Regional Meeting on Future Focus and Cooperation in HAB Research

2nd ASIAN GEOHAB MEETING

Institute of Oceanography Vietnamese Academy of Science and Technology

28 January-1 February 2008

ABSTRACT BOOK









Scientific Committee on Oceanic Research

INSTITUTE OF OCEANOGRAPHY HABVIET / IOC-DANIDA

Training and research activities of the cooperative HABViet Project, 1998-2008



Photo 1. The new ION Plankton Laboratory was built thanks to overhead funds of the HABViet Project . *Photo 2.* Training course on biology and taxonomy of toxic algae. *Photo 3.* HABViet-ION group at a workshop in September 2007. *Photo 4.* PhD student and technician working with SEM at University of Copenhagen, Denmark. *Photo 5.* Identifying phytoplankton species in the lab at ION. *Photo 6 & 7.* Field trips. *Photo 8.* National symposium on HABs and related problems in 2005.

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Organized by Scientific Committee for GEOHAB (GEOHAB SSC) and WESTPACT /HAB in Cooperation with HABViet III Project /Danida-ENRECA Programme

HABViet Project

REGIONAL MEETING ON FUTURE FOCUS AND COOPERATION IN HAB RESEARCH

28 January – 29 January 2008 Institute of Oceanography, Nha Trang, Vietnam

January 28 (Monday), 2008

- 08.00-08.30 Registration
 08.30-09.40 Welcome addresses by: Dr. Pham Van Qui (Vietnamese Academy of Science & Technology), 10' Dr. Le Xuan Than (Local government - People Committee of Khanh Hoa Province), 10' Dr. Pham Huyen (MOST), 10' Dr. Bui Hong Long (Institute of Oceanography), 20'and Mr. Henrik Enevoldsen (IOC-UNESCO), 10' Prof. Ken Furuya (Asian-GEOHAB), 10'
- 09.40-10.00 Giving the Campaign Medals to: Prof. Jacob Larsen Mr. Henrik Enevoldsen

Chairperson: Per Andersen

- 10.00-10.30 *Lam Nguyen-Ngoc, Hai Doan-Nhu, J. Larsen, and H. Enevoldsen.* The occurrence, taxonomy and impact of toxic and harmful microalgae in Vietnamese coastal waters what we have learned during 10 years of cooperative research
- 10.30-10.50 *Chu Van Thuoc*. Seasonal variations in cell density of genus <u>Pseudo-nitzschia</u> (Bacillariophyceae) in coastal waters of North Vietnam
- 10.50-11.20 *Coffee break*

Chairperson: Lam Nguyen-Ngoc

11.20-11.40	Zongling Wang, Ruixiang Li, Mingyuan Zhu, Yanju Hao.
	The interspecific competition between two HAB species: -
	Prorocentrum donghaiense and Alexandrium tamarense
11.40-12.00	Nguyen Thi Thu Lien. Toxic cyanobacteria in some
	freshwaters in Thua Thien-Hue province, Vietnam: -
	morphology, toxins and molecular approaches
12.00-12.20	Walker Smith. Phaeocystis as a HAB Species: Controls on
	Colony Formation
	-

12.20-14.30 Lunch break

Chairperson: Jacob Larsen

- 14.30-14.50 *Jinhui Wang, Ren Xu, and Xiuqing Huang.* Which cause the different geographical distribution and bloom formation of harmful algae?
- 14.50-15.10 *M.A. Burford, A. J. Posselt, and K. R. O'Brien.* Why does the toxic cyanobacterium, <u>Cylindrospermopsis</u> raciborskii dominate in so many reservoirs in Australia?
- 15.10-15.30 *Hai Doan-Nhu and Lam Nguyen-Ngoc*. Phytoplankton communities before, during and after a <u>Phaeocystis globosa</u> bloom in the South Central Vietnam
- 15.30-15.50 *Qi Yuzao, Liu Jieshen, Wang Yan, Li Yang, Ou Linjian, Yang Weidong, and Lu Songhui*. <u>Phaeocystis globosa</u> Scheffel blooms on the coastal China and its impacts
- 15.50-16.00 Coffee break
- 16.00-18.00 Snack party

January 29 (Tuesday), 2008

Chairperson: Yoon Lee

09.00-09.20 Ann Anton, Normawaty Mohammad Noor and Sitti Raehanah Muhd Shalleh. Recent harmful algal blooms (HABs) in Sabah, Malaysia

09.20-09.40	Dang Diem Hong, Dao Viet Ha and Yasuwo Fukuyo.
	Application of molecular biological techniques for
	research of Harmful algal blooms in Vietnam
09.40-10.00	Duong Thi Thuy, Dang Hoang Phuoc Hien, Nguyen Sy

- Nguyen, Dang Dinh Kim, Tran Van Tua, Dang Thi Thom and Jens Dahlmann. Toxic cyanobacterial blooms and their toxins in some North Vietnam water bodies
- 10.00-10.20 *F.P. Siringan, R.V. Azanza, C.P.C. David, J.M. Reotita, M.Y.Y. Sta. Maria and P.B. Zamora.* Transport of <u>Pyrodinium bahamense</u> var. <u>compressum</u> in Malampaya Sound, Palawan, Philippines
- 10.20-10.40 Coffee break

Chairperson: Vera Trainer

- 10.40-11.00 *Patricia Glibert and Ji Li*. Nitrogen uptake rates during the HAB events of 2005 in East China Sea and comparisons with recent Dinoflagellate Blooms on the Western Florida Shelf
- 11.00-11.20 Donald M. Anderson, Bruce A. Keafer, Kerry Norton, Dennis J. McGillicuddy, Ruoying He, Cindy H. Pilskaln, Darcie Couture, and Jennifer L. Martin. <u>Alexandrium</u> fundyense cyst dynamics in the gulf of Maine
- 11.20-11.40 *Dao Viet Ha, Shigeru Sato, Yasuwo Fukuyo, and Masaaki Kodama*. Toxic aquatic organisms from Vietnamese coastal waters
- 11.40-12.00 N. Mohammad-Noor, N, Moestrup, Ø., Daugbjerg, N., Caillaud, A. Caňete, E., Paz, B., Mallat, E., Franco, J.M., Diogène, J. and A. Anton. . Marine epibenthic dinoflagellates from Malaysia – Species diversity and future studies
- 12.00-12.15 Group photo
- 12.15-14.00 Lunch break

Chairperson: Doan Nhu Hai

14.00-14.20 *Lee, Y, YS Kang, WA Lim, KH Ahn, YT Park, and HM Bae.* Bloom dynamics of <u>Cochlodinium polykrikoids</u> in Korea coastal waters in 2007

- 14.20-14.40 *Geumog Myung, Hyung S. Kim, Jong S. Park, Myung Gil Park, Sunju Kim, and Wonho Yih.* Mixotrophic ciliate <u>Myrionecta rubra</u> needs certain type of cryptomonad prey to form a massive bloom
- 14.40-15.00 *Zongling Wang, LI Ruixiang, and Mingyuan Zhu.* The Distribution and HAB of <u>Phaeocystis globosa</u> in coastal water of North China
- 15.00-15.20 Coffee break
- 15.20-15.40 *Ruixiang Li, Mingyuan Zhu and Zongling Wang.* Study on HAB in coastal water of Qingdao City
- 15.40-16.00 Vera L. Trainer, William P. Cochlan, Charles G. Trick, Mark L. Wells. The PICES Harmful Algal Bloom International Project
- 16.00-16.30 Discussion
- 1800-20.00 Dinner Party

GEOHAB Global Ecology and Oceanography of Harmful Algal Blooms

SECOND ASIAN GEOHAB MEETING

31 January-1 February 2008 Institute of Oceanography, Nha Trang Vietnam

31 January (Thursday), 2008

Chairperson: Hakgyoon Kim

9:00–9:20	Ken Furuya. Background and overview of the 1st Asian
	GEOHAB Meeting, Tokyo March 2007
9:20–9:50	Patricia M. Glibert. Overview of the GEOHAB Core
	Research Project on HABs in Eutrophic Systems
9:50-10:10	Kazumi Matsuoka. Dinoflagellate cysts as a proxy of
	eutrophication in coastal waters

10:10–10:30 Coffee break

Chairperson: Ming Jiang Zhou

- 10:30–10:50 *Changkyu Lee and Yoon Lee*. Bloom dynamics of harmful algae, <u>Cochlodinium polykrikoides</u> in Korean coastal waters
- 10:50–11:10 Hakgyoon Kim, Changkyu Lee, Yangsoon Kang, Wolae Lim, Sookyang Kim, Youngtae Park, Jeongmin Shim, Kyongho An, Samgeun Lee, Heonmeen Bae and Yoon Lee. Joint EASTHAB and Asian GEOHAB works to clarify the likely route of <u>Cochlodinium polykrikoides</u> in Kuroshio current system
- 11:10–11:30 *Douding Lu, Dedi Zhu, Yuanfen Wang, Songhui Lu and Yuzhao Qi.* Population dynamics of targeted species with special emphasis on <u>Prorocentrum donghaiense</u> Lu in Zhejiang coastal water, China
- 11:30–11:50 *Songhui Lu and Zhengfeng Li*. The eco-physiological studies of phosphorus on the growth of <u>Karenia</u> <u>mikimotoi</u>

11:50–14:00 Lunch break

Chairperson: Kazumi Matsuoka

- 14:00–14:20 Ken Furuya. Green Noctiluca scintillans, successful red tide species in SE Asian waters
 14:00–14:20 Rhodora Azanza. Harmful algal blooms in Manila Bay, Philippines: the Pyrodinium bahamense var. compressum period and the Noctiluca scintillans period
 14:40–15:00 Thaithaworn Lirdwitayaprasid. Eutrophication and Noctiluca red tide in the upper Gulf of Thailand
 15:00–15:20 Paul J. Harrison, Jie Xu, Alvin Ho and Kedong Yin. Why
- are HABs not as bad in Hong Kong waters as expected?
- 15:20–15:40 Coffee break

Chairperson: Rhodora Azanza

- 15:40–16:00 *Gires Usup and Asmat Ahmad.* Dinoflagellateassociated bacteria: is there a common theme and could it be a subject for a regional project?
- 16:00–16:20 *Po-Teen Lim, Chu Van Thuoc, Nguyen Thi Minh Huyen, Atsushi Kobiyama, Ryuichi Sakai, Yoshinobu Takata, Shigeru Sato and Takehiko Ogata.* Growth and toxin production of dinoflagellate <u>Alexandrium minutum</u> (Dinophyceae) isolated from aquaculture pond in northern Vietnam
- 16:20–16:40 *DanLing Tang, JiuJuan Wang and YQ Chen.* Oceanography analysis of annually variation of harmful algal /phytoplankton blooms in the western South China Sea
- 16:40–17:00 *Robin Raine*. On the prediction of harmful algal events

February 1 (Friday), 2008

Chairperson: Robin Raine

9:00–9:20 *Jacob Larsen*. Toxic epiphytic and benthic dinoflagellates

- 09:20–10:40 Introduction to Urea dumping and other issue of relevance to future research
- 10:40-11:00 *Coffee break*
- 11:00-12:30 General discussion and wrap up
- 12:30-14:00 *Lunch break*
- 14:00-16:00 GEOHAB core Research on Eutrophication

Part 1

Regional Meeting on Future Focus and Cooperation in HAB Research

The occurrence, taxonomy and impact of toxic and harmful microalgae in Vietnamese coastal waters – what we have learned during 10 years of cooperative research ?

Lam Nguyen-Ngoc¹, Hai Doan-Nhu¹, Jacob Larsen² Henrik Enevoldsen², and Per Andersen³

¹Institute of Oceanography, 01 Cauda, Nhatrang City, Vietnam ²IOC Science and Communication Centre on Harmful Algae, University of Copenhagen, Øster Farimagsgade 2D, DK-1353 K, Copenhagen, Denmark ³Orbicon A/S, Jens Juuls Vej 18, 8260 Viby J, Denmark

Research on potentially harmful microalgae in Vietnam has during the last decade generated significant results and knowledge. This has been achieved through both national and international research activities, in particular the HABViet Project via the IOC contributed through its three phases from 1998 to 2008. The occurrence of potentially harmful species is widespread along the VN coast. A total of 70 species are recorded with distribution and seasonal abundance. Hereof one species was new to science and other two were renamed and emended. A guide book for identification of HAB species has been published and a phytoplankton database has been established. This is the first time long-term data sets for phytoplankton have been compiled in Vietnam. Vietnam has a flourishing aquaculture industry as well as harvesting of natural shellfish populations. The journal 'Vietnam Economic News' lists seafood products as the fourth most important export goods, and the value is increasing according to the Vietnamese Ministry of Fisheries. Important export markets require compliance to regulations for toxic algae and biotoxins, and the research has underpinned establishment of routine monitoring programmes. Knowledge has also been acquired about autecology of selected toxic species as well as phytoplankton community dynamics and examples of these results will be presented. Fresh water toxic cvanobacteria also present a major challenge and expertise for screening for toxicity and identification of species has been established. In parallel to the strengthened research expertise and network on harmful algae, there has been a focused strengthening of research facilities, MSc and PhD education, as well as broader training through research. Vietnamese scientists are looking forward to continue and expanded international cooperation. Areas of future research focus will be outlined.

Seasonal variations in cell density of genus Pseudo-nitzschia (Bacillariophyceae) in coastal waters of North Vietnam

Chu Van Thuoc¹, Nguyen Thi Minh Huyen¹, Pham The Thu¹, Le Thanh Tung², and Ton That Phap³

¹Institute of Marine Environment and Resources, 246 Da Nang Street, Hai Phong City, Vietnam. ²Research Institute for Marine Fisheries, 170 Le Lai Street, Hai Phong City, Vietnam. ³College of Sciences, Hue University, Hue City, Vietnam.

During 2004-2005, the national project on harmful algal blooms in the coastal concentrated fisheries culture areas of Vietnam (No. code KC.09.19) had been carried out. In the scope of this project, some sampling sites represented for culturing areas of bivalve (clams, green mussel), shrimps and fish cages in the coastal waters belonged to Hai Phong, Thai Binh and Thua Thien Hue provinces (North Vietnam) had been selected to monitoring harmful microalgae by monthly. The studied results showed that in the studied area the diatom *Pseudo-nitzschia* had dominated and obtained the highest density of cell in spring (from January to March 2005) mainly. In addition, at some sampling sites they were also common in August and October but their cell densities were lower than that in spring. In generally, the cell density distribution of Pseudo-nitzschia in the natural water bodies such as estuaries, lagoons was higher than that in the shrimp ponds. A possible relationship between the change of Pseudo-nitzschia cell density and some environmental factors was also discussed in this report.

The interspecific competition between two HAB species: -*Prorocentrum donghaiense* and *Alexandrium tamarense*

Zongling Wang, Ruixiang Li, Mingyuan Zhu, Yanju Hao

First Institute of Oceanography, State Oceanic Administration, Qingdao 266061, P.R. China

We studied the population dynamics and interspecific competition of *Prorocentrum donghaiense* and *Alexandrium tamarense*, two common HAB species in East China Sea, by using semi-continuous dilution experiments. The results show that *Prorocentrum donghaiense* is the winner in low phosphate concentration that it can completely exclude *Alexandrium tamarense*. However, at high phosphate condition, the competitions of the two species have different results, excluding each other or coexistence. The results also show that the competition results depend on the cell densities that the two species can reach in bi-culturing experiments.

Toxic cyanobacteria in some freshwaters in Thua Thien-Hue province, Vietnam: - morphology, toxins and molecular approaches

Nguyen Thi Thu Lien

Section of Biotechnology, Department of Biology, College of Sciences, Hue University, Vietnam

In our survey of potential toxic cyanobacteria in some freshwaters in Thua Thien-Hue province, Vietnam, we found out five toxic species with a new microcystin-producing species that named *Annamia toxica* gen. et sp. nov. Other microcystin-producing species are *Microcystis aeruginosa*, *M. botrys* and *M. panniformis* and one cylindrospermopsin-producing species is *Cylindrospermopsis raciborskii*. Toxin-producing ability of these species were confirmed by HPLC, ELISA and the presence of the genes involved in the toxic synthesis. This paper presents the results of morphology, toxicity and molecular characteristics of these toxic cyanobacterial strains.

Phaeocystis as a HAB Species: Controls on colony formation

Walker Smith

Virginia Institute of Marine Science, College of William and Mary, Glouceser Point, VI, USA

Phaeocystis is a cosmopolitan species, occurring in a variety of habitats, and has recently been implicated as a harmful algal species in Asian coastal systems. It affects local fisheries by accumulating in fish cages and depleting oxygen, exhibits hemolytic properties, and apparently is increasing in frequency. The bloom-forming species have a multiphasic life cycle, including solitary cells as well as large colonies, but the colonial stage is the one that forms massive blooms. Therefore, understanding the environmental factors that regulate colony formation are essential to predicting the appearance of *Phaeocystis* blooms in coastal waters. Our results suggest that a number of factors contribute to colony formation, including nutrients, nutrient ratios, and grazing. The importance of each of these in the dynamics of harmful algal blooms is discussed.

Which cause the different geographical distribution and bloom formation of harmful algae?

Jinhui Wang^{1,2}, Ren Xu¹, and Xiuqing Huang¹

 ¹East China Sea Environmental Monitoring Center, SOA, Dongtang road 630, shanghai, 200137, P.R. China
 ²School of Environmental Science and Engineering, Shanghai Jiao Tong University, Shanghai, 200240, P.R. China

Some new species formed bloom in differen part of China which never species introductions including been found before, issues of anthropogenic sources (e.g. ballast water) or natural systems (e.g. species range extension) will be analyzed based on geographical distribution and long series monitoring data. Large scale blooms of prorocentrum were found in East China sea for several succesive years since 2000 but not in north part and south part of Chinese coast, some other algaes such as Phaeocystis globosa, Heterosigma akashiwa and Chattonella marina formed bloom in South China sea and North part of Chinese coast but not in East China sea which is most eutrophic sea area in China. Which cause the different geographical distribution and bloom formation of harmful algae? The eutrophication condition, nutrient compositon(e.g. N/P ratio), relationship between oceanographic processes and HAB formation (ex. How the physics of nutrients, trace metals tie into bloom formation), global warming caused species range extension will be discussed, the physiological and ecological features of harmful bloom algae (e.g. nutrients absorb efficiency, storage of nutrient) may be another important factor

Why does the toxic cyanobacterium, *Cylindrospermopsis* raciborskii dominate in so many reservoirs in Australia?

M.A. Burford¹, A. J. Posselt¹, and K. R. O'Brien²

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The cyanobacterium, *Cylindrospermopsis raciborskii* has become a key water quality issue in many reservoirs in Australia. There are reports of the species dominating more and more waterbodies throughout the world, and it appears to be expanding it's range from tropical and subtropical areas, to temperate areas. Australian strains vary from those in other countries because it produces the toxin, cylindrospermopsin. Our studies have examined factors responsible for *C. raciborskii* dominance in subtropical southeast Queensland reservoirs. Blooms typically develop in summer months when water temperatures are highest. Our studies suggest that this species gains a competitive advantage by it's ability to effectively utilise a wide range of light regimes, and a superior phosphorus scavenging and storage capability. This contrasts with other reports suggesting that the competitive advantage is due to the capacity to fix nitrogen. Given the adaptability of this species, management control is therefore likely to remain a challenge.

Phytoplankton communities before, during and after a *Phaeocystis globosa* bloom in the South Central Vietnam

Hai Doan-Nhu and Lam Nguyen-Ngoc

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The blooms of haptophyte *Phaeocystis globosa* has scientifically been record since 2002 when causing seriously damage to intertidal flora and fauna in south Central Vietnam. The blooms have then record often in 2005, 2006 and 2007 with the association with upwelling phenomenon at the area. The effects of upwelling on selective growth of phytoplankton has been remain a big mystery. This present study partially try to discover the succession of phytoplankton under affect of this oceanography phenomenon. In September 2007, the Phaeocystis occupied entirely phytoplankton community with low species number recorded and Shannon diversity index was extremely low (0.09, log2). The stations with more abundance of Phaeocystis was 65. 2 % different from stations less dominant of Phaecocystis in species composition. Dinoflagelates were the second contribution to abundance of phytoplankton communities in the less dominant Phaeocystis stations and diatoms were in the place for the dominant *Phaeocystis* stations. During the pre southwest monsoon month (June 2007) the phytoplankton species were recorded much higher (78 species, range from 61 to 99 species/station) with higher biodiversity (H'=2.99). Abundance central to penal diatoms ratio (C/P) is low in the southern transects (4) compare to northern transects (13-228). During the bloom event in September 2007, after the serious period, the plankton communities have gained their species richness but the structure has been much modified. The C/P ratio was averaged to 7 and at most station it was close to 1. Community analysis showed the most southern transect was more stable while the northern transects with bloom of Noctiluca and middle transects with some domination of *Phaeocystis* were less stable. Dissimilarity percentage on species contribution among the transects were greatly varied, and they all some 80 - 96% different to station with the most abundance of Phaeocystis.

Phaeocystis globosa Scheffel blooms on the coastal China and its impacts

Qi Yuzao^{1,2} Liu Jieshen¹ Wang Yan^{1,2} Li Yang² Ou Linjian¹ Yang Weidong¹, and Lu Songhui^{1,2}

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In recent years Phaeocystis globosa blooms along the China's coast have formed and caused substantial fish kills in aquaculture cages and economic losses to the local economics. The blooms are relatively predictable, in that they repeatedly occur in one location each year. On unique feature of the colonies is their size: they reach up to 3 cm in diameter, a full order of magnitude larger than any other known phytoplankter. As such, the colonies are subject to different physical features, and aspects of their nutrient uptake, cell/colonial physiology and carbon nutrition, life history, and ecology must be much different than their smaller counterparts.

P. globosa has been found to have haemolytic activities, and hence have the potential for impacting human health.

Potential allelopathic effects of this alga on three other algae were also studied. The growth of these three algae was considerably inhibited.

Recent harmful algal blooms (HABs) in Sabah, Malaysia

Ann Anton, Normawaty Mohammad Noor and Sitti Raehanah Muhd Shalleh

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Cochlodinium polykrikoids blooms were first reported in the west coast of Sabah in January 2005 which resulted in incidences of fish mortalities. particularly in cage cultures. Cell densities reached the highest values of 6×10^{6} cellsL⁻¹ in March 2005 and then peaked again in June 2006 (2.6 x 10^5 cellsL-1) and in February (3.3 x 10^6 cellsL⁻¹) and December (3.7 x 10^6 cellsL⁻¹) 2007. The blooms were not periodic over the 3-year period and occurred at different times of the year when favourable environmental conditions prevailed. In addition, C. polykrikoides was first recorded in the Sulu Sea off Lahad Datu, eastern Sabah, with cell numbers reaching 2.0 x 10^3 cellsL⁻¹ in November 2007. During the occurrence of Cochlodinium blooms in the west coast of Sabah, G. catenatum was found to co-occur in high numbers with the highest cell densities recorded in December 2007 (2.2 x 10^5 cellsL⁻¹). The identification of G. catenatum is confirmed by light, scanning and DNA sequence. Integrated studies to identify the factors causing HABs in Sabah are being carried out. These include studies on ocean currents, nutrients and other environmental parameters, species dispersal through ship ballasts, cyst mapping studies and GIS and remote sensing for monitoring bloom events.

Application of molecular biological techniques for research of harmful algal blooms in Vietnam

Dang Diem Hong¹, Dao Viet Ha² and Yasuwo Fukuyo³

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 ²Marine Biochemistry Department, Institute of Oceanography, VAST, 01 Cau Da, Vinh Nguyen, Nha Trang, Khanh Hoa, Vietnam
 ³Asian Natural Environmental Science Center, The University of Tokyo, Yayoi 1-1-1, Bunkyo-ku, Tokyo 113-8657, Japan

In Vietnam, coastal area utilization will be expanded. Monitor the change of environment for protection of environment and natural resources and society is very necessary. Although Vietnam has a great biodiversity in fresh water and marine microalgae, application of Biotechnology to studies on harmful microalgae has just started very recently. In order to set up effective countermeasures against HABs, monitoring of harmful algae from the beginning of bloom is necessary.

Identification at species rank is difficult, because identification criteria used in conventional taxonomic system, i.e. morphological difference among species, are not easily observed under regular laboratory condition. It needs observation under an electron microscope or a light microscope with specific sophisticated attachments such as an epifluorescence or a differential interference contrast. However, these works have met a serious problem with species having similar morphological features that is easily changed by environmental factors. The recent trending the world, taxonomy and phylogenetic studies based species specific gene sequence data provides useful and powerful molecular biological method for their identification. Molecular methods include conventional PCR, real-time PCR, denaturing gradient gel (DGGE), electrophoresis fluorescent fragment detection PCR. fluorescent in situ hybridization (FISH), lectin, antibodies and ribosomal RNA (rRNA) - targeted DNA probes, gene sequence data and Single cells PCR have used for detection of the species of HABs becoming easier which will be discussed in this report.

Toxic cyanobacterial blooms and their toxins in some North Vietnam water bodies

Duong Thi Thuy¹, Dang Hoang Phuoc Hien¹, Nguyen Sy Nguyen¹, Dang Dinh Kim¹, Tran Van Tua¹, Dang Thi Thom¹ and Jens Dahlmann²

¹Institute of Environmental Technology, Vietnamese Academy of Science and Technology (VAST).

²Department of Food Chemistry, Institute of Nutrition, Friedrich-Schiller University, Jena, Germany.

Intensive cyanobacterial blooms in surface waters of recreational reservoirs and sources of drinking water resulting from water eutrophication are a problem in many countries. Typical cyanobacterial bloom is often accompanied by the production and release of toxins which are reponsible for animal and human poisoning. In Vietnam, water bodies are polluted by high level of nutrients which are discharged in wastewater without any treatment. Very little is known about the presence and abundance of toxic cyanobacteria and cyanobacterial bloom in Vietnamese water bodies.

Survey of toxic cyanobacterial blooms in several reservoirs, lakes and fishponds in the North of Vietnam were conducted from 2003 to 2005. During the study period the dominant bloom-causing species belong to genera Microcystis, Oscillatoria, and Anabaena. The most frequent species recorded is Microcystis aeruginosa.

The presence of the mcyA (microcystin synthetase A gene) in bloom and isolated samples indicated the genetic potential to produce toxin. Toxicity of natural bloom and isolated samples was studied using Atermia salina bioassay and results showed many of these samples were toxic to brine shrimps. High PP2A activity inhibition of investigated samples is shown by protein phosphatase inhibition assay (PPIA). 6 different MCs have been identified with total MCs content varied from 0.002 to 3.58 mg.g⁻¹ D.W.

The results showed the potential intoxication by toxic cyanobacteria and their toxins in investigated water bodies, especially, during bloom collapse when toxins may release from cells into water environment.

Transport of *Pyrodinium bahamense* var. *compressum* in Malampaya Sound, Palawan, Philippines

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Blooms of *Pyrodinium bahamense* var. *compressum* were reported in Malampaya Sound in the late 1990s. *Pyrodinium* and other cyst-forming dinoflagellate species, once deposited, become part of the bottom sediments. Thus, understanding bottom sediment transport is essential for determining the dispersal and distribution of dinoflagellate cysts, which may act as "seed beds" of future blooms.

We derived the net bottom sediment transport mainly from granulometric parameters; the direction of better sorting indicates the direction of transport. At the mouth of the Outer Sound, the transport pattern is inward. At its central portion, convergence takes place identifying the area as sediment sink. Similarly, the northern Inner Sound is also a zone of convergence. Bulk sediment properties and changes in bathymetry are fairly consistent with these trends.

The *Pyrodinium* bloom in the late 1990s occurred within the mouth of the Outer Sound. The transport pattern here suggests that *Pyrodinium* cysts found by previous workers in the sediments could have been derived from the open ocean.

In this work, *Pyrodinium* cysts were seen only in the upper 2 cm of some core samples in the Inner Sound. This may indicate bayward transport of cysts from the Outer Sound. However, cyst dispersal via bottom transport is unlikely as transport patterns suggest that the central portion of the Outer Sound is a sediment sink. Furthermore, transport patterns indicate that there is no significant transport across the Outer to the Inner Sound. Thus *Pyrodinium* is likely to have been transported into the Inner Sound via the water column and eventually becoming part of the local assemblage. The appearance of cysts in surface sediments of the Inner Sound suggests that *Pyrodinium* may have become abundant in the water column in recent years, thus becoming more detectable in the sediment.

Presence of *Pyrodinium* cysts in recent sediments and entrapment of sediments within the Inner Sound are threats to the shellfish industry in the basin. If conditions become suitable, *Pyrodinium* bloom may occur in the Inner Sound.

Nitrogen uptake rates during the HAB events of 2005 in East China Sea and comparisons with recent Dinoflagellate blooms on the Western Florida Shelf

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During late spring and summer of 2005, large scale (>15,000 km²) mixed dinoflagellate blooms developed in the coastal East China Sea. Karenia mikimotoi was the dominant HAB species in the first stage of the bloom (late May/early June), and was succeeded by Prorocentrum donhaiense several weeks later. Samples were collected during 3 cruises of the Chinese Ecology and Oceanography of Harmful Algal Blooms (CEOHAB) Program. Nitrogen (N) isotope methods were used to measure the rates of NO_3 , NH_4^+ , urea, and glycine uptake during the blooms, although NO_3^- uptake rates were only measured during the early stages. NO₃ uptake contributed about 30% of the total N uptake before the blooms and decreased to <20% at the beginning of the K. mikimotoi blooms. Of the reduced forms of N, NH_4^+ contributed on average ~65% (sum of NH_4^+ , urea and amino acids), and urea contributed ~20%. Both rates were highest during the peak of the K. mikimotoi blooms. The contribution of amino acids to total N uptake increased from <5% before the blooms to $\sim 10\%$ during the K. mikimotoi blooms and to 25% during the P. donghaiense blooms. This is consistent with the progression of ambient concentrations of N during the bloom progression: average ambient NO₃⁻ decreased significantly, while concentrations of NH_4^+ , urea and amino acids increased from early May to late June. Ambient concentrations of phosphate, as both PO_4^{3-} and organic phosphate, were highest during the *K. mikimotoi* blooms, but concentrations of NH₄⁺ were also elevated, leading to average ratios of DIN:DIP that were generally >40 in the early stages of the K. mikimotoi blooms, while DON:DOP ratios were <40. As the ratios of DIN:DIP decreased from >40 to <20, and reached proportions approximating Redfield ratios, bloom dominance shifted from K. mikimotoi to P. donghaiense. However, even when DIN:DIP ratios were near Redfield proportions, evidence of P limitation was shown by enhanced N uptake rates when pre-incubated with PO_4^{3-} .

In keeping with the overall GEOHAB strategy of applying the comparative approach, comparisons are drawn between the 2005 blooms in the East China Sea and recent blooms of Karenia brevis and mixed dinoflagellates, including Prorocentrum minimum, on the western Florida Shelf of the US. While the blooms in the East China Sea were separated in time, those on the western Florida Shelf were separated spatially. Similar types of measurements, but not as comprehensive as suite of measurements were made, precluding exact comparisons. Both the K. mikomotoi blooms and the K. brevis blooms occurred when concentrations of PO_4^{3-} , particularly DOP, increased and were associated with very low DON:DOP ratios. As in China, the K. brevis blooms were also coincident with elevated concentrations of NH₄⁺. The P. donghaiense blooms in China and the mixed dinoflagellate blooms of Florida occurred under similar N:P ratios: DIN:DIP <20 and DON:DOP >40. Both studies underscore the diversity of nutritional strategies by these different dinoflagellate functional groups and demonstrate that organic nutrients play important roles in their dynamics. In these cases, phosphate, particularly DOP was important in the Karenia sp. blooms, while concentrations of DON and rates of DON uptake appeared to be more important in the *P. donghaiense* and mixed dinoflagellate blooms than in the Karenia sp. blooms.

Alexandrium fundyense cyst dynamics in the gulf of Maine

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Studies examining linkages between HAB cyst abundance and blooms have typically covered small areas due to the difficulties and expense of mapping cysts. Here we present five cyst mapping surveys for Alexandrium fundvense in the Gulf of Maine (GOM), each covering hundreds of km in the alongshore direction, and 50-100 km in the offshore. The first survey (1997) revealed a widespread cyst distribution with two centers of abundance. Subsequent surveys showed substantial increases in cyst abundance from 1997 levels, with large interannual variability and an apparent increase in recent years. Hindcasting studies using a numerical model demonstrate that a major cyst deposition or accumulation event was the dominant factor leading to a massive 2005 red tide in the western GOM and southern New England, and that cyst abundance can be a first-order predictor of the magnitude of regional blooms in the GOM. Here we provide evidence for the persistence of the high-abundance cvst population and argue that we have entered a "new era" of significant, recurrent blooms in the coming years. A cvst seedbed-based conceptual model is presented that is consistent with interannual to decadal patterns of variability in the severity of blooms and shellfish toxicity in the GOM.

Toxic aquatic organisms from Vietnamese coastal waters

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Filter feeders such as bivalves and tunicates accumulate toxins through their feeding activity, when toxic microalgae occur in environment. This type of toxin contamination, especially those by *Pyrodinium bahamense*, is often reported in tropical Asia, and monitoring system on toxin has been set in the Philippines and Malaysia. In Vietnam, current situation on contamination of algal toxin is not clear. In our field survey in 2002-2007, no significant PSP and ASP toxins (saxitoxins and domoic acid) were detected in 2 species of clam (*Meretrix meretrix* and *M. lyrata*) and blue mussel *Perna viridis* collected at Nam Dinh, Thai Binh, Thanh Hoa, Nghe An, Ha Tinh, Khanh Hoa and Ho Chi Minh city. However, a significant level of DA has been found to be accumulated in all the specimens of *Spondylus versicolor*, one of the commercial bivalves in Vietnam, collected in Nha Phu lagoon, Khanh Hoa province. The origin and mechanism of DA accumulation in the species are under progress.

In contrast, toxic aquatic organisms endemic in Vietnamese coastal waters are well studied on their toxin components and individual variation, including poisoning cases. During 2002-2007, several species of xanthid crab, fish, horseshoe crab and octopus from various coastal areas were analyzed on their PSP and ASP toxin amounts. High level of PSP toxin was detected in xanthid crab Zosimus aeneus collected from Khanh Hoa. Significant level of PSP toxins was also detected in 3 species of puffer T. pallimaculatus, A. immaculatus and A. nigropunctatus from Khanh Hoa province and horseshoe crab Carcinoscopius rotundicauda from Can Gio, Ho Chi Minh City in addition to a large amount of tetrodotoxin. Tetrodotoxin was found as a principal toxin in a blue-ringed octopus Haplochlaena lunulata from Khanh Hoa province, goby fish Youngeithysis nebolusa from Cau Hai lagoon (Thua Thien - Hue province). Generally, PSP toxin is detected in TTX-bearing organism, but no PSP toxin was detected in these species.

Marine epibenthic dinoflagellates from Malaysia – Species diversity and future studies

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Studies of marine epibenthic dinoflagellates from Malaysia have revealed 24 species representing 8 genera viz. Prorocentrum, Sinophysis, Gambierdiscus, Coolia, Ostreopsis, Amphidinium, Pileidinium and Bysmatrum. Toxicity studies based on LC-MS/MS on selected species have confirmed that P. lima (3 strains collected from different areas) produces okadaic acid and its derivatives and Ostreopsis labens produces palytoxin. Preliminary results based on BGM fibroblasts showed that *Gambierdiscus* cf. *pacificus* has a strong capability to produce maitotoxin but further studies to confirm this is in progress. Future studies on epibenthic dinoflagellates are being conducted. These include: habitat preferences of epibenthic dinoflagellates on various substrates which will be used to study genetic variability among different populations; molecular phylogenetic studies of the genus Prorocentrum based on morphological characteristics, ultrastructure (periflagellar area) and cyt-b sequences. Further taxonomic comparisons will be done between Prorocentrum and Dinophysis to determine the possible homologues between sulcal areas of Dinophysis and the periflagellar area of Prorocentrum.

Bloom dynamics of *Cochlodinium polykrikoids* in Korea coastal waters in 2007

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Biological and hydrographic characters of Cochlodinium polykrikoides were investigated in Southern coast of Korea from June to October in 2007. The swimming cells were first observed at approximately 8 cells mL-1 in June in Narodo of Southern Sea. Initial bloom of C. polykrikoides occurred on 31th July in Southern Yeosu Gamak Bay and lasted for 50 days with maximum concentration of 32,500 cells mL⁻¹ (found in Namhae). The vegetative cells of C. polykrikoides disappeared on 18th September in waters. The blooms were observed in Wando of Southern Sea to Uljin of Eastern Sea with loses of US \$12 million in fisheries industry. Surface temperature was about 20°C in June prior to bloom and weak stratification was formed in 5-20 m depths. The temperature increased during the blooms with strong stratification and peaked in August at 25 $^{\circ}$ C which is a favorable temperature for C. polykrikoides growth. Considering environmental parameters during the time surveyed, the scale and duration of C. polykrikoides blooms might be affected by wind shift from the north to south and Kuroshio currents in mid August

Mixotrophic ciliate *Myrionecta rubra* needs certain type of cryptomonad prey to form a massive bloom

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Myrionecta rubra Jankowski 1976 (=Mesodinium rubrum Lohmann 1908) is a very common mixotrophic ciliate forming recurrent massive blooms in diverse marine environment. Successful cultivation of this species, however, was firstly reported early this century. Thereafter, ecological role and evolutionary importance of this ciliate have been highlighted owing to the available *M. rubra* strains. All the three strains of M. rubra so far known require cryptomonad preys for any notable growth. Among wide ranges of cryptomonad subgroups each of the M. rubra strain seems to select a certain subgroup for their favorite prey species. The Korean M. rubra strain MR-MAL01, for instance, grows far better with Clade 2 cryptomonad preys than with Clade 3 ones. The selected cryptomonad prey serves not only as a nutritional source but also as a plastid donor to *M. rubra*, a temporary plastid retainor. The cryptomonad preys, thus, are needed by *M. rubra* for its rapid growth through active kleptoplastidic photosynthesis as well as heterotrophic uptake of cell material inside the prey cells. In-situ competition over a certain subgroup of cryptomonad prey among *M. rubra*, dinoflagellates, and other ciliate species might be the leverage for the development of a massive *M. rubra* bloom in the sea. Other prevs like and predators of *M*. rubra such as Dinophysis spp. and other metazoans could be important for the secondary fate of a primary massive bloom of *M. rubra*.

The distribution and HAB of *Phaeocystis globosa* in coastal water of North China

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Phaeocystis belongs to Prymnesiophytes. It is widely distributed in the world coastal waters. HAB of Phaeocystis is increasing in recent years. In China, first record of *Phaeocystis* HAB was in Zeling Bay, Guangdong Providence in 1997. Since then, Phaeocystis HABs occurs almost every year in coastal water of South China. In June 2004, Phaeocystis HAB was first observed in Bohai Sea. North China. In 2005, a small scale HAB of Phaeocystis appeared in coastal water of Yantai City, North Huanghai Sea. In autumn of 2006 and 2007, Phaeocystis HAB occurred in Bohai Sea and lasted for about one month with fish kill. The determination of DNA sequence in 18S rDNA and ITS from a strain of *Phaeocystis*, isolated from Bohai Sea was carried out. The results showed that the species of *Phaeocystis* in Bohai Sea is *Phaeocystis globosa*. In coastal water of Qingdao City, *Phaeocystis* HAB happened in summer of 2007. The preliminary study on ecology of Phaeocystis both in Lab and mesocosm showed that Phaeocystis cell was attached on spinae of Chaetoceros cell through its haptonema. Then big colony could be formed by reproduction. Results from monitoring on HAB indicate Phaeocystis becomes one of the main causative species of HAB in North China coastal waters.

Study on HAB in coastal water of Qingdao City

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Qingdao is a coastal city at south coast of Shandong Peninsula. As the development of industry, tourism and harbor, the eutrophication in coastal water of Qingdao city is accelerated. The occurrence of HAB is increasing since 1990's. Before 2000, HAB events were mainly caused by *Noctiluca scintillans* and diatoms. However, after 200, *Mesodinium rublum* becomes the main causative species. The HAB of *Alexandrium* sp. *Gonyaulax polygramma* and *Heterosigma akashiwo* occurred sometimes. For a better control and management on HAB, basic research on ecology and oceanography were carried out as well as monitoring and early warning.

The PICES harmful algal bloom international project

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The Harmful Algal Bloom Section of PICES, the North Pacific Marine Science Organization, has recently proposed a training program to help establish and implement monitoring programs in developing non-PICES nations. The sharing of such monitoring information will ensure the security of imported fisheries products, and will protect the coastal environment and ecosystems of Japan and other importing nations. HAB International will focus on assisting in the development of capabilities in Vietnam, Hong Kong, Malaysia, Philippines, Thailand, Mexico, Peru, Chile, but the project is not exclusive of other developing nations that are interested in participating. The primary aims for the pilot training workshops are to establish effective protocols, determine proper equipment, develop appropriate databases, and to design ideal teaching strategies. We seek the support of key national and local regional officials in developing nations, and an understanding of the regional operational structure and laboratory facilities in the countries suitable for hosting the workshops. Preliminary site visits in Year 1 of the project by the workshop organizers will be used to work with country representatives to design the framework for the workshops. We will begin the training workshops in Vietnam during Year 2 using established contacts at regional and local levels that members of the organizing committee have established

Part 2

2nd ASIAN GEOHAB MEETING
Overview of the GEOHAB Core Research Project on HABs in Eutrophic Systems

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Eutrophication- related HABs are growing in frequency, geographic extent and duration throughout the world, but this increase is especially apparent throughout much of Asia. Although we have good quantitative estimates of many sources and forms of nutrient loads, the transformation processes of these nutrients and how they are affected by landscape changes, food web alterations, and climatic variations are not well understood. Through an international symposium in Baltimore, Maryland, USA, in 2005 and publication of the symposium report in 2006, the GEOHAB Programme has identified and justified the following questions that are central to the betterment of understanding of the relationship between HABs and eutrophication:

- 1. Are there clusters or specific types of HAB species that are indicative of global nutrient increases?
- 2. To what extent do residence time and other physical processes impact the relationship between nutrient loading and HAB proliferation?
- 3. How do feedbacks and interactions between nutrients and the planktonic, microbial food web impacts HABs and their detrimental effects?
- 4. Do anthropogenic alterations of the food web, including overfishing and aquaculture activities, synergistically interact with nutrients to favor HABs?
- 5. How do anthropogenic changes in land use, agricultural use of fertilizer, NOx emissions from vehicles, and global changes in land cover affect the delivery and stoichiometry of nutrients to coastal waters and the resulting incidences of HABs?
- 6. Do climate change and climate variability have impacts on ecosystems that augment the impacts of eutrophication on the formation of HABs?

Much progress has been made over the past several years in many aspects of HAB biology and ecology. In particular, the advancements in

understand of HAB physiology related to nutrition has greatly developed in the past decade. Additionally, significant advancements in understanding of nutrient loads from land have also developed, as has the understanding of the impacts of changing landscapes on nutrient delivery from land. At a global scale, nutrient export models are now available for inorganic nitrogen and phosphorus as well as organic nitrogen, phosphorus and carbon. As many questions related to HABs are of critical importance to the Asian waters, comparative approaches may aid in advancing the understanding of these relationships and ultimately in the management of nutrient loads and improved prediction and management of HABs.

Dinoflagellate cysts as a proxy of eutrophication in coastal waters

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Eutrophication is one of critical issues for coastal environments. Recent increase of harmful algal blooms (HABs) in the coasts of the world is closely related to nutrient enrichment caused mainly by anthropogenic activity. However, development of eutrophication has been not yet well understood because of misunderstanding the meaning of "eutrophication".

The word "eutrophication" means tendency of nutrient increase, and then does not indicate a certain nutrient level. For example, a phenomenon of nutrient increasing from oligotrophic to mesotrophic level is called "eutrophication" as well as from mesotrophic to eutrophic level. If this usage for "eutrophication" is adequate, we can understand why many different indexes of dinofalgellate cysts or assemblages have been suggested until now. For enstance, Dale et al. (1999) concluded Lingulodinium machaerophorum is the index species for cultural eutrophication in the case of Oslo Fjord. However, Matsuoka (1999) suggested the increase of heterotrophic dinoflagellate cysts reflecting the eutrophication in Tokyo Bay where is one of the highest nutrient level in the world. Thus, as Matsuoka (2001) have already mentioned, these differences in index species or assemblages for eutrophication suggest the different nutrient levels at the start of eutrophication.

In order to understand possibility of occurrence of HAB, it is very important to know on temporal dynamics of HAB species with the history of nutrient level. Particularly in East and Southeast Asia, many eutrophicated areas have rapidly appeared by recent anthropologic activities, and then expansion and extension of HAB events are feared. Analyses on phytoplankton including diatoms and/or dinoflagellate cysts preserved in sediments of coastal waters is very useful for understanding the present condition of phytoplankton communities related with nutrient levels from the past.

Bloom dynamics of harmful algae, *Cochlodinium polykrikoides* in Korean coastal waters

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In order to understand bloom dynamics of *Cochlodinium polykrikoides* the species, we studied the variation of biological and hydrographic characteristics before and during the bloom around the Korean coastal waters, mainly focused on the Narodo area from which the species always shows its initial outbreak and spreads near by. One or two chained young swimming cells of the species were initially observed in the South Sea of Korea from July with low densities less than 50 cells/L, where it took three to six weeks until initial bloom. In addition, although the swimming cells of the species appeared almost simultaneously around the southwestern part of the South Sea, the elapsed time to initial bloom was the shortest only near the Narodo area indicating that Narodo area was the most environmentally favorable place for the growth of the species.

The abundance of *Sagita enflata* was comparatively higher in the offshore between Jejudo and Geomoondo than that of inshore between Kwangdo and Narodo. Also, the abundance of *C. polykrikoides* swimming cells near Kwangdo and Geomoondo at the early growing stage before bloom outnumbered that of other areas. Therefore, it was assumed that distinct geographical and environmental characteristics in Narodo area could act as an important keys for *C. polykrikoides* to grow faster and make initial bloom than among other areas. In addition, the wind shifts from northwind to southwind, playing an important role in the accumulation of swimming cells, and the strength of kurushio currents and cold water mass, annually concurring near Jindo area during the summer season are might deeply affect to the beginning of first outbreak, scale and duration of the bloom in the Korean coastal waters.

Joint EASTHAB and Asian GEOHAB works to clarify the likely route of *Cochlodinium polykrikoides* in Kuroshio current system

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A dinoflagellate, Cochlodinium polykrikoides, has recently become more prevalent and posing a threat to marine life in Asian coastal waters in Korea, Japan, Philippine, and Malaysia. A tripartite research group, EASTHAB, regards this species as one of key harmful dinoflagellates, and has organized four consecutive meetings to collect and compile scientific findings essential to build actual prediction model for this species since 2004. Recent finding leads collaborative works of joint EASTHAB and Asian GEOHAB in order to establish prediction model. So far, the life cycle and the likely transport route have not yet been clarified. One hypothesis is that the northeastwards Kuroshio warm current transport the vegetative cells to Korean coastal waters and then developed into subsequent dense blooms after mixing with eutrophic coastal waters. It witnessed that this upwarding Kuroshio current would intervene in the transport of C. polykrikoides whose likely route supposed to be from western to northern Pacific as did in the transport of Chaetognaths and Leptocephalus, an early life stage of Pacific eel. It needs to take collaboration of EASTHAB and Asian GEOHAB in HAB monitoring to cover Kuroshio current system to track the likely route of this dinoflagellate.

Population dynamics of targeted species with special emphasis on *Prorocentrum donghaiense* Lu in

Zhejiang coastal water, China

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A couple of micro-algae cause blooms in Zhejiang coastal waters of the East China Sea. Among them, the most well known species is Prorocentrum donghaiense which form extensive blooms in the last decade. Field observations have suggested that populations of these dinoflagellates have initiated in the subsurface layer in offshore waters where they are influenced by the Taiwan warm current (TWC). For instance, the density of P. donghaiense exceeded 10,000cells/L in early April 2004 and became the dominant phytoplankton species in subsurface micro thin layers near the 50 m isobath where an inoculum of the causative species was found and resulted in the subsequent development of massive blooms. Environmental factors such as direction change of monsoons, water stratification, and the advance of the TWC play an important role in promoting development of large-scale blooms of dinoflagellates in the ECS. Seasonal pattern of population P. donghaiense is found as follows: it is abundant and forms extensive blooms in spring; widely appears with relatively high cell density at some part of area in summer; occur sporadically in autumn and very few in winter. The annual variation of population and bloom pattern such as species succession, time of bloom initiation, persisting etc are controlled by physical processes. More interdisciplinary studies in the near future are really required for further elucidating the population dynamics on targeted HAB species

The eco-physiological studies of phosphorus on the growth of *Karenia mikimotoi*

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Karenia mikimotoi caused frequently harmful algal blooms in Chinese coasts in recent years. The present research studied phosphorus eco-physiological characters of *K. mikimotoi* in batch cultures and semi-continuous cultures.

The utilization characters of K. mikimotoi to phosphorus indicated that *K.mikimotoi* can use NaH2PO4·2H2O, G-P, ATP. The algae prefer higher phosphorus concentration and lower N/P ratios.

The absorption character of *K. mikimotoi* to phosphorus indicated that the Ks of *K. mikimotoi* to DIP is 0.258 μ M, which is lower than many other dinoflagellates. The affinity index (α) is 417.6, which is much higher than many other dinoflagellates. sIt is higher competitive ability for *K. mikimotoi* to absorb phosphate.

The results of cellular growth kinetics research indicated that μ max of *K. mikimotoi* is 0.443 day⁻¹, which is within the ordinary range of dinoflgellate growth rates. So *K. mikimotoi* is not a harmful algae which depend on growth strategy to bloom.

The orthogonal experiment L9(33) of *K. mikimotoi* indicated that optimal temperature, salinity and phosphorus for *K. mikimotoi* growth were 25°C, 25‰ and 2.5 μ M respectively under 100 μ E m⁻²·s⁻¹ illumination.

We can conclude that although the low μ max could not be trigger for K. mikimotoi blooming,the features that utilization of several types of DIP and DOP, lower Ks and higher α made K. mikimotoi easier survival in low nutrition. The blooms occur when the environmental factors approach the best conditions.

Green *Noctiluca scintillans*, successful red tide species in SE Asian waters

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Noctiluca scintillans (Macartney) Kofoid et Swezy forms a distinct greenish red tide in the tropical Southeast Asia and adjacent waters due to the presence of the photosynthetic endosymbiont Pedinomonas For this reason, it is locally called green Noctiluca. noctilucae. Although the cell size of green *Noctiluca* tends to be smaller than that of asymbiotic *N. scintillans* in temperate waters, the morphology, life cycle, bioluminescence and volume specific respiration of green *Noctiluca* are similar to asymbiotic N. scintillans, and both are considered to be an identical species. Green Noctiluca exhibits two types of nutritional mode according to clones: one requiring an external food supply and the other not. The latter grows photoautotrophically for generations, but they also feed on food particles. It grows faster with foods than without. Furthermore, sexual reproduction does not occur during its autotrophic growth under laboratory conditions. Thus, P. noctilucae assures subsistence of the host under food-limited conditions, but the symbiont does not give higher growth rates than in asymbiotic N. scintillans. Phagotrophy makes its growth fast.

Green Noctiluca is one of the most recurrent red tide species in the Southeast Asian waters, particularly in Manila Bay, the upper Gulf of Thailand, and Jakarta Bay. In Manila Bay, green Noctiluca became the most frequent causative species of red tides in 2001 and since 2003 the organism forms perennial red tides in the bay, occasionally covering Our observations suggest that although green almost whole area. Noctiluca grows slower than co-existing fast growing species, once it becomes dominant, its active grazing prevents population growth of coexisting phytoplankton. In the upper Gulf of Thailand, green Noctiluca is the most recurring red tide species. Its growth was first recorded in 1957, and the frequency and intensity of its red tide have been increasing over the past 2 decades. A seasonal study in the upper Gulf indicates that active growth of N. scintillans by both sexual and asexual reproduction is supported by phagotrophy. While nutrient supply is sufficient in the upper Gulf throughout the year, the dependence of N. scintillans on the

nutrient concentration is not significant. Instead, this nutrient condition produces a favorable food condition through growth of phytoplankton as prey.

Thus, on-going eutrophication in the SE Asian coastal waters appears to provide a competitive advantage to green *Noctiluca* over coexisting species by increasing food availability. A comparative study on the ecology of green Noctiluca within the SE Asia and with asymbiotic *N. scintillans* is expected to provide insight on the expansion of this species in Asian waters.

Harmful algal blooms in Manila Bay, Philippines: - the Pyrodinium bahamense var. compressum period and the Noctiluca scintillans period

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Manila Bay has been one of the areas frequently affected by harmful algal blooms in the Philippines/Southeast Asia. The recurring blooms of *Pyrodinium bahamense* var. *compressum*, first experienced in 1988 and almost yearly thereafter, overwhelmed the country with public health and economic problems. From 1999 up to the present, however, a"take over" of *Noctiluca scintillans in* Manila Bay has been observed. Because of these events, this semi-enclosed bay has become one of the most studied sites in terms of *Pyrodinium* (and *Noctiluca*) bloom dynamics.

Pyrodinium- related studies in Manila bay include long term estimates of its *motile* cell density and toxicity; vertical and horizontal cyst distribution; and hydrodynamics/modeling. A study on *Noctiluca-Pyrodinium* has also been done, results of which reveal that *N. scintillans* show high growth rate utilizing *Pyrodinium* as source of food, thus indicating that *N. scintillans* is a potential grazer of *Pyrodinium* in the field. Recently, the phytoplankton composition and physico-chemical conditions in the area during a *Noctiluca scintillans* bloom has been undertaken as part of an on-going research that aims to contribute to the understanding of the bloom dynamics of the species in the bay.

This paper is a review of *Pyrodinium bahamense* var. *compressum's* and *Noctiluca scintillan's* bloom occurrences in Manila Bay, Philippines with brief discussions on available knowledge relevant to the understanding and management of their blooms.

Eutrophication and *Noctiluca* red tide in the upper Gulf of Thailand

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The Gulf of Thailand is a tropical semi enclosed sea which located in the southwestern part of the South China Sea. The gulf covers an area of about 350,000 square kilometers and divided into three parts which are the upper, middle and lower part. The area of the Upper Gulf (another name is the Inner Gulf) is about some what of 100×100 square kilometers with average depth of 20 meters. There are four main rivers (the Mae Klong, the Ta Chin, the Choa Phraya and the Bang-pa-kong Rivers) flow through the densely-populated, agricultural and industrial areas of the Central Plain, draining to the north of the Upper Gulf of Thailand. The total flow from these four major rivers is between 59×10^6 m³ /day to 150×10^{-6} m³ /day. With its nutrient-rich, confined nature and shallow water, the eutrophication of coastal waters has become apparent as a problem since the past two decades. The long term study on ecological changes in the gulf from 1971 to 1987 revealed that the mean cell density of phytoplankton has increased more than two times from 1976 to 1987. The increasing trend was also noticed in chlorophyll a content in seawater. The concentration of nitrate and phosphate has also increased from 1974 to1989; red tides were observed in wider areas and trend to increase in the river mouth but the frequency of occurrences has fluctuated during 1981 to 1989.

Generally, *Noctiluca scintillans* and *Ceratium furca* is the main causative red tide organism in coastal areas of the Upper Gulf, however the blooms of some diatoms are also observed near the river mouth. The recent studies showed that red tides were often observed in the eastern and the western part of the Upper Gulf during the southwest monsoon and the northeast monsoon, respectively. These observations were well supported by the satellite images of chlorophyll *a* concentration taken by SeaWIFS (1997-2002) and MODIS (2002-2006). The studies on population dynamics of *Noctiluca* in the eastern coast of the Upper Gulf during 2002-2005 clearly showed that this species often formed red tide during southwest monsoon period (raining season). Although there was no any red tide in dry season (low nutrients and low phytoplankton cell density) but a few cells of green *Noctiluca* can be observed in the water column. These observation suggesting that the ednosymbiont, *Pedinomonas noctilucae*, may play an important role as the food source for *Noctiluca* to survive in dry season. To prove this hypothesis the preliminary study was conducted in the clonal culture of *Pedinomonas* and the results showed that this tiny symbiont could release amino acids and oligosaccharides into the culture medium. Therefore green *Noctiluca* may obtain the dissolved organic compounds from the symbiont and perhaps may also digest the dead *Pedinomonas* cell as the particulate food. To obtain more understanding on the physiology and ecology of *Noctiluca* the comparative study between the red and green *Noctiluca* is needed.

In situ and potential eutrophication in Hong Kong waters

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The coastal waters of Hong Kong are profoundly influenced by 3 nutrient inputs: the N-rich Pearl River discharge, relatively nutrient-poor oceanic waters from the South China Coastal Current in winter, and year round domestic sewage effluent. Actual and potential nutrient limitation were determined in four representative stations on seasonal basis to assess phytoplankton response to seasonal changes in nutrient concentrations and physical processes in Hong Kong waters.

In winter, the NE monsoon winds and downwelling enhances the invasion of coastal shelf waters that are relatively low in nutrients. Ambient chlorophyll concentrations are relatively low (< 5 µg chl $a L^{-1}$) because the water column is vertically mixed and phytoplankton are light-limited. During this period, potential eutrophication (the maximum yield of chl a during the incubation) was < 44 µg chl $a L^{-1}$ due to relatively low nitrogen concentrations.

In contrast, in summer, Hong Kong waters receive high nutrient inputs from the Pearl River discharge with high N:P ratio (up to 100:1). In situ and potential eutrophication varied spatially. High potential eutrophication (> 90 µg chl a L⁻¹) was attributed to high nutrient influx from the Pearl River outflow and sewage effluent in western waters and Victoria Harbor, while in situ concentrations of chl a was low (< 13 μ g L⁻ ¹) in these areas due to strong hydrodynamic mixing and high flushing rate. In more stable areas, the degree of in situ eutrophication was primarily regulated by PO₄ availability, with high chl a (~ 40 μ g L⁻¹) in the southern waters due to relatively high PO₄ input from the Pearl River discharge and with low chl a (~ 11 µg L⁻¹) in eastern waters due to low nutrient concentrations in more oceanic influenced waters. Our results implies that PO₄ removal in sewage effluent is necessary to control the eutrophication impacts during summer while nutrient removal is less important in winter since phytoplankton biomass was restricted by physical processes during winter.

Dinoflagellate-associated bacteria: is there a common theme and could it be a subject for a regional project?

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In recent years there have been renewed interest in bacteria associated with marine dinoflagellates. Analysis of isolates from various regions indicate that the bacteria flora comprise a fairly conservative group of Proteobacteria, CFB and Firmicutes. Furthermore in at least one case 16S rDNA sequence of a bacterium associated with Alexadrium minutum from Malaysia had 99% similarity with the sequence of Oceanicaulis alexandrii that was first described from a culture of A. tamarense from England. Analysis of genome fragments of bacteria associated with A. minutum and Coolia monotis from Malaysia also showed that most of the genes matched most significantly with genes of Roseobacters. At present these bacteria-dinoflagellate associations are considered based primarily on trophic underpinnings, such as DMSP metabolism but they could have a wider range of influence on the dinoflagellate life cycle and success. Advances in molecular biology and metagenomics enable these associations to be studied at a deeper level than previously possible. Perhaps the suitability of this subject as a basis for comparative studies under GEOHAB should be explored.

Growth and toxin production of dinoflagellate Alexandrium minutum (Dinophyceae) isolated from aquaculture pond in northern Vietnam

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In October 2004, six clonal cultures of Alexandrium minutum established from Hai Phong, Northern Vietnam and were examined for their toxicity. Toxicity analysis showed these cultures of A. minutum contained predominantly GTX4 while other congeners GTX1 - 3, NEO and dcSTX were also detected in small or trace amount. Interestingly, a toxin congener was consistently detected at different growth stages and also in all the strain. The congener was further purified using column Bio-Gel P2 and Bio-rex 70 and was identified as GTX4-12ol using LCMS/MS and NMR. However, the congener showed absence of biological activity with AOAC Mouse bioassay. In this study, growth and toxin production of A. minutum strain Am17 was examined under different salinity regimes. Experiments were carried out in duplicate in seven salinity conditions (5 to 35 psu) with constant temperatures of 25°C and illumination of 150 ± 10.0 µmol m⁻² s⁻¹ at 12:12 light:dark photo-cycle. Growth of A. minutum was optimum at 10-15 psu with 0.21-0.24 day⁻¹, but suppressed under salinities 5 and 35 psu (0.06-0.08 day⁻¹). Toxin production rate, R_{tox} at salinity 5 psu (1.10 ± 0.54 fmol PSTs cell⁻¹ dav⁻¹) was 2-3 folds lower than high salinity regimes (10-35psu)(3.30 - 5.30)fmol PSTs cell⁻¹ day⁻¹). Rtox of GTX4-12ol was lowest at 10 PSU that coincided with highest growth rate. However, no significant change in Rtox of GTX4 was observed in the range of salinities 10-35 PSU. Based on these results, it was postulated that the GTX4-12ol was the transformation product of GTX4. Further study of enzymatic activities involved in GTX4-GTX4-12ol transformation is crucial in understanding the regulating mechanism of this unique congener.

Oceanography analysis of annually variation of harmful algal /phytoplankton blooms in the western South China Sea

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Harmful Algal Blooms (HABs) in the South China Sea (SCS) coastal waters have caused large economic losses in aquacultured and wild fisheries: phytoplankton blooms in a narrow jet-like protrusion into the western SCS appear in the western SCS in summary season almost every year. The present study reviewed HABs with the oceanographic conditions in western SCS, and analyzed annually variation of the jet-like phytoplankton bloom using satellite remote sensing data. The HAB had high Chlorophyll-a (Chl-a) concentrations, occurring about 200 km off the coast and about 200 km north-east of the Mekong River mouth, for a period of about 6 weeks. In the same period, Sea Surface Temperature (SST) imagery showed a coldwater plume extending from the coast to the open sea, and QuikScat data showed strong southwesterly winds blowing parallel with the coastline. The study indicated the HAB was induced and supported by offshore upwelling that bring nutrients from the deep ocean to the surface and from coastal water to the offshore, and the upwelling was driven by strong wind through Ekman transport when winds were parallel with the coastline. During June- Sep, regional phytoplankton biomass appeared as a large jet-like shape from the coastal waters of Vietnam eastward toward the SCS; this feature intensified and extended as a large jet or a gyre from July to September, decayed in October, and disappeared entirely in November. The gyre was about 400 km in diameter with Chl-a concentrations from 0.5 to 2 mg m-3. Data on sea surface temperature (SST), winds, and sea surface height anomalies indicated a strong offshore upwelling during the period of strong southwesterly winds alongshore. The upwelling coincided with the regional increase of phytoplankton biomass in terms of shape, timing, and location. We observed this phenomenon during every southwesterly monsoon season from 1997 to 2002 using Chl-a images from three ocean color sensors; it appeared to be related with the cross-shelf upwelling that deliver nutrients to surface waters. The phytoplankton then flowed along with a large anticyclonic gyre into the SCS. Annually variation of the jetlike bloom/HAB in the western SCS is influenced by varies condition, such as El Nino. This study is jointed supported by The CAS/SAFEA International Partnership Program for Creative Research Teams, and One Talents Program, Chinese Academy of Science.

On the prediction of harmful algal events

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Prediction of harmful algal events is a necessity in order to minimise or mitigate their effects. This is usually enabled through models which are generated from a thorough understanding of the physical and biological dynamics of the relevant marine systems. Two models are presented whereby prediction of specific events is possible over different time scales. One of these, for the initiation of *Alexandrium* blooms, requires parameterisation of transitions between stages in the life cycle of the genus. The other, principally for *Dinophysis* but also includes other species, is based on an understanding of the physical dynamics of the affected region. These examples are shown in the case of harmful algal blooms around Ireland, but the approaches have global relevance and their application in other locations is quite simple.

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Red tides in south central coast of Vietnam: - Biodiversity and socio-economical consequences

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The blooms of a Haptophyte – *Phaeocystis globosa* in open waters of Binh Thuan province were recorded in the years of 2002, 2005, 2006 and 2007. About 90% of animal and plant species in tidal reefs of Phanri Bay were eliminated by bloom of *P. globosa* in July 2002. Cage-raised fish and lobster died, and losses were estimated to be over VND10 billion (ca. \$US 650,000). Because of the seriousness of the situation, surveys of biological and hydrochemical patterns were carried out in July 2002, August 2005, and August-September 2007. Data from these surveys will be presented.

In July 2002, colonies and cells of *Phaeocystis* cf. *globosa* were absolutely dominant in phytoplankton communities, with average concentrations of motile cell (free motile stage) and non-motile colony (colony stage) aty 3 x 10⁹ cells.1⁻¹ and 45 x 10³ colonies.1⁻¹, respectively. In 2005, the concentration of *P. globosa* ranged from 145 x 10⁶ to 320 x 10⁶ cells.1⁻¹, with an average concentration of chlorophyll-a at 45.6 \pm 7.6 µg.1⁻¹. Concentration of phosphorous and nitrates were low in the bloom areas, 12.2 and 49.0 µg.1⁻¹, respectively, but the pheophytine concentration reached the highest value, at 250 µg.1⁻¹. During the bloom of *P. globosa* in Sept 2007, diversity index of phytoplankton was less than 0.5. An analysis of cummulative dominance (K) showed that the phytoplankton community was very old and in a senescent stage in comparison with the pre-bloom period.

To minimize the bloom's affects on aquaculture industry and public health, a monitoring programme is needed to propose an early warning..

Data on Amnesic Shellfish Poisons (ASP) in bivalves at some aquaculture areas in the northern and north-central coastal waters of Vietnam

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In the framework of national project coded KC.09-19 (2004-2005), a study on the accumulation of ASP toxin in bivalves (clam and green mussel) at some aquaculture areas in the northern and north-central coastal waters of Vietnam had been carried out. The study results showed that the accumulation of ASP toxin concentration on viscera tissues of clam and green mussel at Do Son, Cat Ba, Tien Hai, Lang Co areas usually fluctuate by time not strongly in 2004-2005. Of which the average concentration of ASP toxin per month in Do Son clam was the lowest, and ranged from 0 to 2.8 ng/g tissue. The highest concentration (78.38ng/g) was found in Cat Ba mussel in August 2004. The concentration of ASP toxin detected in clam and mussel tissues in the sampling areas were still lower than the permit limit and safety for consumers. There was the relationship between concentration of ASP toxin and biomass of *Pseudo-nitzschia* diatom at the sampling areas although it was not clearly. The accumulation of toxin in shellfish tissue depend on the toxin accumulation capacity and depuration of each shellfish species, on Pseudo-nitzschia biomass and toxin produce capacity of toxic phytoplankton species under the effect of environmental factors in each sampling area as well.

Biodiversity of planktic Oscillatoriales (Cyanobacteria) in La-Nga river, South Vietnam

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Eighty-eight cyanobacterial species belonging to order Oscillatoriales from La-Nga river were examned, in which, two genera of *Oscillatoria* and *Phormidium* were high diversity with 26 and 24 species, respectively. Other 30 species were found in genera such as *Planktothrix*, *Lyngbya*, *Homoeothrix*, *Geitlerinema*, *Komvophoron*, *Pseudanabaena*, *Planktolyngbya*, *Spirulina*, and *Borzia*. Genera of *Borzia* and *Homoeothrix* were first time recorded to Cyanobacterial flora of Vietnam. Descriptions and photographs of these species were made.

On a toxic benthic dinoflagellte *Gambierdiscus toxicus* isolated from Cau Island, south central Viet Nam

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Gambierdiscus toxicus was isolated from *Avrainvillea sp.* (Chlorophyta) arround Cau Island of Binh Thuan Province, south central coast of Vietnam. Growth rates were low at salinity of 20 % in all experimental temperatures (20, 23, 26 and 29°C), and were rather high at salinities 25, 30 and 35 % in all temperatures. Maximal division rates ranged from 0.3 – 0.4 per day at salinity of 30 %, temperature of 26 °C; and at salinity of 35 %; temperature of 23°C. The species was confirmed to produce toxins of P-CTX and P-CTX-4B. Morphology of species was also described and photographed.

Species composition and ability to produce toxins of cyanobacteria of genus *Microcystis* Kützing & Lemm. in Tri An reservoir, Dong Nai province

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Six species of genus Microcystis found in Tri An Reservoir: Microcystis aeruginosa, Microcvstis botrys, Microcvstis wesenbergii, Microcvstis paniformis, Microcystis flosaquae, Microcystis sp1. Distintive characteristics of colony morphology of each species in different stages of development have been decribed. Of 14 varieties (isolated strains) from 4 species: M. aeruginosa, M. botrys, M. wesenbergii, M. panniformis, 13 strains are found to contain toxins microcystin. Mp1 and Ma4 (M. aeruginosa) have the highest level of microcystin. Mw and Mw2 (M. wesenbergii), in particular, contain toxin microcystin with concentrations of 19.48 (ng/l) and 1077.25 (ng/l), respectively .The relationship between the ability to produce toxins of these bacteria and the culture medium pH is rather conspicuous with pH = 8 and pH = 9 (no microcystin) versus pH = 7 and pH = 10 (microcystin concentrations: 15.47 (ng/l) and 2.92 (ng/l) respectively). With Mb, the highest microcystin concentration is found with pH = 8 (143.79 ng/l) and the lowest with pH = 7 (3.17ng/l). T1 shows the highest concentration of microcystin with pH = 9 (808.13ng/l), and lowest with pH = 8 (71.61 ng/l).

Phylogenetic relationship and distribution of two noxious dinoflagellate *Cochlodinium polykrikoides* and *C. fulvescens* (Gymnodiniales, Dinophyceae)

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Phylogenetic relationships among several chain-forming Cochlodinium species, including the harmful red tide forming dinoflagellate Cochlodinium polykrikoides and C. fulvescens recently described from Asian coastal waters, were examined using specimens collected from coastal waters of Canada, Hong Kong, Japan, Korea, Malaysia, Mexico, Philippines, Puerto Rico, and USA. The phylogenetic tree inferred from partial (D1-D6 regions) large subunit ribosomal RNA gene (LSU rDNA) sequences clearly differentiated between C. polykrikoides and C. fulvescens. Two samples collected from the Pacific coasts of North America (British Columbia, Canada and California, USA) having typical morphological characters of C. fulvescens such as the sulcus located in the intermediate region of the cingulum, were closely related to C. fulvescens from western Japan in the phylogenetic tree. However, ecological characteristics of these strains collected from western and eastern Pacific sides are different particularly in response for water temperature. Cochlodinium polykrikoides formed a monophyletic group positioned as a sister group of the C. fulvescens clade with three wellsupported sub-clades. These three clades were composed of, 1) East Asian, including specimens collected from Hong Kong, western Japan, and southern Korea, 2) Philippines, from Manila Bay, Philippines and Omura Bay, Japan, and 3) American/Malaysian, from the Atlantic coasts of USA, the Pacific coast of Mexico, Puerto Rico, and Sabah, Malaysia. For the moment, each of these clades is considered to be a so-called "ribotype" representing the population inhabiting each region, which is distinguished based on ribosomal RNA gene sequences in the species despite of similarities in their morphological characters.

Potentially Toxic Cyanobacteria and Health Risk to Community in the South and Central Highland of Vietnam

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Recently, toxic cyanobacteria and cyanobacterial compounds have been recorded in water bodies in Vietnam. Surface water from lakes, reservoirs and rivers mainly supplies for daily domestic and industrial activities. So that citizens are dealing with a health risk related to cyanotoxins in Vietnam.

Nine potentially toxic cyanobacteria have been found in Bien Ho Lake, Tri An Reservoir and in a shrimp pond in the South and Central Highland of Vietnam. These species belonged to four genera, *Microcystis, Anabaena, Aphanizomenon* and *Cylindrospermopsis*, and presented all year round in the lake and reservoir. Scum of *Microcystis* spp. was recorded in Tri An Reservoir. So that millions of local people have been facing to a health problem via daily water and food consume. *Anabaena smithii* was the first time confirmed based on morphological characteristics and *Cylindrospermopsis* cf. *curvispora* was the first time recorded in Vietnam. List of paricipants

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Front cover: Surface bloom of *Noctiluca scintillans* in South central coast of Vietnam (Binh Thuan Province), Sep. 2007 Photo by Hal Doan-Nhu Back cover: Foam produced following the bloom of *Phaeocystis globosa* in South central chast of Vietnam (Binh Thuan Province), August 2005, Photo by Lam Nguyen-Ngoc