:1, thereafter followed by a similar decrease and recovery pattern. There were two distinct cycles during the 37-year time series exhibiting this pattern. Nutrient measurements begun in 1973, and detrended, also exhibited long-term patterns: mean annual phosphorus concentrations decreased and silicate concentrations increased, while inorganic nitrogen (NH<sub>4</sub> and NO<sub>3</sub>) concentrations were cyclical. The trends in the mean annual ratios of Si:N (range ca. 4.5 to 0.45) and Si:P (range ca. 20 to < 10:1) and the diatom:flagellate ratio were strongly and positively correlated, indicative of the importance of Si in regulating the functional group selection and long-term patterns observed. The Spearman r (non parametric) correlation coefficient between the trends in the diatom:flagellate ratio and the Si:P ratio was +0.79, and +0.71 with the Si:N ratio, both statistically significant. The interannual patterns of selected flagellate species representative of different nutritional modes that occurred within this functional group oscillation will be shown. The results of chemostat experiments with the three major diatom species in Narragansett Bay, conducted to examine the influence of Si:N supply ratios on their interspecific competition and species selection, will also be presented, and discussed from the perspective of the strengths and limitation of the nutrient ratio theory, and related current concepts of eutrophication, to explain harmful algal bloom species selection.

# ORAL

### Urease kinetics of several harmful algal species from the Chesapeake Bay, USA

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Urea  $(CO(NH_2)_2)$  is a source of nitrogen and carbon for marine bacteria and phytoplankton in marine environments. Many phytoplankton taxa utilize urea and many 'harmful' species have been observed to be stimulated by elevated urea concentrations. The ability of phytoplankton to use urea depends on the activity of the enzyme urease to catabolize urea into ammonium that is subsequently used by various biochemical pathways in the cell. Preliminary field studies in the Chesapeake Bay show that urease activity rates are highest in the summer and are positively correlated with dinoflagellate biomass. To investigate further possible relationships, we examined urease enzyme kinetics in five different phytoplankton species that have been designated harmful or toxic species in the Chesapeake Bay: Prorocentrum minimum (dinoflagellate), Heterocapsa triquetra (dinoflagellate), Karlodinium micrum (dinoflagellate), Storeatula major (cryptophyte; not toxic by itself but a prey of K. micrum), and Pseudo-nitzschia The non-harmful flagellate *Isochrysis galbana*, with previously observed high sp. (diatom). urease activity rates was included as a positive control species. All species were grown in f/20 nitrate, ammonium, and urea media and were analyzed during exponential growth for maximum urease activity rates  $(V_{max})$  and competitive ability at low urea concentrations  $(K_m)$ . S. major had the highest urease activity rates when grown on ammonium and urea (>1  $\mu$ g at N l<sup>-1</sup> hr<sup>-1</sup>), while *P. minimum* had the highest urease rates when grown on nitrate. *P. minimum* is a better competitor for urea when grown on ammonium and urea, indicated by having the lowest K<sub>m</sub> (<270 µg at N  $l^{-1}$ ) among the five species. *H. triquetra*, a winter species, had the highest K<sub>m</sub>. From this study, we hope to better understand how enzymes involved in nitrogen acquisition pathways play a role in ecological success of a given species under varying environmental conditions.

# **POSTER – Session 1, Poster 15**

# Harmful algal blooms in the context of major ocean initiatives

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The National Oceanic and Atmospheric Administration (NOAA) is engaged in, and leading many of the critical activities in ocean and coastal science and resource management. Several major ocean initiatives along with recent Congressional legislation frame NOAA's programs in monitoring, research, and forecasting of harmful algal blooms and coastal eutrophication. The U.S. Ocean Action Plan calls for U.S. agencies to build an Integrated Ocean Observing System (IOOS) as a fundamental contribution to the Global Earth Observing System of Systems (GEOSS), create a National Water Quality Monitoring Network, implement new legislation on Oceans and Human Health, and Harmful Algal Blooms and Hypoxia, and share expertise abroad. NOAA, in collaboration with other federal, state, and regional organizations, continues to support ocean observation system building, foster technology development, advance the understanding harmful algal bloom phenomena, and the impacts of eutrophication on coastal ecosystems. The integration of research and observations has lead to the development of operational ecological models and forecasts. Recent successes in harmful algal bloom and hypoxia forecasting demonstrate the benefits of integrated technologies and scientific collaboration. NOAA's vision is the integrated management of the oceans. The direction of U.S. ocean science and management programs is toward greater integration and collaboration, domestically and internationally.

# ORAL

# Freshwater flow and nutrients: Effects on top-down control of bloom-forming dinoflagellates

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The potential of zooplankton grazing to regulate dinoflagellate net population growth in a record drought (low nutrient) and a record wet (high nutrient) year in a mesohaline tributary of Chesapeake Bay (Choptank River) was investigated. During the record high freshwater flow year (2003), dissolved inorganic nitrogen and phytoplankton biomass in spring were  $\sim 10X >$ than in 2002, a record low flow year. No blooms occurred in spring 2002, but there were two dinoflagellate blooms in 2003. Species composition and abundance of micro and mesozooplankton differed significantly between the two years. In both years microzooplankton were the dominant source of grazing mortality for <25 micron dinoflagellates. Although populations of small dinoflagellates were low in spring 2002, potential community grazing coefficients averaged ~  $0.3 \text{ d}^{-1}$  for Karlodinium micrum and Prorocentrum cordatum. In spring 2003, microzooplankton biomass and community grazing coefficients were highly variable but averaged 2-3X those in 2002. Although average grazing coefficients were ~ 1 d<sup>-1</sup> (similar to the max. reported growth coefficient for most dinoflagellates), a bloom of Heterocapsa rotundatum (peak density ~ 4 X  $10^4$  cells ml-1) occurred in March and a bloom of *P. cordatum* (peak density  $\sim 2 \times 10^5$  cells ml<sup>-1</sup>) occurred in May 2003. We hypothesize that the spatial and temporal variability in grazing, coupled with the high nutrient levels, provided windows of opportunity for small dinoflagellate blooms in 2003.

# ORAL

### Iron induced development pathway of HABs community and its consequence on mitigation of eutrophication

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Three types of iron enrich experiments, in situ batch culture, semi- in situ "all-minus-one" nutrients enrichment combining dilution technique, simulation of the vertical migration cessation of mesozooplankton by adding extra stocks of mesozooplankton to the iron enrichment cultures after certain lag times, on phytoplankton community development was carried out in coastal of China Sea Waters. All these in vitro iron enrich experiments indicate that iron enrichments significantly promote the growth of the phytoplankton and change the communities structure. These iron enrich experiments indicated that the biomass of diatoms, especially the big or chain form ones, such as, Pseudo-nitzschia pungens, Guinardia flaccida, Chaetoecerso curvisetus, Thalassiosira routa, Skeletonema costatum, was promoted much faster, and diatom becomes the predominant species soon. Iron enrichments also promote the phytoplankton utilization of nitrate and phosphate, reduce the phytoplankton requirement of silicate. Consequently, the bioavailability of iron may influence the biogeochemical cycle of these macro-nutrient elements in the seawater. The "all-minus-one" nutrients enrichment experiments showed that the phytoplankton community growth was most promoted by iron addition in coastal waters. And, it was the major contribution by the iron pool of microzooplankton recycling. The mesozooplankton lag addition experiments indicated that the increasing mesozooplankton grazing pressure only delays the start time of phytoplankton exponential growth period, and the iron-mediated increases in algal biomass are not found to be prevented by the increasing grazing pressure. The above two types of experiment showed that zooplankton also play an important role in big-celled or chain-form diatom development in iron enriched phytoplankton community. This study suggests that iron induced phytoplankton growth may contribute significantly not only to the CO<sub>2</sub> drawdown caused by the faster sinking of carbon down to the deep waters, but also could be potential mitigation of eutrophication by sequestration of extra nutrients through faster sinking of these harmless big-celled diatoms.

# ORAL

# A conceptual model for ecosystem disruptive algal blooms: The interactive roles of eutrophication, algal toxicity, and limitation by nutrients and light

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Ecosystem disruptive algal blooms (EDABs) are occurring with increasing frequency and intensity in conjunction with increased eutrophication of coastal waters. Such blooms are often caused by toxic species that severely disrupt food web dynamics. Three EDAB species include Auereococcus anophagefferens and Aureoumbra lagunensis (brown tide species), and Chrysochromulina polyepsis. These species are all small-sized and well-adapted for growth at low nutrient and light levels. Because such blooms are often associated with eutrophication, one is left with a paradox: Why would the growth of low-nutrient species be stimulated by increased nutrient inputs? To solve this puzzle we note that algal blooms ultimately lead to resource limitation. Increased nutrients initially stimulate the growth of species (e.g., diatoms) capable of rapid growth rates, whose large size allows them to minimize grazing. As the bloom progresses these algae consume essential nutrients and/or attenuate light to limiting levels. This in turn selects for smaller phytoplankton that are capable of growth at low levels of limiting resources but are efficiently grazed by microzooplankton. Eventually a steady-state evolves in which algal growth and grazing rates roughly balance, and nutrient concentrations remain stable because nutrient loss from algal uptake is balanced by recycled inputs from zooplankton grazing. Such systems, however, are susceptible to disruption by small species adapted for growth at low light and/or nutrients that are also toxic or unpalatable to grazers. Low grazing rates allow these species to proliferate, and as the new bloom progresses, zooplankton grazers diminish through poisoning or starvation. The decrease in grazing pressure leads to even higher algal biomass and demand for nutrients, and less recycling. This combination causes even more severe nutrient limitation, which further selects for the EDAB species. Eventually this species may occur as a virtual monoculture with little transfer of nutrients and energy to higher trophic levels.

#### ORAL

# Assessing the validation of a preliminary *Karlodinium micrum* Nowcast model system in Chesapeake Bay and its tributaries: A framework for HAB Nowcasts and Forecasts

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A hybrid statistical-mechanistic model built on the NOAA/NOS approach for modeling the likelihood of encountering sea nettles was enhanced with neural network predictions for weekly Nowcasts of Karlodinium micrum bloom probabilities in Chesapeake Bay. A habitat model for the ichthyotoxic dinoflagellate K. micrum was formulated from the Maryland Department of Natural Resources long-term phytoplankton and water quality monitoring program data sets. A 3-dimensional hydrodynamic computer model (QUODDY) is used to provide near real-time simulations of salinity and temperature to drive the spatiotemporal phytoplankton relative abundance predictions (i.e. Nowcasts) throughout the year. A near real-time and real-time data source network of the Maryland Department of Natural Resources continuous water quality monitoring program and USGS flow monitoring drives the model hydrodynamics. A qualitative assessment of presence or absence for K. micrum in Maryland tidewaters during 2004 provided a good preliminary distributional replication of high and low bloom likelihoods during the year. Here we provide an overview of the modeling framework with a quantitative error assessment of phytoplankton monitoring results for the 2004 Nowcasts and insights into refining the preliminary model for improving predictions in Nowcast and Forecast modes for this and other HAB species.

### ORAL

# History of HAB monitoring in Maryland tidewaters: Monitoring, response, nowcasting and forecasting

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HAB monitoring in Maryland evolved from a longstanding routine water quality and phytoplankton assessment program with Maryland Department of the Environment, the Chesapeake Bay Program and Maryland Department of Natural Resources (MD DNR). Living resource events and human health issues in Maryland tidewaters linked with potentially toxic algal blooms in 1996-97 intensified interest and concern regarding HAB species for the State. An Aquatic Health Hotline maintained by the Maryland Department of Health and Mental Hygiene in cooperation with MD DNR was initiated in 1997. The hotline remains crucial to providing an avenue for citizen reporting of unusual habitat conditions for investigation under the State response network. The program provides a safety net to insure major HABs are identified and appropriate management actions taken. Management actions have involved response sampling to specific events, waterway, beach and precautionary shellfish bed closures and active communication with area academic researchers, managers and the media on events and issues. The HAB monitoring program has progressed to incorporate the use of new monitoring technologies to complement existing methods and include the use of genetic probes for algal identification (e.g. PCR), continuous monitoring stations, phycotoxin evaluations and worldwide-web water quality monitoring updates with an automated alert system. In addition, implementation and testing of a preliminary hybrid statistical-mechanistic model developed through federal and academic partnerships to nowcast the probability of blooms of the ichthyotoxic dinoflagellate Karlodinium micrum are underway. A framework for forecasting K. *micrum* and other HABs has been proposed. The forecast and habitat model tools are expected to aid managers planning and conducting sampling responses, protecting human health and enhancement of communication links with the public.

# POSTER – Session 2, Poster 8

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# Top down control and demise of a nutrient driven dinoflagellate bloom

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*Heterocapsa triquetra* is one of the most common bloom forming dinoflagellates found in estuaries and near shore regions around the world. The prevailing environmental conditions associated with wintertime blooms in temperate regions are largely the result of atmospheric forcing. Runoff following the rainfall events supplies nutrients critical for bloom initiation and development. Winter *Heterocapsa triquetra* blooms can reach chl *a* levels >100 mg L<sup>-1</sup> and cell densities between 1 to 6 x 10<sup>6</sup> L<sup>-1</sup> and account for as much as half the annual carbon production in shallow estuaries. In order to bloom, *H. triquetra* optimizes a suite of factors. The most important factors controlling *Heterocapsa*'s winter niche are low temperatures that limit micro- and macrozooplankton populations so that grazing losses are minimal. During

ecosystem disruptions (e.g. after hurricanes) *Heterocapsa* reappears in response to high nutrient concentrations and persists until the grazers are reestablished.

### **POSTER – Session 2, Poster 1**

### Nitrogen preference of fhe fish-killing flagellate Chattonella cf. verruculosa

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The species *Chattonella cf. verruculosa*, associated with extensive fish-kills in Rehoboth Bay, Delaware in 2000, and more recently in the New River, North Carolina, was examined for its preference in nutrient substrates for growth and terminal biomass. Blooms of *C. cf. verruculosa* commonly occurred in inland bays and estuarine regions where nutrient enriched waters are common. Clonal cultures of this species were studied at preferred salinities of 15 and 20 using media enriched with different nitrogen substrates. Nutrient preferences as expressed as most rapid growth (k) and maximal terminal density confirmed that *C. cf. verruculosa* grew well (>0.75 div·day<sup>-1</sup>) on various nitrogen sources and several forms of phosphorus as well. Single nitrogen sources in the form of ammonia and urea gave best growth with lower values than nitrate alone. When in combination, ammonia and nitrate did show moderate growth but the combination of ammonia and urea or urea alone gave the best growth patterns and highest terminal densities. Nutrient data from bloom sites confirmed the presence of both urea and ammonia in excess of nitrate. These studies strongly suggest that not only will the species of nutrients (nitrogen) influence growth of specific a HAB species but that combinations of them may have a synergistic effect in initiating and supporting blooms.

# ORAL

#### The research of the eutrophication status of East China Sea

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A series of four cruises were conducted during 2002-2003 at four seasons of a year in Changjiang estuary and adjacent coastal areas in which HAB frequently occur, in order to research of the relationship between eutrophication status of East China Sea (ECS) and Harmful Algal Blooms (HAB) occurrences.

On the basis of measurements of important nutrient substance, the inter-seasonal fluctuation and spatial distribution characteristics, the structure and the composition of these nutrients, the

regulating factors, and the ECS eutrophication state are discussed. The results show a clear characteristic that the nutrients' concentration decreased from inshore areas to open sea related to the river inputs. Maximum values of chlorophyll-a were typically observed at intermediate salinities and coincided with non-conservative decreases in nutrients along salinity gradient, indicating that depletion of nutrients was related to phytoplankton uptake. The seasonal fluctuations of nutrient concentrations were just opposite to that of chlorophyll, which indicated that the seasonal variations of nutrients were mainly controlled by phytoplankton uptake, whereas riverine inputs merely weakened or balanced the extent of the seasonal variations of nutrients. The estuarine and coastal waters was impacted by the Changjiang plume with high value of N/P ratio (>30), but rates of primary production were apparently not constrained by any kind of nutrient elements. However, the low SiO<sub>3</sub>/DIN ratios (<1) in most of the studied area might be linked with the rapidly increasing frequency of HAB incidents in recent years of the research area. As for the nutrient sources of the research areas, the nutrients fluxes of the Changjiang River, the exchange at the sediment-water interface and the wet/dry deposition of atmosphere are calculated. The flux of three sources is most typical of this pattern: Changjiang > Sediment > Atmosphere.

**POSTER – Session 1, Poster 2** 

Interannual variability of brown tide, *Aurecoccus anophagefferens*, blooms in the Maryland Coastal Bays

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The Maryland Coastal Bays have experienced recurring brown tide blooms, Aureococcus anophagefferens, since 1999 (and has been present since 1993). Bloom intensity varied temporally and spatially among sub-embayments with brown tide blooms occurring more frequently and at higher concentrations in the southern bays. Specifically, one area in the southern bays has had recurring Category three blooms (> 200,000 cells ml-1) while sites in the northern bays only reached Category two or three levels in 1999, 2002, and 2003. Seasonal cycles of dissolved organic nitrogen (DON), phosphorus (DOP) and carbon (DOC) were observed in the south with peaks typically occurring in July. An analysis of dissolved organic and inorganic nutrients was conducted across brown tide sampling areas and years to test the hypothesis that differences in nutrient levels may be driving the differences in bloom intensities. Although pre-bloom inputs of dissolved inorganic nitrogen (DIN) were generally greater in the northern bays, no significant relationships to brown tide densities were observed. DON was higher at the southern bay sites that had recurring blooms compared to other southern bay sites. Data from one southern site (Public Landing) suggest that A. anophagefferens is utilizing DON to bloom. An increase in DIN may have been a factor in the widely distributed bloom in Chincoteague Bay during 2003. Differences in flushing rates, climatic parameters (rain, wind) or zooplankton grazing may be the primary factors contributing to bloom formation among the MD Coastal Bay embayments.

**POSTER – Session 1, Poster 19** 

### The synergy of iron, copper and the toxicity of diatoms

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Outbreaks of toxigenic diatoms have caused massive marine mammal mortalities and closures of important fisheries. Here we show that toxigenic diatoms have an unusual ability to adapt to iron stress, and that this adaptation, regulated by the availability of copper, is linked to cell toxicity. The toxin, domoic acid, chelates both iron and copper and while toxin production is enhanced by iron deficiency, it is stimulated more by shortages of copper. Our findings suggest there is a synergistic linkage between iron and copper metabolism, whereby domoic acid release enhances copper acquisition that, in turn, facilitates cell access to otherwise unavailable organically complexed iron in seawater. This view is supported by field incubations of toxic diatoms from the Juan de Fuca eddy region off British Columbia, in which reductions of cell growth upon the addition of known siderophores is reversed with small amendments of Cu. Although common in prokaryotic organisms, high affinity iron uptake systems have not been reported for eukaryotic phytoplankton, and this unusual trait may separate toxigenic from non-toxic diatoms. We conclude that while excess macronutrients may elevate the biomass of potentially toxic diatoms, it is the secondary nutrients such as iron and copper that regulate the levels of cell toxicity and ultimately the impact of the bloom.

# ORAL

Volunteer HAB monitoring provides a "First Watch" for rresource managers and researchers in the Delaware Inland Bays, U.S.A.

Whereat, Edward, and Muns Farestad

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The Delaware Inland Bays Citizen Monitoring Program has been collecting water quality data since 1991 to augment monitoring efforts by the State in compliance with the Clean Water Act.

The program is managed by the University of Delaware Sea Grant Marine Advisory Service. In 2001, we began training volunteers to identify and enumerate living HAB cells in water samples using light microscopes. Since then, we have collected thousands of samples from the Inland Bays, many of which contained potentially toxic or bloom forming phytoplankton species. Regular reports of our observations are distributed by email to resource managers and HAB researchers. The Delaware Inland Bays are among the most eutrophic estuaries in the Mid-Altantic States. The most prevalent potentially toxic HABs that we see in the Inland Bays belong to the class Raphidophyceae: Chattonella cf. verruculosa, C. subsalsa, Heterosigma akashiwo and more rarely Fibrocapsa japonica. Over the past 4 years, blooms of Raphidophytes have been found in upper tributaries, particularly in residential canals, although their presence at low cell density is more widespread. Water from bloom locations has elevated nutrient levels, and in the case of residential canals, long residence times. Differences between species have emerged in terms of the bloom locations, phenology, and the ranges of temperature and salinity associated with blooms or mere presence in the water column. Volunteers have provided episodic sampling in response to blooms, and have routinely forwarded samples to University researchers for culturing, laboratory research, molecular probe development, and in the case of C. cf. verruculosa, toxin testing.

### **POSTER – Session 2, Poster 17**

#### A behaving drifter for simulating transport of mobile HAB organisms in nature

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We have developed a "Plankton Mimic", a behaving Lagrangian drifter that will help elucidate the biological/physical interactions in harmful algal blooms. This autonomous device senses its environment, dynamically adjusts its preferred depth, and swims toward that depth at a rate realistic for the organism being mimicked. Sensors include depth, temperature, salinity (conductivity), vertical velocity (both relative to the surface and relative to the surrounding water), light (PAR estimated from 4 color bands), and a real-time clock. From PAR and temperature data the mimic calculates photosynthesis, respiration and carbon pools. From a vertical concentration profile for nutrients, it can calculate uptake rates and nutrient pools. Using these pool values, it can estimate growth rate and doubling time. The device includes an ultrasonic pinger for tracking while submerged, and radio and LED beacons for locating it at the surface. An integrated GPS receiver allows the mimic to log its trajectory by taking position fixes during brief "pop-ups" to the surface. While at the surface it modulates the radio beacon signal with lat/lon data to announce its position and to facilitate recovery at the end of a deployment. Non-volatile memory (up to 256KB) allows logging of any selected suite of data at user-selected intervals. Rechargeable NiMH battery capacity is sufficient for 1-3 weeks of deployment. We intend to program mimics with several behavioral models (clock- and physiologically driven) to compare transport of mimics with that of real *Karenia brevis* blooms in the Gulf of Mexico.

#### **POSTER – Session 2, Poster 15**

#### Influence of monsoons and oceanographic processes on red tides in Hong Kong waters

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Hundreds of red tides have occurred in Hong Kong waters during 1983 to 2001, and show a clear spatial and temporal distribution. Most (74%) occurred in semi-enclosed northeast bays away from the Pearl River estuary, and fewer occurred in western estuarine waters. Most red tides (>70%) occurred between December and May, and fewer in summer. However, nutrients are high in the Pearl River estuary, whereas nutrients are generally low in the northeast bays and cannot support the high biomass of red tides. This suggests that the formation of red tides in Hong Kong waters is due to factors other than nutrients. Hong Kong waters in the northern part of the South China Sea are subjected to seasonal oceanographic processes due to monsoonal winds and Pearl River outflow. The prevailing NE monsoon winds in winter and spring result in downwelling and a longer residence time of waters in the NE bays, creating conditions similar to those of a batch culture, allowing vertical migration of phytoplankton in shallow waters to play a dominant role in favor of local red tides. This may be one of the reasons why more red tides occur in winter and spring in semi-enclosed waters. In summer, the SW monsoon winds result in upwelling along the coast, and high river discharge and rainfall cause an increased estuarine circulation in the Pearl River estuary and rapid outflow of the surface water from these semienclosed waters. As a result, residence time of these waters decreases, and they may be analogous to semi-continuous or continuous cultures. This may explain why there are fewer red tides in summer. The species occurring most frequently are (in descending order) Noctiluca scintillans, Gonyaulax polygramma, Skeletonema costatum, Mesodinium rubrum, Prorocentrum minimum and Ceratium furca. In general, dinoflagellate red tides occur mostly in April when Si is low, whereas diatom red tides occur in June when Si increases due to the freshwater discharge. Deep oceanic water on the continental shelf is drawn into the NE bays during the upwelling; it is poor in nutrients, and hence does not favour the strategy of vertical migration by dinoflagellates for the acquisition of nutrients in deep waters.

### **ORAL- INVITED**

#### Geographic distribution of Pfiesteria spp. and environmental factors

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Dual-gene (mtDNA-18S rDNA) Real-Time quantitative PCR assays were developed for Pfiesteria piscicida and P. shumwayae. After species-specificity and sensitivity were verified, these assays were used to investigate distribution of these two species from North Carolina to Maine in the east coast of the US, Hawaii, and Chilean and Chinese coasts. Both species were detected from most of the locations except in Hawaii and China, but occurred at <1.5 cells/ml for *P. piscicida* and < 10 (mostly <5) cells/ml for *P. shumwayae*. Sequence analysis confirmed the identity of the detected organisms and revealed genetic diversity. P. piscicida in Neuse River mostly occurred when water was 25-30 °C in temperature and 5-20‰ in salinity. In LIS, P. piscicida was detected more frequently in the nutrient-rich western Sound. P. shumwayae in Neuse River was detected less frequently than P. piscicida, and mostly at a temperature of 20-30 °C and a salinity of 5-20%. No correlation between *Pfiesteria* spp. abundance and chlorophyll concentration was found. Bioassays with Rhodomonas sp. addition for Neuse River samples elevated abundance of *Pfiesteria* spp. in some occasions. We conclude that 1) dual-gene PCR is useful in providing accurate estimation of Pfiesteria spp. abundances; 2) both Pfiesteria species are widely present but usually at low abundance; 3) control mechanisms of both Pfiesteria species may involve both bottom-up and top-down factors and no significant correlation between *Pfiesteria* spp abundances and nutrient or prev availability has been observed.

#### **POSTER – Session 2, Poster 4**

# A comparison of nutrient effects on the growth of *Chattonella subsalsa* and *Heterosigma akashiwo* (Raphodophyceae) isolated from the Inland Bays, Delaware (U.S.A)

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During the past several years, novel blooms of the toxic raphidophytes *Chattonella* and *Heterosigma* have caused fish kills and raised concerns for human and ecosystem health in the highly eutrophied Inland Bays of Delaware, USA. We compared the effects of nitrate, ammonium, dissolved organic nitrogen and phosphate availability on the biomass yield and growth rates of unialgal *Chattonella subsalsa* and *Heterosigma akashiwo* cultures isolated from the bays, using classic Monod-type experiments. *C. subsalsa* was unable to grow on urea, but *H. akashiwo* can use this organic N source. Both are capable of growing on glutamic acid. Experiments varying nitrate to phosphate ratios suggest that neither species grows well at ratios below Redfield values. Maximum nutrient-saturated growth rates ( $\mu_{max}$ ) for *C. subsalsa* ranged from 0.6 to 0.7 d<sup>-1</sup> and half-saturation concentrations for growth (K<sub>1/2</sub>) were 8  $\mu$ M for nitrate, 3  $\mu$ M for ammonium, and 0.7  $\mu$ M for phosphate.  $\mu_{max}$  of *H. akashiwo* was slightly higher than

*Chattonella* (0.8 to 0.9 d<sup>-1</sup>), but  $K_{1/2}$  values were nearly an order of magnitude lower at 0.9  $\mu$ M for nitrate, 0.1  $\mu$ M for ammonium, and 0.01 $\mu$ M for phosphate. Our results suggest that the smaller *Heterosigma* is much better adapted to grow at low nutrient levels, and the larger cells of *Chattonella* may have a competitive advantage only under highly eutrophic conditions such as those currently found in the Inland Bays.

#### **POSTER – Session 1, Poster 16**

## Which is the trigger factor to the outbreak of large scale *Prorocentrum* blooms in the East China Sea?

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For at least successive 7 years, *Prorocentrum donghaiense* produced large scale blooms in spring covering areas from 2,000 to 10,000km<sup>2</sup> in the East China Sea. Several cruises, consisting of profile stations and towing multi-parameter measurements were carried out to investigate the dynamics of the bloom and the ecological and oceanographic factors which could be related to the formation of the bloom. The results showed that the causative species, low-nutrient tolerant *Prorocentrum donghaiense*, spent at least 30 days growing at the sub-surface layer before forming large scale blooms at the sea surface. The blooming area was very well fit with the plume front and upwelling front outside the Yangtze River estuary and along the east coast of Zhejiang province, where the intensifying Taiwan warm current, Yangtze River diluted water and the weakening coastal current met to form the fronts along a line of 30-50m water depth, a sudden change area of sea bottom topography. The co-existence of warm water mass and stratified structure of temperature, salinity and Chlorophyll-a, and rapid change of phosphorus levels in early spring in this area supported the hypothesis that both physical and chemical processes played important roles in the forming of this large scale *Prorocentrum* blooming.

### **ORAL-Invited**

Nutrient regulation of toxin production: Comparison of hemolytic activity of *Amphidinium carterae* and *Amphidinium klebsii* 

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A recently emerging topic of interest within HABs is the production of hemolytic toxins by marine microalgae. Hemolytic toxins, resulting in disruption of gill membranes and blood

vessels in fish, are but one of many ichthyotoxic compounds. *Prymnesium parvum*, known to cause massive fish kills in Texas and North Carolina, produces a suite of toxins, including the extremely hemolytic prymnesins. Toxin activity of the prymnesins can be influenced by the environmental variables in which the species is grown. The dynamics of these toxins and the role they play in the environment are still being evaluated. *Amphidinium carterae* and *Amphidinium klebsii* are two unarmored, benthic dinoflagellates that are distributed worldwide and produce hemolytic compounds. Recently isolated clonal cultures of both species were used in experiments designed to determine the effect of nutrient limitation on toxin expression. These experiments employed nitrogen, phosphorous limited, and nutrient replete growth media. Hemolytic activity was quantified using the erythrocyte lysis assay, performed in the 96 well plate format and measured with a Biotek microtitre plate reader. Preliminary studies show that nutrient availability affects the hemolytic activity in *A. carterae* and *A. klebsii* and that the hemolytic activity varies between the two species.

#### **POSTER – Session 1, Poster 10**

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