

ANNUAL REPORT 2012

年度報告



海洋污染國家重點實驗室

State Key Laboratory in
Marine Pollution

The State Key Laboratory in Marine Pollution 海洋污染國家重點實驗室

envisions a solid base where coordinated and long-term research can be conducted to tackle marine pollution problems.

希望通過堅實的長期協作研究平台以解決海洋污染問題

The mission of the SKLMP is to protect the marine environment of Hong Kong and South China by identifying major threats such as algal toxins and contaminants of emerging environmental concern, and developing tools and technologies to address and solve these problems.

海洋污染國家重點實驗室的使命為通過明確威脅海洋環境的主要問題，如藻毒素和新興環境污染物等，並發展相關設備和技術致力於這些問題的解決，以保護香港和華南地區的海洋環境安全。



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A Message from the Director

主任致辭

State Key Laboratories (SKLs) are key components of China's science and technology research system. They are the foundation for top-level basic and applied research development nurturing outstanding researchers and helping to promote the development of scholarly exchanges for the country. SKLs carry a high status and signify recognition from the Ministry of Science and Technology of China (MOST) for research undertaken by a specific laboratory in a specific location. These laboratories produce innovative research aligned with national technology development directions, the national economy, social development goals and national security priorities.

Consequently, in each round of applications elite institutions, research centres and universities from the whole of China make significant efforts to compete for the honour of being or retaining the title of State Key Laboratory. For example, there were more than twenty applications from universities for partner State Key Laboratories this year in Hong Kong alone, demonstrating active participation and the determination of universities in the SAR to engage fully with the national scientific research system.

The State Key Laboratory in Marine Pollution (SKLMP) was established in December 2009 and our particular strength lies in the multidisciplinary nature of the team that has been assembled. This team draws from the expertise of 31 members and the shared facilities of six collaborating universities bringing together a profound depth of collective expertise. Since inception, the SKLMP has been in a less advantageous situation financially, when compared with most of our counterparts on the Mainland, who are receiving RMB 10 million each year from MOST. As a result of this disadvantage, helping participating members achieve a sense of being a team member in the SKLMP remains one of our biggest challenges.

Despite this, the SKLMP is proud of its reputation as an exemplary multidisciplinary team in terms of tackling marine pollution problems. We have remained committed to assembling our group of renowned professionals and researchers, and equipping them with state-of-the-art research facilities to ensure we continue to develop the complementary expertise that will increasingly place us at the forefront of marine environmental research. These concerted efforts will enable us to continue to take a lead in promoting scientific development, and assist us to help solve complicated environmental problems over larger temporal and spatial scales.

In the past three years, the aims of the SKLMP have been:

1. To provide state-of-the-art technological support for researchers to international standards, and to promote the development, deployment, operation and transfer of advanced technology for innovative research and educational excellence in the marine environment.
2. To adopt a strategy for providing seed funding through the SKLMP Seed Collaborative Research Fund (SCRF), CityU Internal Research Fellowship Programme (RFP) and CityU Internal Research Fund (IRF) with the twin objectives of helping members to acquire necessary preliminary data and better prepare their applications for major external grants, and bringing together members by undertaking key research projects. The mixing of expertise and members from different institutions will guarantee full utilisation of complementary expertise, cross fertilisation and an exceptionally high level of synergy.

Our members have won the 2012 First and Second-Class award in the Natural Science category at the Higher Education Outstanding Scientific Research Output Awards (Science and Technology) from the Ministry of Education of China. Many of the outstanding achievements of the SKLMP are the result of the collaborative effort, professional knowledge, commitment and unreserved contributions of our strong team of core members who have been very successful in attracting research grants and consultancies, and in producing a good number of high quality publications. The cohesion of our team of experts has been recognised as the backbone of the SKLMP and also the key to our success.

We are pleased that the Research Centre for the Oceans and Human Health (H2O) has been established in Shenzhen with the approval of the Shenzhen Municipal Government in 2012. With SKLMP members, CityU and Shenzhen Virtual University Park working as advocates of H2O, we have received three grants from the National Natural Science Foundation of China (NSFC), one grant under the Major National Science and Technology Programme in the “Twelfth Five-Year” Plan period, a Shenzhen Strategic Emerging Industry Development Special Project and three collaborative projects from the Provincial and Municipal Authorities in China, with an estimated grant total of RMB 6.88 million in an eight-month period. H2O has already served as an excellent platform for academics in Hong Kong and Mainland China to develop closer collaboration through academic and technological exchanges.

In addition, under the full support of the Shenzhen Virtual University Park and Prof. Ji-lan Su, the Shenzhen Marine Research and Technology Consortium (SMART) was established. H2O, together with nine other universities and research centres which conduct marine-related

research based in Shenzhen, and in collaboration with the environmental industry, took the lead in stimulating the formation of marine innovative technology ventures in China by turning some of our novel cost-effective technology 'know how' into real-life applications for the benefit of the State's economy and social development. SMART is expected to foster the commercialisation of our intellectual property through the Shenzhen-Hong Kong Innovative Technology Circle to enable China to capitalise on the rapidly growing commercial opportunities presented by the vast world marine environmental market, and to ensure we are ready to play a leading role in managing marine environments.

The SKLMP is the only state key laboratory focusing on marine environmental research in Hong Kong and, in addition to its geographic advantages, has a mission to promote academic exchange and collaboration with first-class academic institutions in Hong Kong, Taiwan and Mainland China. Grasping the opportunities provided by the peaceful historic development between China and Taiwan, the SKMLP has organised the First Xiangjiang Marine Forum in 2012. The Forum has invited distinguished speakers and participants from different sectors, including government departments, corporations, local universities and research institutes to present research topics and reports, and to participate in free discussion. The goal of the Forum is to create a relaxed environment and to provide a platform of "enlightening new ideas and promoting academic fusion" for researchers through academic discussion and hands-on workshops. The Forum aims to encourage the questioning of original theories and the promotion of free academic exchange through the expression of different opinions. By investigating the frontiers of scientific research, the Forum will foster original ideas and insights and contribute to the development of interdisciplinary collaboration. In purely practical terms, the Forum will promote knowledge innovation and improved laboratory techniques and practices. Collaboration developed during the forum between SKLMP members will be directed toward application for SKLMP internal strategic research funds, including the SCRF and the IRF. As a performance indicator, each of these strategic research projects should aim at bidding for major outside competitive grants. Through discussion and collaboration between scientists, government officials, NGOs and educators, this forum will deliver state-of-the-art marine science knowledge to the public, aimed at enhancing public awareness of marine conservation, and promoting the development of marine science research and education.

In addition to working actively at the frontier of marine pollution research, social responsibility is also one of the key missions of the SKLMP. In order to raise public awareness of marine conservation and environmental protection, and help marine researchers to gain further understanding of the ocean, a charity programme which conducts Scuba Diving Training, "The

Spin Kid Project”, was organised in September 2012. At present, our members number over 50 professionals, and we have won praise and support from more than ten universities and academic institutions in China.

As a result of the unreserved efforts of our members and our friends in China and overseas, the SKLMP has made an excellent start in 2012. Looking forward to 2013, there is still a long road ahead with many responsibilities and challenges to be faced. These require us to be united, committed and selfless, in order to continue to strive for perfection in our influential marine pollution research.

I sincerely hope that the SKLMP will become a base for us to contribute to the economy and social development of the State through research, technology innovation and knowledge transfer; and a base for assembling and nurturing outstanding young researchers. I sincerely hope that with our continued commitment and efforts, Hong Kong and Shenzhen will become one of the most important marine science research centres in South China.

Finally, I would like to share my favorite motto from Mr. Lin Zexu as a New Year Greeting for all of us:

“海納百川，有容乃大；壁立千仞，無欲則剛。”

(All rivers and streams will run into the sea, its greatness contains everything; it implies tolerance and capacity fosters greatness. The cliffs will rise steeply for a thousand feet; our motivation and strength come from no personal desire.)

I wish you and your family a Happy New Year and a very successful and prosperous 2013.

Yours sincerely,

Paul Kwan Sing Lam

Director of the State Key Laboratory in Marine Pollution

31st December 2012

在中國內地，國家重點實驗室是國家科技創新體系的重要組成部分。國家重點實驗室圍繞國家發展戰略目標、面向國際競爭，從事高水準基礎研究和應用基礎研究、聚集和培養傑出科學家、開展高層次學術交流，是增強國家科技儲備和原始創新能力的重要基地，具有崇高的學術榮譽。所以每輪國家重點實驗室的申請和評估，各高校和科研院所都傾盡全力爭取，競爭異常激烈。香港高校在2012年也遞交了二十多份夥伴國家重點實驗室的申請書，足以體現香港高校積極融入國家科研體系的決心和努力。

海洋污染國家重點實驗室(SKLM)自二零零九年獲批准成立以來，至今已走完第三個年頭。在缺乏大規模縱向經費的投入之下，如何加強實驗室成員的重點實驗室團結意識至今仍然是一項挑戰。海洋污染研究是多學科交叉、大空間尺度及需持之以恆的工作，我們堅信凝聚團隊的力量是唯一能令我們屹立於國家一流實驗室之列的堅實基礎和未來發展的動力源泉。

本年度，SKLM以“SKLM種子協作研究基金”(SCRF)、“城大內部博士後基金”(RFP)、“城大內部研究經費”(IRF)及儀器和科技開放平台，支援著成員們開展各類科研工作。儘管這樣的資助及支持對成員的研究工作仍只是杯水車薪，但充分展現了我們團結及凝聚團隊力量的策略和決心。SKLM成員在2012年度高等學校科學研究優秀成果獎（科學技術）評選中，獲得一項自然科學一等獎及一項自然科學二等獎。坦白說，目前SKLM取得的很多研究成果，除了取決於大多數成員精深的專業學識以外，更仰仗大家難能可貴的無私奉獻之品格。成員們高尚的人格魅力和強大的團隊凝聚力，已成為內地很多從事海洋科學研究高校和科研院所的楷模，大家是SKLM引以為傲的珍貴資源。

另外，我們欣慰地看到，海洋與人類健康研究中心（Research Centre for the Oceans and Human Health，簡稱H2O）經深圳市政府和香港城市大學的批准，已經正式在深圳成立了。雖然H2O成立至今只歷時短短的八個月，但是在SKLM各成員的努力以及香港城市大學、深圳虛擬大學園的支持下，H2O的各項工作已全面展開，成績斐然。現已獲得三項國家自然科學基金項目、一項“十二五”國家科技重大專項、一項深圳市戰略性新興產業發展專項項目，以及三項省市單位間合作項目等，經費總額達人民幣688萬元。這充分顯示了H2O在與內地各高校、科研機構開展學術交流、加強項目合作中所發揮的橋樑作用；同時也為SKLM成員在內地的發展構建了一個廣闊的平台。目前，在深圳虛擬大學園與國家海洋局第二海洋研究所蘇紀蘭院士的大力支持下，H2O已與九家從事海洋科學研究並落戶深圳的知名高等研究院校成立了“深圳海洋研究與技術聯盟”（Shenzhen Marine Research and Technology Consortium，簡稱SMART），旨在滿足深圳海洋研究與產業化發展的重大需求，加強深港創新圈科技合作，為國家實現海洋強國的夢想貢獻綿薄之力。

作為香港唯一從事海洋環境研究的國家重點實驗室，SKLMP對於推動兩岸三地海洋環境科學的學術交流與研究合作具有無可比擬的地緣優勢和責無旁貸的使命感。SKLMP抓住目前海峽兩岸前所未有的和平發展歷史契機，於2012年在香港舉辦了首屆“香江海洋論壇”，邀請海峽兩岸及香港的海洋生態環境專家與學者參與：共同探討了海洋生態環境的可持續發展戰略；通過提倡百家齊放、百家爭鳴，鼓勵有獨創性的思路與見解，從而啟迪各方挑戰傳統理論，提出創新思維，弘揚學術民主風氣；探索科學前沿，促進學科交叉與融合，共同提高兩岸三地整體海洋環境的研究水準。我們真誠希望各成員能以“香江海洋論壇”作為活躍的學術交流平台，定期開展海洋環境科學的學術論壇，從而確定“SKLMP種子協作研究基金”(SCRF)及“城大內部研究經費”(IRF)的資助項目，並挖掘面向國家重大基金申請的科學問題。

在開展科研工作的同時，SKLMP也不忘肩負的社會責任。為了提升公民海洋保護意識、幫助海洋科學工作者進一步瞭解海洋，以及通過開展社會公益活動回饋社會，SKLMP大力推廣名為“哪吒計畫”的潛水活動，該活動自9月開展至今參與人數已超過五十人，獲得了廣泛的好評及國內外十多家高校和科研院所知名學者的大力支持。

回顧2012，在各成員的不懈努力及海內外朋友的無私支持下，SKLMP有了良好的開端。展望2013，依舊前路漫漫、任重道遠。這更需要我們精誠團結，多一點包容，少一點功利，爭取在新的一年中取得長足進步。

衷心希望SKLMP將會成為我們向國家貢獻力量的基地、年輕科學家的夢工廠。期盼在大家的共同努力下，使香港及深圳成為中國南方海洋科學的重要研究中心!

謹在此借林則徐先生的名句為新春寄語，與各位同儕共勉：

海納百川，有容乃大；壁立千仞，無欲則剛。

並在此祝大家新年快樂!

謝謝大家!

林群聲

海洋污染國家重點實驗室(SKMP)主任

二零一二年十二月三十一日

Research Scopes in SKLMP

實驗室研究範疇

Academic Committee

Research Group 1

Research Group 2

Research Group 3

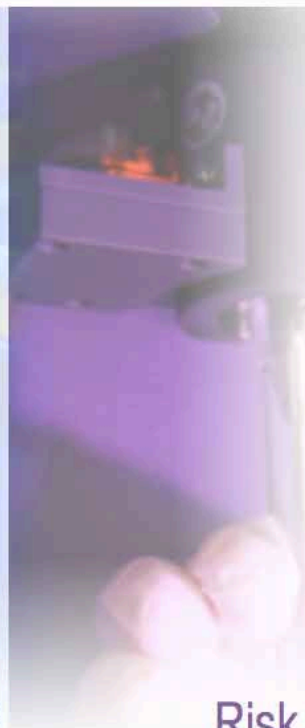
Research Group 4



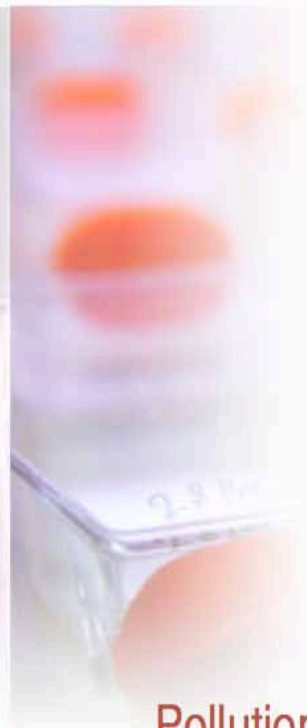
Pollution
Monitoring
Technology
污染檢測技術



Marine
Ecosystem
海洋生態
系統



Risk
Assessment
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Pollution
Control and
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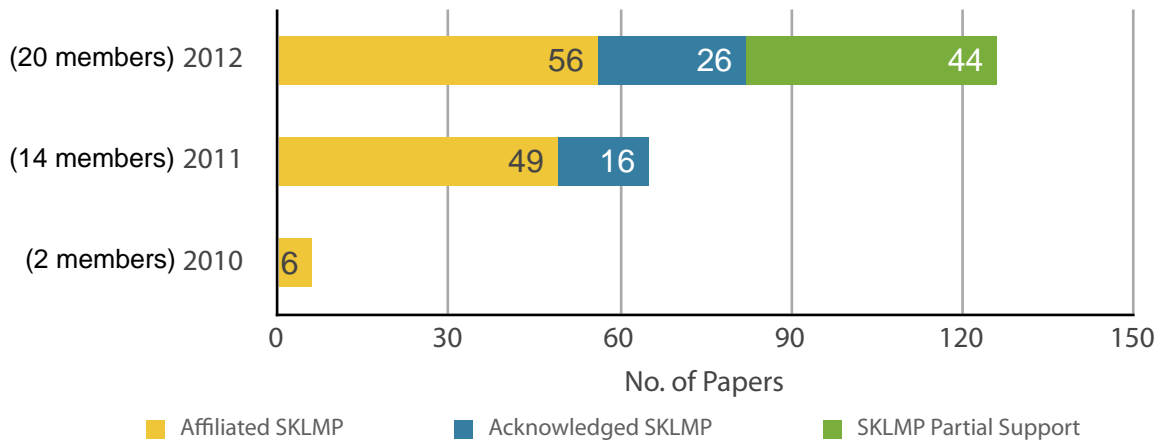
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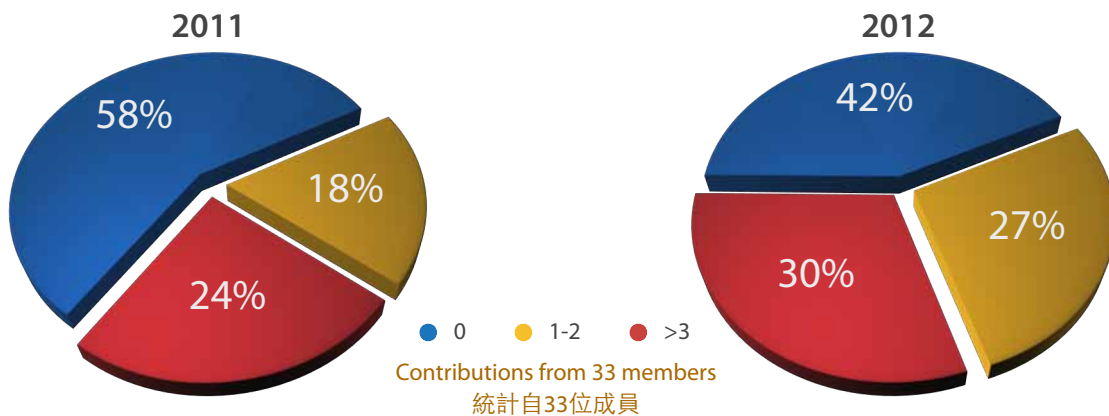
Research Highlights 研究亮點

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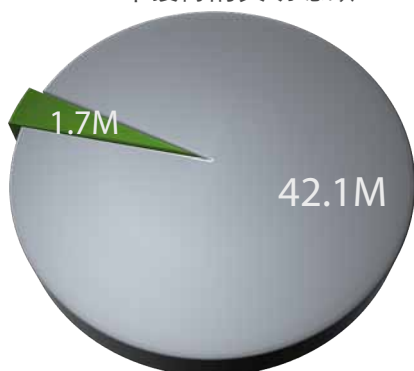
No. of Papers Published in Peer-reviewed SCI Journals with SKLMP included as the author's affiliation (2010-2012)
2010-2012 於SCI期刊發表的論文數目



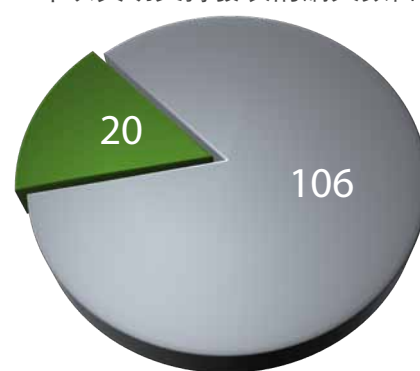
No. of SCI publications (with SKLMP affiliation) per member
成員發表的SCI論文數 (以SKLMP為科研合作單位)



Total Research Fund Received (2012)
2012年獲得的資助總額



Publications Arising from Different Fund Sources (2012)
2012年以資助支持發表的論文數目



● SKLMP Research Grants (IRSF and SCRF)
● Competitive External Grants

Part 1. Papers with the SKLMP included as the author's affiliation

以SKLMP為作者單位的期刊論文

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Part 3. Papers with the SKLMP Partial Support

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General and Comparative Endocrinology, (In Press)
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Phytotherapy Research, (In Press)
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Levels, Spatial Distribution and Sources of Selected Antibiotics in the East River (Dongjiang), South China.
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Androgen Rather than Estrogen Up-regulates Brain-type Cytochrome P450 Aromatase(*cyp19a1b*) Gene via Tissue-specific Promoters in the Hermaphrodite Teleost Ricefield Eel *Monopterus albus*.
Molecular and Cellular Endocrinology, 350:125-135.

Attendance at International Conferences & Titles of Presentations 出席的國際會議與標題介紹

Keynote Lecture

- 1 **Yang, M.M.** (Keynote Lecture)
Development of Multi-functional Nanoparticles for Cancer Diagnosis and Therapy.
Biomedical Engineering International Conference (BME2012).
5 Dec 2012. Hong Kong.
- 2 **Yang, M.M.** (Keynote Lecture)
Advances in Molecular Diagnostics and Perspectives on Personalized Medicine.
Forum on In Vitro Diagnostics.
24 Nov 2012, Chongqing, China.
- 3 **Au, D.W.T.** (Keynote Lecture)
Gender-Specific Modulation of Innate Immune Responses in Fish Under Environmental Stresses.
28th European Society for Comparative Physiology and Biochemistry.
2-5 Sep 2012, Bilbao, Spain.
- 4 **Yang M.M.** (Keynote Lecture)
Cell-cell Communication in Integrated Microfluidics and Differentiation of Mesenchymal Stem Cells on Nanomaterials.
9th World Biomaterials Congress.
1-5 Jun 2012, Chengdu, China.

Invited Lecture

- 1 Yang, M.M. (Invited Lecture)
Development of Multi-functional Nanoparticles for Target Destruction of Cancer Stem Cells.
Third Military Medical University.
23 Nov 2012, Chongqing, China.
- 2 Wu, R.S.S. (Invited Lecture)
Hypoxia: Nothing Could Be Worst!
The 2nd Congress on Climate Change.
15 Oct 2012, Mazatlan, Mexico.
- 3 Wu, R.S.S. (Invited Lecture)
Emerging Issues in Ocean Management: Contribution from Science and Solution.
The 6th East Asian Seas Congress.
9 Jul 2012, Changwon City, Korea.
- 4 Yu, P.K.N. (Invited Plenary Lecture)
Study of Low-Dose Radiation Effects in Zebrafish Embryos.
Hefei DNA Repair Mini-Symposium, Hefei Institutes of Physical Science, CAS.
19-20 Apr 2012, Hefei, Anhui, China.
- 5 Wu, R.S.S. (Invited Lecture)
Hypoxia: Problems and Scientific Challenges.
The United Nations Public forum on "Ocean Hypoxia and its Impact on Ecosystems and Economics"
18 April 2012, New York, USA.

- 6 Yang, M.M. (Invited Lecture)
Development of Microfluidics Technology for Drug Screening and Molecular Pharmacological Studies.
Baiyunshan Pharmaceuticals (Group) Ltd.
15 Mar 2012, Guangzhou, China.

Oral Presentation

- 1 **Wong, M.H.**
Removal Efficiencies of Toxic Chemicals in Sewage Treatment Works.
Annual Conference of Drainage Service Department.
Nov 2012, Hong Kong SAR.
- 2 Fei, Y.H., Li, X.Y.
Adsorption of Tetracyclines on Marine Sediment During Organic Matter Diagenesis.
IWA World Water Congress and Exhibition 2012.
Nov 2012, Busan, Korea.
- 3 **Li, X.D.**
Differentiating Anthropogenic Impacts on ARGs using Suitable Gene Indicators in the Pearl River Estuary, South China
International Conference on Persistent Toxic Substances (PTS)
22-26 Oct 2012, Miami, USA.
- 4 **Leung, K.M.Y.**
Water Effect Ratio and Its Application on Setting Site-specific Water Quality Criteria for Protecting Marine Ecosystems of Hong Kong.
SETAC Asia Pacific 2012 Meeting.
24-27 Sep 2012, Kumamoto, Japan.
- 5 **Murphy, M.B.**
Occurrence and Removal Efficiencies of Multiple Classes of UV filters and their Metabolites in Wastewater Treatment Plants.
SETAC Asia/Pacific Annual Meeting.
24-27 Sep 2012, Kumamoto, Japan.
- 6 Chen, W., Lau, S.W., Wu, R.S.S., Ge, W.
Neonatal Exposure to BPA or E2 Increases Female Ratio but Suppresses Ovarian Growth in the Zebrafish.
7th International Symposium on Fish Endocrinology.
3-6 Sep 2012, Buenos Aries, Argentina.
- 7 Lau, S.W., Zhang, L., Ngai, S.M., Ge, W.
Roles of Multi-functional Y-Box Binding Protein 1 (YB-1) in Zebrafish Ovarian Follicle Activation.
7th International Symposium on Fish Endocrinology.
3-6 Sep 2012, Buenos Aries, Argentina.
- 8 **Ge, W., Lau, S. W., Ngai, S. M., Zhang, L.**
Roles of Y-Box Binding Protein 1 (YB-1) in Zebrafish Ovarian Follicle Activation.
26th Conference of European Comparative Endocrinologists (CECE).
21-25 Aug 2012, Zurich, Switzerland.

- 9 Ip, R.H.L., Li, W.K.
Seemingly Unrelated Intervention Time Series Model for Effectiveness Evaluation of Large Scale Environmental Remediation.
Asia-Oceania Geosciences Society (AOGS-AGU) Joint Assembly.
15 Aug 2012, Singapore.
- 10 Wong, M.H.
Emerging Chemicals of Concern: Sources, Fates and Effects.
2012 Sino-European Symposium on Environment and Health.
Aug 2012, Galway, Ireland.
- 11 Rocke, E., Jing, H., Kataoka, T., Kong, L., Liu, H.B.
Phylogenetic Composition and Distribution of Picoeukaryotes in the Hypoxic Northwestern Coast of the Gulf of Mexico.
Protist 2012 conference.
29 Jul- 3 Aug 2012, Oslo, Norway.
- 12 Li, X.D.
Migration of Cu and Pb along the Soil Columns from E-waste Contaminated Soil.
International Symposium on Environmental Geochemistry
15-20 Jul 2012, Avevio, Portugal.
- 13 Ng, W.H., Rivkin, R.B., Liu, H.B., Chen, B.Z., Guo, C., Sun, M.M.
Effect of Nutrient Enrichment to Phytoplankton and Bacterial Growth and Grazing Mortality in Coastal Waters.
2012 ASLO Aquatic Sciences Meeting.
8-13 Jul 2012, Lake Biwa, Shiga, Japan.
- 14 Gui, C., Liu, H.B., Song, S., Zheng, L., Chen, B.
Dynamics of Picophytoplankton in the East China Sea.
2012 ASLO Aquatic Sciences Meeting.
8-13 Jul 2012, Lake Biwa, Shiga, Japan.
- 15 Kong, L., Kataoka, T., Buchwald, C., Jing, H., Liu, H.B.
Phylogenetic Diversity and Spatial Distribution of Hydrazine Oxidoreductase (HZO) Gene in the OMZ Off Costa Rica.
2012 ASLO Aquatic Sciences Meeting.
8-13 Jul 2012, Lake Biwa, Shiga, Japan.
- 16 Li, X.D.
Trace Metals in Atmospheric Particular Matters over the Northern South China Sea (SCS): Regional Sources and Long-range Atmospheric Transport.
Goldschmidt Geochemistry Conference 2012
25-29 Jun 2012, Montreal, Canada.
- 17 Yang, M.M.
Using Integrated Microfluidics to Study Intracellular Calcium Response to Mechanical Stimulation.
The 4th International Symposium on Microchemistry and Microsystems.
10-13 Jun 2012, Hsingchu, Taiwan.
- 18 Yang, M.M.
Screen Printing of Solder Resist on Master Substrates for Fabrication of Multi-Level Microfluidic Channels and Flask-shaped Microstructures.
The 4th International Symposium on Microchemistry and Microsystems.
10-13 Jun 2012, Hsingchu, Taiwan.

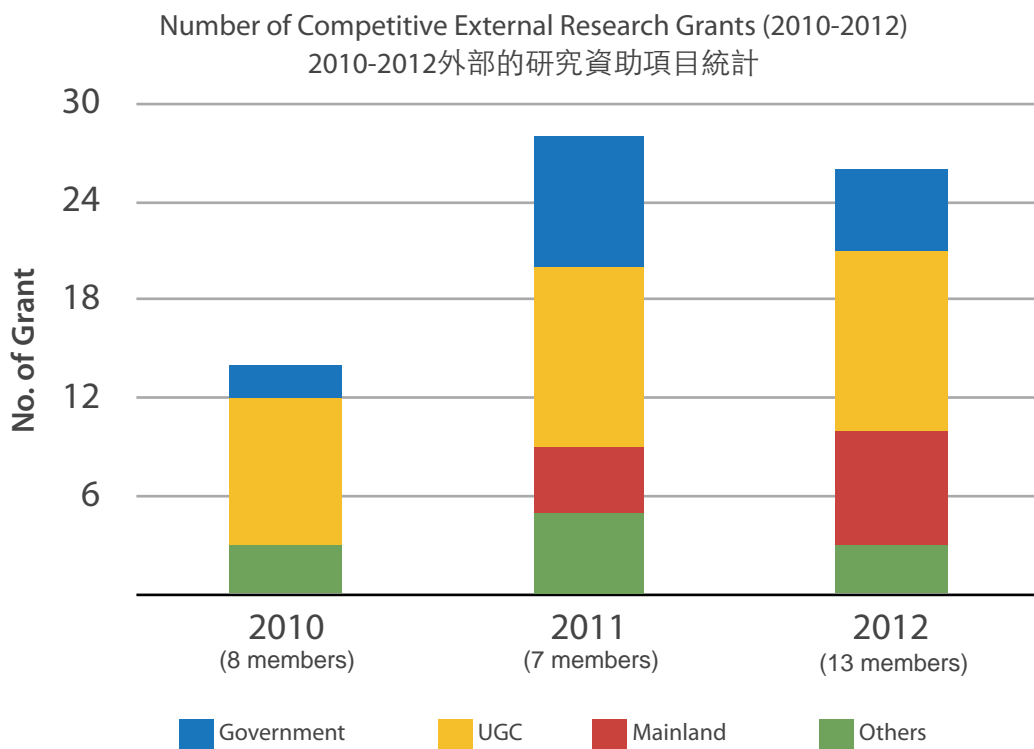
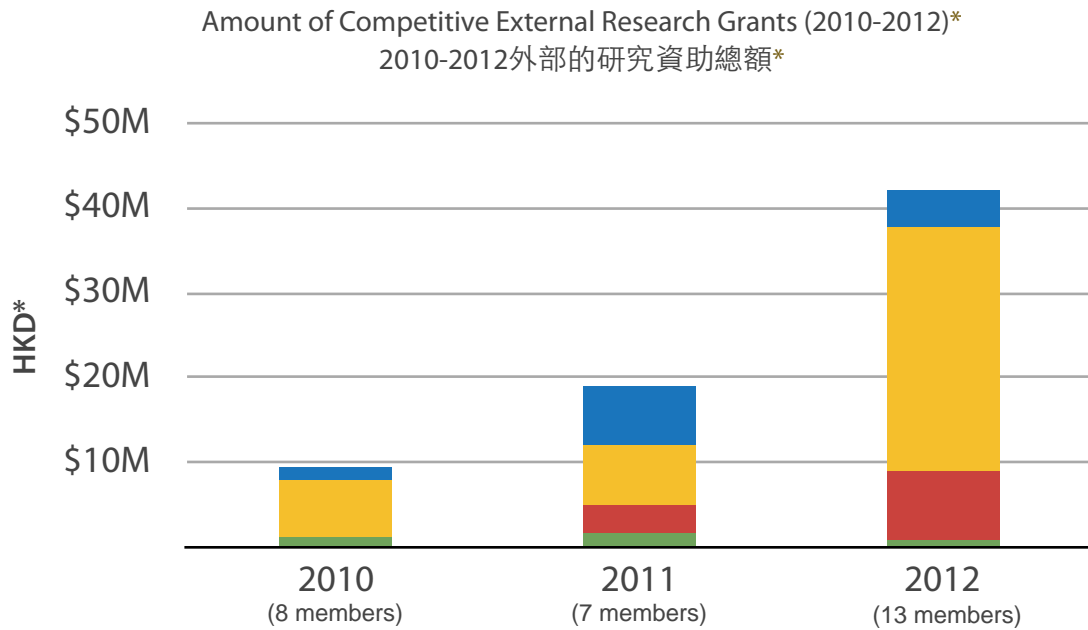
- 19 **Leung, K.M.Y.**
A Closer Look at the Temperature-Dependent Chemical Toxicity to Aquatic Organisms and its Implication on Derivation of Water Quality Guidelines for Protecting Aquatic Life.
6th SETAC World Congress.
20-24 May 2012, Berlin, Germany.
- 20 **Murphy, M.B.**
Measurement of Trace Element and Perfluorinated Compound (PFC) Concentrations and Species Identification of Shark Fins from the Hong Kong Market.
6th SETAC World Congress.
20-24 May 2012, Berlin, Germany.
- 21 **Ge, W., Lau, S.W., Zhang, L.**
Molecular Mechanism of Follicle Recruitment in the Zebrafish Ovary – Potential Gate-Keeping Roles of Y-box Binding Protein 1 (YB-1).
7th Congress of Asia and Oceania Society for Comparative Endocrinology (AOSCE).
3-7 Mar 2012, Kuala Lumpur, Malaysia.
- 22 **Choi, V.W.Y., Konish, T., Oikawa, M., Cheng, S.H., Yu, K.N.**
Threshold Number of Protons for Inducing Adaptive Response in Zebrafish Embryos.
International Symposium on the Natural Radiation Exposures and Low Dose Radiation Epidemiological Studies, Hirosaki University.
29 Feb - 3 Mar 2012, Aomori, Japan.
- 23 **Choi, V.W.Y., Ng, C.Y.P., Kong, M.K.Y., Cheng, S.H., Yu, K.N.**
Adaptive Response to Ionizing Radiation Induced by Cadmium in Zebrafish Embryos.
International Symposium on the Natural Radiation Exposures and Low Dose Radiation Epidemiological Studies, Hirosaki University.
29 Feb - 3 Mar 2012, Aomori, Japan.
- 24 **Ng, C.Y.P., Choi, V.W.Y., Lam, A.C.L., Cheng, S.H. Yu, K.N.**
Multiple Stressor Effect in Zebrafish Embryos from Simultaneous Exposures to Ionizing Radiation and Cadmium.
International Symposium on the Natural Radiation Exposures and Low Dose Radiation Epidemiological Studies, Hirosaki University.
29 Feb -3 Mar 2012, Aomori, Japan.
- 25 **Wu, H., Yang, Z., Au, D.W.T., Guo, B., Lin, Z., Liu, Z., Wei, F. Li, G., Liao, W., Qin, L., Hung, L., Wei, L., Zhang, G.**
A Novel Ionizable Cationic Lipid Nanoparticle-based Delivery System for Potential RNAi Therapy in Age-related Cartilage Degradation.
Annual Meeting of Orthopedics Research Society.
7-11 Feb 2012, San Francisco, USA.

Research Grants

研究資助

Competitive External Research Grants

外部的研究資助



*1RMB=1.24HKD

Grants from Hong Kong 香港科研資助

Government 香港政府

Project Title *	Grant Type	Investigator(s)	Amount (HK\$)
2012			
1 Survey of Juvenile Fish Resources at the Three Marine Parks at Northeast Hong Kong 香港東北地區三個海岸公園的幼魚資源調查	Agriculture Fisheries and Conservation Department 漁農自然護理署	PI: Leung, K.M.Y.	798,000
2 Dosing of Ferric Iron for the Control of Odour Problems in Typhoon Shelters 三價鐵離子用於解決颱風避難所的空氣臭味問題的研究	Environment and Conservation Fund 環境及自然保育基金	PI: Li, X.Y.	1,058,500
3 Field Sampling, Species Identification and Data Analysis of Benthic Infaunal Communities of Hong Kong Marine Waters 香港水域底棲水生動物群落的採樣、種類鑒定和數據分析	Environmental Protection Department 環境保護署	Co-I: Leung, K.M.Y.	800,000
4 Analysis of the Relationship between Marine Water Quality Parameters and Climatic and Other Environmental Factors 海洋水質參數與氣候及其他環境因子的關係研究	Environmental Protection Department 環境保護署	PI: Li, W.K. Co-I: Leung, K.M.Y.	515,000
5 Fish Resources Study for the Proposed Marin Park in the Brothers Islands	Highways Department 路政署	PI: Lam, P.K.S.	1,180,000
Subtotal			4,351,500
2011			
6 Effect of Vaccine Combined with Traditional Chinese Medicine on Prevention of Disease in Grey Mullet 結合傳統中藥的疫苗對鯔魚疾病防治的研究	Agriculture Fisheries and Conservation Department 漁農自然護理署	PI: Wong, M.H.	410,000
7 Provision of Services for the Technical Review and Statistical Analysis of the Datasets of Waterbird Monitoring Programme for the Deep Bay Area and Baseline Ecological monitoring programme for the Mai Po Inner Deep Bay Ramsar Site 后海灣水鳥監控程序數據集和米埔內后海灣拉姆薩爾國際重要濕地生態基線監控程序數據集的技術評審和統計分析服務供給	Agriculture Fisheries and Conservation Department 漁農自然護理署	PI: Lam, P.K.S.	793,500
8 Removal Efficiencies of Toxic Chemicals in Sewage Treatment Works in Hong Kong 香港污水處理廠有毒化學物質的清除效率研究	Drainage Services Department 渠務署	PI: Wong, M.H.	900,000

* 項目名稱以英文譯本為準

Government 香港政府

	Project Title *	Grant Type	Investigator(s)	Amount (HK\$)
9	Integrated Fish Pond Farming Using Food Processing Waste: for Quality Fish Production and Habitat Conservation 利用食品加工廢棄物用於池塘漁業養殖：用於高質量魚產品的養殖以及生境保護	Environment and Conservation Fund 環境及自然保育基金	PI: Wong, M.H.	1,900,000
10	International conference on deriving environmental quality standards for the protection of aquatic ecosystems (EQSPA-2011)	Environment and Conservation Fund 環境及自然保育基金	PI: Leung, K.M.Y.	495,800
11	A review of marine biodiversity and ecological surveys in Hong Kong	Environment and Conservation Fund 環境及自然保育基金	PI: Leung, K.M.Y.	456,380
12	Review and Development of Marine Water Quality Objectives-feasibility Study 海水水質目標的回顧與發展-可行性分析	Environmental Protection Department 環境保護署	Co-I: Kong, R.Y.C.	860,000
13	High Efficiency-multifunction-green-vertical Municipal Wastewater Treatment System: Development and Demonstration 高效多功能綠色直立城市廢水處理系統的發展與示範	Innovation and Technology Fund 創新及科技基金	PI: Tam, N.F.Y.	1,080,000
Subtotal				6,895,680
2010				
14	Qualification of Antibiotic Residues and Microbial Antibiotic-resistant Genes in Fish Ponds and Marine Culture Zones of Hong Kong 抗生素殘留以及香港魚塘和海洋養殖區微生物抗生素耐藥基因的定量分析	Environment and Conservation Fund 環境及自然保育基金	Co-I: Leung, K.M.Y.	499,880
15	Development of Highly Efficient Semiconductor Nanoparticles as Photocatalysts for the Degradation of Organic Pollutants in Water under Visible Light 可作光催化劑對水中有機污染物進行可見光降解的高效半導體納米粒子的開發	Innovation and Technology Fund 創新及科技基金	PI: Lau, T.C.	998,430
Subtotal				1,498,310

UGC 教資會

	Project Title *	Grant Type	Investigator(s)	Amount (HK\$)
2012				
1	Centre for Marine Environmental Research and Innovative Technology	Areas of Excellence Scheme 卓越學科領域計劃	PI: Wu, R.S.S.	20,500,000
2	Air-surface Exchange of Persistent Organic Pollutants (POPs) and Heavy Metals (MNs) in Peri-urban Agricultural Ecosystems of the Pearl River Delta, South China	NSFC/RGC 國家自然科學基金/研究資助局	PI: Li, X.D.	863,800
3	Pharmaceuticals in Municipal Sewage Treatment Works of China: Behaviour and Risk Assessment 城市污水處理廠污水中殘留藥物的性狀及其風險評估	NSFC/RGC 國家自然科學基金/研究資助局	PI: Lam, P.K.S.	820,000
4	Health Risk Assessment of Toxic Trace Elements and Polycyclic Aromatic Hydrocarbons (PAHs) via Indoor Dust from Coal-burning Households	NSFC/RGC 國家自然科學基金/研究資助局	PI: Wong, M.H.	780,000
5	Sources and Biodynamics of Mercury in Marine Fish in Hong Kong Coastal Waters 香港沿海水域海洋魚體內汞的來源和生物動力學研究	General Research Fund 優配研究金	PI: Wang, W.X.	1,200,000
6	PBDE: Bioaccumulation, Maternal Transfer and Effects on Darwinian Fitness Traits in Multiple Generations of Marine Gastropod	General Research Fund 優配研究金	Co-I: Wu, R.S.S	764,711
7	Response of the Phytoplankton, Microzooplankton and the Prokaryotic Communities to Atmospheric Dust Deposition: an -Omics Approach 採用組學方法研究大氣灰塵沈積物對浮游植物、微型浮游動物和原核生物群落的影響	General Research Fund 優配研究金	PI: Liu, H.B.	950,000
8	Mechanistic Study on the Organotin-Mediated Imposex in the Rock Shell Thais Clavigera using RNA-sequencing Analysis 採用RNA測序分析技術研究有機錫誘發的荔枝螺性畸形機理	General Research Fund 優配研究金	PI: Leung, K.M.Y.	850,000
9	Oxidation Chemistry of Manganese Nitrido Complexes 氮化錳複合物的化學氧化研究	General Research Fund 優配研究金	PI: Lau, T.C.	813,750

* 項目名稱以英文譯本為準

UGC 教資會

	Project Title *	Grant Type	Investigator(s)	Amount (HK\$)
10	Integration of Biomimetic Microparticle-based Multiplexed Assays and Microfluidic Systems for High Throughput Quantitative Detection and Discrimination of Endocrine Disrupting Chemicals 聯合基於多復路分析的仿生微粒技術與微流體系統用於高通量辨別與檢測內分泌干擾物的研究	General Research Fund 優配研究金	Co-I: Yang, M.M.S.	775,000
11	Photoelectrode for (MEC) Concurrent Hydrogen Generation and Waste Organic Degradation 光電極用於微生物電解池的研究：制氫和有機廢棄物的降解	General Research Fund 優配研究金	PI: Li, X.Y.	500,000
Subtotal				28,817,261
2011				
12	Functional Analyses of HIF Transcription Factors and Associated MicroRNAs in the Human H295R Adrenocortical Cell Line 人腎上腺皮質細胞系H295R的HIF轉錄因子及其關聯微RNAs的功能分析研究	Areas of Excellence Scheme 卓越學科領域計劃	PI: Kong, R.Y.C.	800,000
13	Comparison of the acute sensitivity to chemicals of tropical and temperate aquatic animal species: Meta analysis and mechanistic studies	General Research Fund 優配研究金	PI: Leung, K.M.Y. Co-I: Lam, M.H.W.	710,000
14	Waterborne Viral Pathogen Detection Technologies 水源病原菌檢測技術研究	Areas of Excellence Scheme 卓越學科領域計劃	PI: Kong, R.Y.C.	200,000
15	Human Leukocyte Antigen (HLA) Homologues in Zebrafish: a Key to Understanding Hypoxia-mediated Modulation of Offspring Sex Ratios and Reproductive Functions in Fish 斑馬魚中的人白細胞抗原同系物是解釋為甚麼低氧可誘導調節斑馬魚後代性別比例以及生殖功能的關鍵	Areas of Excellence Scheme 卓越學科領域計劃	PI: Kong, R.Y.C.	100,000
16	Biokinetics, Bioavailability, and Chronic Toxicity of Metal Nanoparticles in Aquatic Organism 水生生物中金屬納米粒子的生物動力學、生物利用度以及慢性毒性的研究	General Research Fund 優配研究金	PI: Wang, W.X.	1,050,000
17	Uncovering the Molecular Links between Hypoxia and Endocrine Disruption: a Functional Study of Zebrafish Leptin 低氧與內分泌物分解之間的分子聯繫：斑馬魚瘦素的功能研究	General Research Fund 優配研究金	PI: Kong, R.Y.C.	1,000,000

UGC 教資會

	Project Title *	Grant Type	Investigator(s)	Amount (HK\$)
18	The Functional Roles of Zebrafish irx1a Gene in Heart Development and Regeneration 斑馬魚irx1a基因在心臟發育和再生中的作用研究	General Research Fund 優配研究金	PI: Cheng, S.H.	820,000
19	Risk Assessment and Remediation of Cadmium Contamination in Registered Vegetable Farms in the Pearl River Delta Region 珠江三角洲區域蔬菜農場的鎘污染風險評估及其修復	General Research Fund 優配研究金	PI: Wong, M.H.	780,000
20	Assessment of Benthic Community Health in Subtropical Waters Using Biological Indices and Life-trait Analysis 應用生物指數和生命特徵分析法評估亞熱帶海域底棲生物群落健康狀況	General Research Fund 優配研究金	PI: Shin, P.K.S.	700,000
21	Measurement and Assessment of Novel Halogenated Flame Retardants in Waterbirds and Marine Cetaceans in Hong Kong 香港水域水鳥和鯨類動物中新興阻燃劑的測量和評估	General Research Fund 優配研究金	PI: Lam, P.K.S.	460,000
22	UGC AoE Sustained Funding Sub-project 大學教育資助委員會-卓越學科領域計劃維持的子研究項目	University Grants Committee 大學教育資助委員會	PI: Tam, N.F.Y.	410,000
Subtotal				7,030,000
2010				
23	Marine Environmental Research and Innovative Technology, MERIT	Areas of Excellence Scheme 卓越學科領域計劃	PI: Au, D.W.T.	700,000
24	Automated Micro/Nano-scale Execution of Tasks with Multiple Biological Cells Using a Table-Top Robotic Bio-manipulation System 使用台式機械人生物操縱系統對多種生物細胞自動執行微/納米級任務	Competitive Earmarked Research Grant 角逐研究用途補助金	PI: Cheng, S.H.	1,484,960
25	C-H Bond Activation by Nitrido Imido and Amido Complexes of Ruthenium bearing Salen Ligands 具有Salen配基的鈦的Nitrido, Imido, Amido 絡合物對碳氫鍵的活化作用	Competitive Earmarked Research Grant 角逐研究用途補助金	PI: Lau, T.C.	1,115,000
26	From Genes to Behavior: the Roles of Iroquois Genes in the Development and Function of Zebrafish Embryonic Retinal Neurons 從基因至行為: Iroquois 基因在斑馬魚胚胎視網膜神經發育與功能中的作用	Competitive Earmarked Research Grant 角逐研究用途補助金	PI: Cheng, S.H.	1,034,994

* 項目名稱以英文譯本為準

UGC 教資會

	Project Title *	Grant Type	Investigator(s)	Amount (HK\$)
27	Nanotherapeutics in Angiogenesis: Synthesis and in Vivo Assessment of Drug Efficacy and Biocompatibility in the Zebrafish Embryos 血管新生的納米療法: 合成及在斑馬魚胚胎體內評估藥物療效及生物相容性	Competitive Earmarked Research Grant 角逐研究用途補助金	PI: Cheng, S.H.	740,565
28	Understanding the relationship between gender, estrogen and telomere biology in fish: Towards developing an alternative vertebrate model for aging studies	General Research Fund 優配研究金	PI: Au, D.W.T	730,000
29	Action Mechanisms of Endocrine Disrupting Chemicals in Fish Reproductive Axis 內分泌乾擾物對魚類生殖的作用機制	University Grants Committee 大學教育資助委員會	PI: Ge, W. Co-I: Wu, R.S.S. Co-I: AU, D.W.T.	500,000
30	UGC AoE Sustained Funding Sub-project 大學教育資助委員會-卓越學科領域計劃維持的子研究項目	University Grants Committee 大學教育資助委員會	PI: Cheng, S.H.	285,000
31	Chinese Medicine Research and Further Development 中藥研究與發展	University Grants Committee 大學教育資助委員會	PI: Cheng, S.H.	166,500
Subtotal				6,757,019

Others 其他

	Project Title *	Grant Type	Investigator(s)	Amount (HK\$)
2012				
1	Identification of Mammalian Species Used in Food by Flow-Through DNA Hybridization System 採用導流DNA雜交技術識別食物中哺乳動物的種類	CityU Applied Research Grant	PI: Cheng, S.H.	170,235
2	Photosynthetic and Proteomic Responses of the Marine Diatom <i>Thalassiosira Pseudonana</i> to Triphenyltin Exposure 假微型海鏈藻對三苯基錫的光合以及蛋白組學的響應研究	HKU (small project grant)	PI: Leung, K.M.Y.	80,000
3	Interactions among Biodegradable Chelants, Soil Microbes, and Plant Roots in the Phytomanagement Process of Metal-contaminated Soils	Hong Kong Scholar Program 香江學者計劃	PI: Li, X.D.	600,000
Subtotal				850,235
2011				

Others 其他

	Project Title *	Grant Type	Investigator(s)	Amount (HK\$)
4	Futian-CityU Mangrove Research and Development Centre 香港城市大學福田城大紅樹林研發中心	CityU Applied R&D Grant	PI: Tam, N.F.Y.	850,000
5	The Developmental Toxicity of UV Sunscreens 紫外光遮光劑的發育毒性研究	CityU Strategic Research Grant	PI: Cheng, S.H.	180,000
6	Understanding the Estrogen Dynamics and Longevity Gender Gap in Medaka 青鱈魚雌性激素動力學以及雌雄壽命差異的研究	CityU Strategic Research Grant	PI: Au, D.W.T.	180,000
7	An Ecological Study of Horseshoe Crab Spawning and Nursery Beach for Conservation Purposes: Ha Pak Nai, Hong Kong 香港下白泥馬蹄蟹的產卵及其哺育海灘的生態學保護研究	Ocean Park Conservation Foundation Hong Kong 香港海洋公園保育基金	PI: Shin, P.K.S.	250,000
8	Impact of Sea-level Rise on Protection and Management of Coastal Mangrove Wetland 海平面上升對沿海紅樹林濕地保護與管理的影響研究	Ocean Park Conservation Foundation Hong Kong 香港海洋公園保育基金	PI: Tam, N.F.Y.	200,000
Subtotal				1,660,000
2010				
9	Time-series and Spatial Statistical Studies on Marine Water Quality Monitoring Data in Hong Kong: Implications of the Effectiveness of Environmental Policy and Management, and Definition of Water Pollution Control Zones 對香港海洋水質監測數據的時間和空間統計研究: 對環境政策及管理有效性的影響, 以及水污染管制區的定義	HKU Seeding Funding for Small Project	PI: Leung, K.M.Y. Co-I: Li, W.K.	72,000
10	Smart Ambience for Affective Learning (SAMAL): An Innovative Exploration of Smart Ambience for Integrating Affect and Cognition in Learning Life Science and Information Management 智慧情境的情感學習: 智慧情境對整合在學習生命科學與信息管理中產生的情感與認知的創新探索	Teaching Development Grant, CityU	Co-I: Cheng, S.H.	800,000
11	Ocean Dynamics and Modeling Program	Research Project Competition (HKUST)	PI: Gan, J.P.	300,000
Subtotal				1,172,000

* 項目名稱以英文譯本為準

Grants from Mainland China 內地科研資助

Mainland 內地

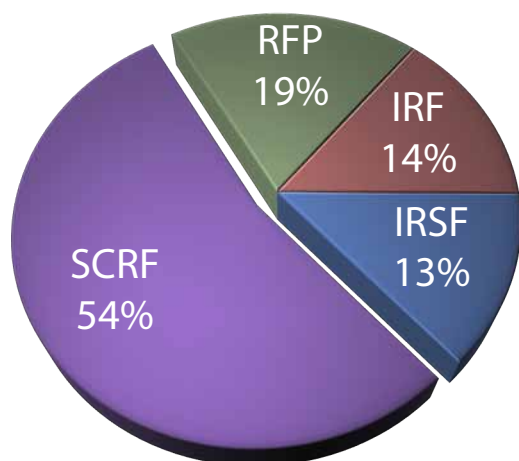
	Project Title *	Grant Type	Investigator(s)	Amount (RMB¥)
2012				
1	分解水和還原二氧化碳的高效光/電催化體系研發及機理研究	2012年度深圳市基礎研究項目	PI: Lau, T.C.	600,000
2	Development of Nanotechnology-based Detection Platform for Early Diagnosis of Lung Cancer"	National Program on Key Basic Research Project of China (973 Program) 國家重點基礎研究發展計劃項目(973計劃)	Co-I: Yang, M.M.S.	830,000
3	Biokinetics and Toxicology of Heavy Metals in Southern China Estuaries	National Science Foundation of China 國家自然科學基金	PI: Wang, W.X.	3,000,000
4	Study of the Key Factors that Influence the Flux of Ciguatera Toxins through Marine Food Webs	National Science Foundation of China 國家自然科學基金	PI: Chan, L.L.	760,000
5	Assessment and Characterization of Novel and Unknown Per- and Polyfluorinated Compounds in Pearl River and Yangtze River Delta (2013-2016) 珠江三角洲和長江三角洲新型與未知全氟化合物污染狀況及其生態風險 (2013-2016)	National Science Foundation of China 國家自然科學基金	PI: Lam, P.K.S.	740,000
6	Assessment of Conventional & Emerging Halogenated Flame Retardants in Two Estuaries of China: Pearl River Delta and Yangtze River Delta (2013-2015) 中國珠江與長江河口地區傳統及新興鹵系阻燃劑環境分布特徵與生態風險評估 (2013-2015)	NSFC Project for Young Scientists 國家自然科學基金青年基金	PI: Lam, J.C.W.	280,000
7	Development, Optimization and Validation of Methods for Isolation, Purification and Trace Analysis of CTXs 雪卡毒素高純度提取與痕量分析技術研究	Shenzhen strategic emerging industry development special project 深圳市戰略性新興產業發展專項資金	PI: Lam, P.K.S. Co-I: Chan, L.L.	300,000
Subtotal				6,510,000
2011				
8	Health Risk Assessment of Toxic Trace Elements and PAHs via Indoor Dust from Coal-burning Households in Rural China	National Science Foundation of China 國家自然科學基金	PI: Wong, M.H.	900,000

Mainland 內地

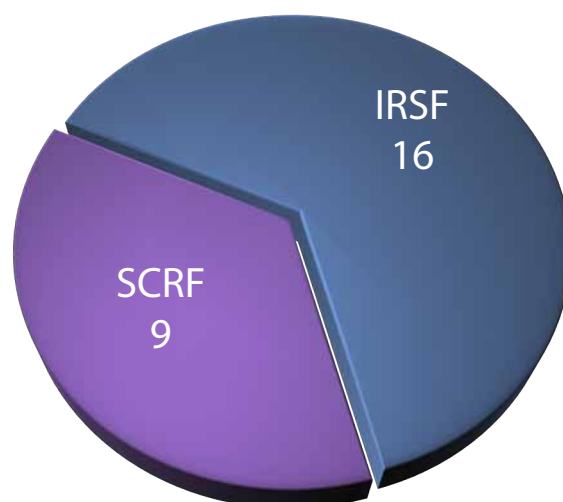
	Project Title *	Grant Type	Investigator(s)	Amount (RMB¥)
9	Establish the Research Centre for Ocean and Human Health	Shenzhen Municipality	PI: Lam, P.K.S.	400,000
10	Eco-remediation Technology and Demonstration of the Coastal Wetland in Shenzhen Bay	Shenzhen Municipality	PI: Tam, N.F.Y.	200,000
11	Demonstration of Circular Economy: Study on the Eco-remediation of Shenzhen Overseas Chinese Town	Shenzhen Overseas Chinese Town Holding Company	PI: Tam, N.F.Y.	1,200,000
Subtotal				2,700,000

The SKLMP Research Grants SKLMP的研究資助

Distribution of SKLMP Grants (2010-2012)
SKLMP科研經費分配(2010-2012)



Publications Arising from SKLMP Grants (2010-2012)
研究經費支持發表的論文(2010-2012)



Funded by the CityU Research Centre Annual Fund 獲研究中心年度基金資助的項目情況

CityU Internal Research Fellowship Programme (RFP) 城大內部博士後基金

	Project Title	Supervisor	Amount (HKD\$)
Jan 2013 - Dec 2014			
1	A Holistic Approach to Unravel Xenoestrogen Induced Immunosuppressive Effects and Reproductive Impairment in Fish: Implications for Risk Assessment and Monitoring of Immunosuppressants in Waters	Au, D.W.T.	400,000
2	Comparative Investigation on Resistance and Defense Mechanism of Coral Reef Fishes to Ciguatoxins (CTXs)	Chan, L.L.	400,000
3	Significance of Roots and Microorganisms in Phytoremediation of Polybrominated Diphenyl Ethers (PBDEs) by Mangrove Plants	Tam, N.F.Y.	400,000
4	The Regenerative Toxicology of Alpha Emitters in the Caudal Fin of Marine Medaka: Linking Radiation Induced Effects from Molecular to Physiological Levels	Cheng, S.H.	400,000

CityU Internal Research Fund (IRF)
城大內部研究經費

	Project Title	Investigator	Amount (HKD\$)
Jan 2013 - Dec 2014			
1	Expression of Plasma Immune Proteins is Gender-dependent and Related to Fish Susceptibility to Pathogen	Au, D.W.T.	200,000
2	In-situ Measurement of Variations in Solar Ultraviolet Radiation Penetration through Marine Water	Yu, P.K.N.	200,000
3	Phytoremediation of Polybrominated Diphenyl Ethers (PBDEs) by Mangrove Wetlands	Tam, N.F.Y.	200,000
4	Responses of Marine Organisms to Ocean Acidification: Does Ecosystem Matter?	Cheng, S.G.	200,000
5	Risk Assessment of Pharmaceutical Residues and Occurrence of Antibiotic-resistant Bacterial Genes and Strains in Hong Kong Surface Waters and Sediments	Murphy, M.B.	200,000
6	The Regenerative Toxicology of Alpha Emitters in the Caudal Fin of Marine Medaka: Linking Radiation Induced Effects from Molecular to Physiological Levels	Cheng, S.H.	200,000

CityU Internal Research Seed Fund (IRSF) 城大內部研究種子基金

	Project Title	Investigator	Amount (HKD\$)
Jan 2011 - Dec 2012			
1	Development of Novel Technology for Early Diagnosis and Monitoring of Immunotoxic Pollutants in Marine Environment	Au, D.W.T.	100,000
2	Quantification of Poly- and Perfluorinated Compounds (PFCs) and Species Identification of Shark Fins Purchased from Hong Kong Seafood Shops	Murphy, M.B.	100,000
Jan 2010 - Dec 2011			
3	Development of Highly Efficient ZnO Tetrapods Nanoparticles for Photodegradation of Organic Pollutants in Water under Visible Light	Lau, T.C.	150,000
4	International Collaborative Research on Endocrine Disrupting Compounds (EDC) and Emerging Persistent Organic Pollutants (POPs) in South China Sea	Cheng, S.H.	200,000
5	Long-term Measurements of Ultraviolet Radiation in Marine Environments in Hong Kong	Yu, P.K.N.	150,000
6	Sorption and Degradation of Polybrominated Diphenyl Ethers (PBDEs) by Green Microalgae with and without the Effect of Metals	Tam, N.F.Y.	200,000
7	Unraveling Tissue-specific Mechanisms for <i>in vivo</i> Regulation of Estrogen Target Genes in Medaka	Au, D.W.T.	200,000

Funded by the Innovation and Technology Fund for Partner State Key Laboratory (PSKLs) from Innovation and Technology Commission
獲創新科技署國家重點實驗室專項基金資助的項目

SKLMP Seed Collaborative Research Fund (SCRF)

SKLMP種子協作研究基金

Project Title	Investigator	Amount (HKD)
Jan 2012 - Dec 2014		
1 Assessing the Impacts of Organic and Metal Pollution on Symbiotic Microbial Communities in Marine Corals and Sponges by Metagenomics and Transcriptomics Approaches	Qian, P.Y. (HKUST) Wang, W.X. (HKUST) WU, R.S.S. (HKU) Qiu, J.W. (HKBU) Lee, O.O. (HKUST) Chiu, J.M.Y. (HKU)	900,000
2 Establishing the Green Lipped Mussel <i>Perna viridis</i> as a Universal Marine Model Organism and Pollution Biomonitor for Ecotoxicology and Environmental Genomics	Leung, K.M.Y. (HKU) Lam, P.K.S. (CityU) Wong, C.K.C. (HKBU) Chan, L.L. (CityU)	900,000
3 Health Risk Assessments of Residents in the Pearl River Delta exposed to Brominated Flame Retardants (BFRs)	Wong, M.H. (HKBU) Wang, H.S. (HKBU) Man, B.Y.B. (HKBU) Wu, S.C. (CityU) Lam, P.K.S. (CityU) Wong, C.K.C. (HKBU) Jones, P.D.(University of Saskatchewan, Canada) Giesy, J.P. (University of Saskatchewan, Canada)	900,000
4 Interactive Effects of Climate Change and Hypoxia on Fish Sex Determination: Estrogen synthesis and Masculinisation 2011	Kong, R.Y.C. (CityU) Wu, R.S.S. (HKU) Yu, R.M.K. (University of Newcastle, Australia)	900,000
5 Sources and Bioaccumulation of Mercury and Cadmium in the Pearl River Estuary (PRE) and Hong Kong Coastal Waters	Li, X.D. (PolyU) Wang, W.X. (HKUST)	900,000

Abstracts of the Research Fellowship Programme (RFP) Projects

RFP 項目摘要

Jan 2013 - Dec 2014 (Newly Funded)

RFP

A HOLISTIC APPROACH TO UNRAVEL XENOESTROGEN INDUCED IMMUNOSUPPRESSIVE EFFECTS AND REPRODUCTIVE IMPAIRMENT IN FISH: IMPLICATIONS FOR RISK ASSESSMENT AND MONITORING OF IMMUNOSUPPRESSANTS IN WATERS

Doris Wai Ting AU, Terrance Chi Kong LAU, Joseph Liam HUMBLE

Earlier studies on chronic toxicity of xenoestrogens or estrogen active chemicals (EACs) in fish were largely focused on adverse outcomes related to reproductive impairment. An increasing number of field and laboratory studies have shown that many EACs in environment could depress the expressions of major immune proteins and increase fish susceptibility to pathogens. The findings support the immunosuppressive role of EACs in fish. Surprisingly, no studies have ever been conducted to evaluate the manifestation of EACs induced adverse effects on fish immune function and reproduction concomitantly in the same fish population. Such information is crucial for a holistic assessment of the risk of EACs in waters. Moreover, female fish have a much higher level of endogenous E2 than the male counterpart. It is not known how and in what ways the immune system of male and female fish may respond differently to xenoestrogens. Could such disparity lead to a gender-difference in host susceptibility to pathogens? The answers will shed light on the impact of EACs on reducing population density and shifting the sex-ratio of a population, which are of great implication for ecological risk assessment. 17 α -Ethinylestradiol (EE2), a ubiquitous xenoestrogen in sewage waters, will be used as the model EAC to investigate the dose response, NOEC and LOEC of EE2 on immune and reproductive impairments in both male and female marine medaka (*Oryzias melastigma*) which has recently been established as a model fish for immunotoxicology. Rifampicin (RIF) will be used as the reference immunosuppressant to determine the immunosuppressive effects and potency of EE2 in both male and female medaka.

Development of immune biomarkers are urgently needed for early detection, risk assessment and monitoring of immunosuppressive chemicals in aquatic environments. A comprehensive understanding on the major biological pathways/network that can be modulated by immunosuppressant and their link to the adverse outcome on fish immunity (i.e. increased susceptibility to pathogens infection) are essential for the identification of major immune genes and relevant immune function endpoints (biomarkers) in fish that can be extrapolated to significant ecological effect. Next generation sequencing technology will be used to screen and

identify major immune pathways and genes that are altered by RIF / EE2. The relationship between perturbations of these molecular endpoints and impairment of host immunocompetence will be established. The findings of this study will contribute a major breakthrough in risk assessment of EACs and immunosuppressants in waters.

COMPARATIVE INVESTIGATION ON RESISTANCE AND DEFENSE MECHANISM OF CORAL REEF FISHES TO CIGUATOXINS (CTXS)

Leo Lai CHAN, Yim Ling MAK

Ciguatoxins (CTXs) are a group of natural marine biotoxins that are commonly found in marketed coral reef fishes. They are of great concerns as consumption of CTX-contaminated coral reef fishes can induce ciguatera fish poisoning (CFP) in human. Because of global warming and an increase in nutrient input in the marine environment, there is a growing likelihood of CTX-causative dinoflagellates, and hence CTXs spreading into new regions of the globe. More importantly, CTXs may negatively affect wildlife since a laboratory study has demonstrated that CTX-exposed fish embryos exhibited cardiovascular, muscular and skeletal abnormalities. A reduction of larval survivability in fishes was also observed when greater CTX levels were exposed to fish embryos. Additionally, mortality of piscivorous marine mammals such as Hawaiian monk seals (*Monachus schauinslandi*) was reported in a CFP-prone area in Hawaii. CTXs may therefore represent an unrecognized threat to marine life and eventually pose an ecological risk to coral reef system in the CFP-prone regions. In the present study, we aim to define a list of CTX-sensitive coral reef fishes that may become less common in an ecosystem of the CFP-prone region (due to natural selection) based on comprehensive laboratory studies. Besides, pharmacokinetics (i.e. accumulation, subsequent change in tissue distribution and elimination of CTXs) of CTXs in CTX-resistant and sensitive coral reef fishes will be examined in order to provide insight on species-specific variations in physiological mechanisms that render CTX-resistant coral reef fishes ability to mediate harms after CTX exposure. The study of genomic and the complementary DNA sequence of the excitable tissue such as nerve, skeletal muscle, brain and heart of CTX-resistant and sensitive coral reef fish species will also be carried out to understand the molecular basis of the defense mechanism of fishes against CTXs. All the results will provide systematic scientific information for clarifying the role of CTXs to act as natural selection agents which can result in long-term changes of coral reef community and ecosystem.

SIGNIFICANCE OF ROOTS AND MICROORGANISMS IN PHYTOREMEDIATION OF POLYBROMINATED DIPHENYL ETHERS (PBDEs) BY MANGROVE PLANTS (AREA D: POLLUTION CONTROL AND BIOREMEDIATION)

Nora Fung Yee TAM, Hai Chao ZHOU

Toxic, recalcitrant and newly emerged organic pollutants such as polybrominated diphenyl ethers (PBDEs) are common contaminants in coastal sediments, and their carcinogenic properties have led to increasing attention on their remediation. Over the last few decades, phytoremediation has been suggested as an innovative, non-intrusive and inexpensive technology that utilises plant systems and associated microorganisms to remove toxic contaminants from the environment. Previous work on phytoremediation, however, focused mainly on the uptake and accumulation of pollutants (mostly heavy metals) within tissues of terrestrial plants. Little is known about the phytoremediation of toxic organic pollutants by wetland plants in coastal environments. Unlike metal removal, the most significant phytoremediation process for organic pollutants such as PBDEs is rhizosphere degradation, the breakdown of pollutants by microorganisms in the root zone environment (the rhizosphere). Because coastal wetland plants are subject to tidal flushing with alternating aerobic and anaerobic environments which support both aerobic and reductive transformation/degradation of PBDEs. Wetland plants are also known to transfer oxygen from the above-ground tissue to the roots for internal respiration. Excess oxygen is released to the surroundings (rhizosphere) to provide aerobic pockets for microorganisms to degrade pollutants and the formation of iron plaque, which could immobilize pollutants on root surface and uptake. However, the roles of roots and the rhizosphere effects of wetland plants, such as mangroves, have never been explored. The proposed study therefore aims to investigate the significance of roots, including the effects of roots in releasing oxygen to create an aerobic environment in the rhizosphere, the formation of iron plaque on root surface, the roles of root exudates on the degradation, mobility, bioavailability of PBDEs, the selection of PBDE-degraders and the changes of the microbial community structure in rhizosphere and bulk sediments during phytoremediation by mangrove plants. The detailed functions of roots in different mangrove plant species on the remediation of coastal sediments contaminated with PBDEs will be compared. The results from the proposed work will provide a thorough scientific understanding on the significance and mechanisms of mangrove roots, the rhizosphere and the associated microorganisms in the cleanup of coastal sediments contaminated with PBDEs.

THE REGENERATIVE TOXICOLOGY OF ALPHA EMITTERS IN THE CAUDAL FIN OF MARINE MEDAKA: LINKING RADIATION INDUCED EFFECTS FROM MOLECULAR TO PHYSIOLOGICAL LEVELS

Shuk Han CHENG, Jinping CHENG

The Tsunami-driven accidental meltdown of the Fukushima Dai-ichi nuclear power plant (NPP) has illustrated the wide-ranging effects on the marine ecosystems associated with airborne fallout over the ocean and oceanic discharge of radionuclides found in contaminated water. Hong Kong receives part of its electricity supply from the Daya Bay nuclear plant, one of the first commercial NPP operated by the China Guangdong Nuclear Power Group. It is known that this Nuclear Power Group is planning to build over 20 more NPP in Guangdong, posing an ever increasing need for research on the radioecology in marine pollution. Among the radionuclides, the Americium-241 and Plutonium-240 are alpha emitters commonly found in nuclear reactors and in nuclear explosions. Their much longer decay half lives (at 432 years and 6,563 years respectively), compared to the gamma emitter Cesium-137 (at 30 years) means that these alpha emitters persist much longer in the environment. The alpha particles have much larger relative biologic effectiveness, at about 20 times when compared to beta particles and gamma ray photons. Therefore, special efforts will be required to monitor their existence and to determine the ecological consequences under realistic exposure conditions. Alpha particles are highly radiotoxic when ingested or come into direct contacts, generating reactive oxygen species and double strand DNA breaks. They are also most difficult to monitor by equipment due to its short range and the complex processes involved. With this seed funding, we will gather the biological data to measure a biological relevant endpoint under conditions of alpha particle exposures and co-stressors. This exploratory project will enable us to design a transgenic marine medaka to be used to perform biomonitoring of alpha particles in seawater and its potential effects with mixtures of stressors such as metals and endocrine disruptors.

Abstracts of the Internal Research Fund (IRF) Projects

IRF項目摘要

Jan 2013 - Dec 2014 (Newly Funded)

IRF

EXPRESSION OF PLASMA IMMUNE PROTEINS IS GENDER-DEPENDENT AND RELATED TO FISH SUSCEPTIBILITY TO PATHOGEN

Doris Wai Ting AU, Yun Wah LAM

In teleost fish, the innate immune system serves as the first line of host defense to pathogens before the adaptive immune system is ready and potent enough to take over. Plasma complement proteins are known to provide a bridge between innate and adaptive immunity in vertebrates. Liver is the key organ for complement proteins synthesis. Our recent studies have clearly demonstrated that endogenous expression of six major complement system genes in liver of medaka *Oryzias melastigma* was gender-dependent (males > females). Moreover, the immunomodulatory effects of environmental contaminant BDE-47 on complement genes expression in liver of *O. melastigma* varied between male (downregulation) and female (upregulation). The results suggest that gender-dependent immune response is an important consideration when assessing the risk of immunosuppressive chemicals in waters. It is however unclear whether the expression of complement system genes in liver is coupled with their proteins expression in plasma. The recent advent of proteomics technology allows proteins profiling and absolute quantification of single protein in fish plasma, which is useful to verify whether or in what ways gender-dependent expression of plasma immune proteins may occur in medaka. To understand the involvement of estrogen (E2) on the expression of plasma immune proteins in medaka, fish at 4-months to 15 months old with different levels of plasma E2 (F > M; peak at 8-month old females) will be used to study the correlation between the level of plasma E2 and expression of plasma immune proteins in medaka. E2 exposure experiment will be conducted to investigate the modulatory role of E2 in expression of plasma immune/complement proteins. To understand the predictability of plasma immune proteins expression on fish susceptibility to microbial challenge, pathogen challenging experiments will be conducted using fish expressing different levels of plasma immune proteins. The levels of plasma immune proteins (in particular complement system proteins) in medaka (resting and/or pathogen-challenged) will be correlated with fish susceptibility to infectious pathogen. If the levels of targeted plasma immune proteins exhibit a good relationship with fish susceptibility to pathogen, these plasma immune proteins will be promising molecular biomarkers for early detection, monitoring and assessing the risk of immunosuppressive chemicals in aquatic environments.

IN-SITU MEASUREMENT OF VARIATIONS IN SOLAR ULTRAVIOLET RADIATION PENETRATION THROUGH MARINE WATER

Peter Kwan Ngok YU

In this project, we aim to utilise our newly planned ultraviolet (UV) radiation measurement technique for evaluation of UV radiation penetration through water of varying qualities and conditions. Radiochromic films, first developed for the measurement of ionizing radiation have been discovered by us to also measure non-ionizing broad spectrum UV radiation. We have performed initial testing on these films in water environments and found potential for use as a UV dosimeter due to their characteristics such as ability to measure UV over an extended period of time, their relative water resistance and a level of accuracy and reproducibility required for such measurements. We plan to use these films for evaluation of UV penetration into water of differing qualities in-situ over a daily period. To perform this task, we will create a dosimetry string line, which can be lowered into the aquatic environment to measure accumulated UV radiation over a period of time during the day. Detectors will be positioned on the string line at defined depths and lowered into the aquatic area for analysis. Our film detectors change color based on the level of UV exposure they have received and relative reading can be performed by comparing optical density color changes of the films at a later stage. Our initial aims will be to firstly produce baseline measurements for clean water (swimming pool results) followed by evaluation of UV penetration differences for marine and aquatic environments where water quality is compromised to differing levels by dissolved organic matter and pollution. Of significance with these techniques is the ability of our system to perform measurements of solar UV radiation for a range of depths over a given time period without the need for many expensive solid-state UV detectors. The use of our film style detectors (which are relatively inexpensive) allows for multiple point measurements to be made at the same time, which is not practically accomplishable using solid-state UV meters. We believe that this work has potential applications for use in all marine environments for analysis of UV radiation levels and its variation caused by water pollution and quality.

PHYTOREMEDIATION OF POLYBROMINATED DIPHENYL ETHERS (PBDES) BY MANGROVE WETLANDS

Nora Fung Yee TAM, Hantao ZHOU

Polybrominated Diphenyl ethers (PBDEs), the widely used flame-retardant additives, are ubiquitous, persistent and toxic contaminants. Due to their hydrophobicity, PBDEs released to environments are accumulated in sediments, particularly in estuaries, the special coastal zones strongly influenced by anthropogenic activities. Extremely high concentrations of PBDEs have been recorded in estuarine sediments in South China. Although penta- and octa-BDEs have been banned since 2000s due to their toxicities, deca-BDEs are still produced and applied around the world. The persistence, toxicity and carcinogenic properties of PBDEs have led to increasing attention on their remediation. Over the last few decades, phytoremediation has been suggested as an innovative, non-intrusive and inexpensive technology that utilises plant systems to remove toxic contaminants from the environment. Previous work on phytoremediation has focused on the cleanup of metal contaminated upland soil using terrestrial plants, concentrating on phytoextraction (uptake and accumulation of pollutants within plant tissues). Little is known about the phytoremediation of toxic organic pollutants by wetland plants in coastal environments. Our recent research demonstrates that mangrove plants had some tolerance to polycyclic aromatic hydrocarbons (PAHs) and the wetland system was capable of removing and degrading PAHs in contaminated sediments. However, the performance of mangrove wetlands in the cleanup of PBDE-contaminated sediments and their tolerance to PBDE stresses has never been reported. Without a clear understanding of how mangrove plants tolerate PBDE toxicity, their ability to remove and degrade PBDEs, it is difficult to promote and apply phytoremediation, an emerging technology. The proposed study therefore aims to explore the feasibility of using mangrove wetland systems to cleanup sediments contaminated with PBDEs and understand the fate of PBDEs in contaminated sediments. The study also compares the performance of different mangrove plant species, aiming to identify the most tolerant and effective species for phytoremediation purposes. The results from the proposed work will not only fill the knowledge gap on the fate of PBDEs in estuarine sediments, it will also provide very useful scientific information for developing phytoremediation strategies to clean-up contaminated sediments.

RESPONSES OF MARINE ORGANISMS TO OCEAN ACIDIFICATION: DOES ECOSYSTEM MATTER?

CHEUNG, Siu Gin, Kam Shing Paul SHIN

A large proportion of the carbon dioxide (CO₂) released into Earth's atmosphere by human activities is absorbed by the oceans. When dissolved in water, CO₂ forms carbonic acid, increasing the concentration of H⁺ (hydrogen ion) and lowering the pH in ocean waters. Under the Intergovernmental Panel on Climate Change (IPCC) future CO₂ emission scenarios, a drop in ocean pH of up to 0.5 units by 2100 has been predicted with "with a high level of confidence". There is a growing concern of the effect of ocean acidification on marine organisms with relevant publications increasing gradually in recent years. Recent data suggest that ocean acidification impacts the performance of marine organisms via changes in calcification rates (a drop in pH will cause a corresponding decrease in carbonate saturation level, adversely affecting organisms with calcified shells) and other physiological responses. Acidification, however, does not occur in isolation but in concert with other stresses, such as warming and reduced salinity. Few studies, however, have examined the interactive effect of acidification and other direct or indirect results of global change, which may aggravate the effect of ocean acidification on marine organisms.

Intertidal species are subjected to greater fluctuations in temperature, salinity, pH, etc. Therefore, they are expected to develop greater tolerances to environmental changes, for example, ocean acidification. As far as we know, no study has been carried out to test this hypothesis. Most of the previous studies on ocean acidification only focused on a single species or compared closely related species in the same habitat. If the hypothesis is correct, this helps us predict relative tolerances of closely-related species based on the environment they inhabit. Besides, it implies that intertidal communities may be more resistant to ocean acidification than subtidal communities. We propose to test the above hypothesis by comparing two species of *nassariid* gastropods in the same genus. One of them is intertidal species whereas the other is subtidal. The combined effects of ocean acidification and low salinity on the physiological responses, as well as growth and development of critical life stages of key species will be investigated. The substantial improvements made by the proposed research efforts to the existing predictive framework will be central to the management and conservation of living marine ecosystems in the face of climate change.

RISK ASSESSMENT OF PHARMACEUTICAL RESIDUES AND OCCURRENCE OF ANTIBIOTIC-RESISTANT BACTERIAL GENES AND STRAINS IN HONG KONG SURFACE WATERS AND SEDIMENTS

Margaret B. MURPHY, Maureen V. BOOST

The occurrence of pharmaceuticals in the natural environment due to extensive human use and release has been documented worldwide in recent years. The widespread occurrence of these biologically active compounds in both abiotic and biotic samples has raised concern about their potential toxic effects with regard to both ecological and human health. In particular, the ubiquitous environmental occurrence of human and veterinary antibiotics, coupled with clinical data showing a global rise in drug-resistant bacterial strains, has prompted research into the presence of antibiotic-resistance genes (ARGs) and resistant bacteria in natural systems. Our previous work has shown that antibiotic loadings from sewage treatment plants into the Hong Kong environment are among the highest in the world for some antibiotics; likewise, clinical studies have shown that the rate of antibiotic resistance among some bacterial strains is very high in Hong Kong. This study will therefore investigate the presence of antibiotic resistance in the Hong Kong environment in order to determine the potential impacts of pharmaceutical release. Year-long sampling of surface seawater and marine sediment will be carried out to examine seasonal variation in environmental pharmaceutical concentrations. Seawater samples will be extracted and analyzed based on our previously published method with modifications, while a new method will be developed for pharmaceutical quantification in marine sediments. Seawater and sediment samples will also be analyzed for the presence of ARGs and for resistant bacterial strains using genotypic and phenotypic methods based on sample filtration, bacterial culture on MacConkey agar and sensitivity tests for Gram-negative, Gram-positive and anaerobic strains, as well as polymerase chain reaction for the amplification of ARGs. Concentrations in seawater and marine sediments will be compared with the occurrence data for the ARGs and resistant bacterial strains to determine correlations between antibiotic levels and the presence of resistance in the environment. The results of these analyses will be used to carry out a risk assessment of pharmaceuticals in the Hong Kong environment using a probabilistic approach and the use of species sensitivity distributions. This approach can then be applied to future research in China and in the Asia-Pacific region.

THE REGENERATIVE TOXICOLOGY OF ALPHA EMITTERS IN THE CAUDAL FIN OF MARINE MEDAKA: LINKING RADIATION INDUCED EFFECTS FROM MOLECULAR TO PHYSIOLOGICAL LEVELS

Shuk Han CHENG, Peter Kwan Ngok YU

The Tsunami-driven accidental meltdown of the Fukushima Dai-ichi nuclear power plant (NPP) has illustrated the wide-ranging effects on the marine ecosystems associated with airborne fallout over the ocean and oceanic discharge of radionuclides found in contaminated water. Hong Kong receives part of its electricity supply from the Daya Bay nuclear plant, one of the first commercial NPP operated by the China Guangdong Nuclear Power Group. It is known that this Nuclear Power Group is planning to build over 20 more NPP in Guangdong, posing an ever increasing need for research on the radioecology in marine pollution. Among the radionuclides, the Americium-241 and Plutonium-240 are alpha emitters commonly found in nuclear reactors and in nuclear explosions. Their much longer decay half lives (at 432 years and 6,563 years respectively), compared to the gamma emitter Cesium-137 (at 30 years) means that these alpha emitters persist much longer in the environment. The alpha particles have much larger relative biologic effectiveness, at about 20 times when compared to beta particles and gamma ray photons. Therefore, special efforts will be required to monitor their existence and to determine the ecological consequences under realistic exposure conditions. Alpha particles are highly radiotoxic when ingested or come into direct contacts, generating reactive oxygen species and double strand DNA breaks. They are also most difficult to monitor by equipment due to its short range and the complex processes involved. With this seed funding, we will gather the biological data to measure a biological relevant endpoint under conditions of alpha particle exposures and co-stressors. This exploratory project will enable us to design a transgenic marine medaka to be used to perform biomonitoring of alpha particles in seawater and its potential effects with mixtures of stressors such as metals and endocrine disruptors.

Summary of the Internal Research Seed Fund (IRSF) Projects

IRSF項目概要

SKLMP Funded Project Progress Report

Jan 2011 - Dec 2012 (Ongoing)

DEVELOPMENT OF NOVEL TECHNOLOGY FOR EARLY DIAGNOSIS AND MONITORING OF IMMUNOTOXIC POLLUTANTS IN MARINE ENVIRONMENT**Doris Wai Ting AU**

Impairment of immune system reduces the ability of an organism to defend itself against pathogens, resulting an adverse impact on fitness and survival success of individuals. Endocrine disruptive chemicals (EDCs) are present in large quantities and widespread in the marine ecosystems. The mammalian immune system has been found to be sensitive to environmental estrogens. Immune competence of vertebrates can be compromised more readily by EDCs as compared to their long term consequence on reproductive success. However, to date, there is a lack of monitoring technology for early diagnosis of immunotoxic effects in marine organisms. There is an urgent need to develop novel and sensitive tools for early detection and monitoring of immunotoxicants and their effects in marine vertebrates. A longstanding barrier to progress in biomarker development for ecotoxicology has been the inability of most sub-organismal responses (including molecular and biochemical changes) to indicate significant effects at the population level. Current strategy is to employ a two-step approach to tackle this challenge: i) formulate a conceptual framework to predict —adverse outcome pathway|| (AOP) which represents a sequence of events that begins with a molecular initiating event, spans multiple levels of biological organization, and ends with an adverse outcome at the level of an individual; ii) develop biologically based, quantitative extrapolation models that allow us to apply cell- or tissue-level data to individuals, and individual-level data to entire populations. In the present study, attempt will be made to develop a conceptual adverse outcome pathway (AOP) and biologically-based, quantitative dose response models for predicting environmental estrogens induced immunotoxic effects, using the marine medaka as model organism. Experiments will be conducted to determine dose-response, sensitive life stage and lifelong impacts of estrogen agonist, 17 α -ethinylestradiol (EE2), on immune competence in marine medaka. A battery of molecular, cellular and histological endpoints will be used to assess change in immune function and competence in marine medaka upon short-term and long-term exposure to EE2. The

ultimate goal is to identify novel immune biomarkers that gain both predictive and diagnostic credibility through their links to both the initiating event and the adverse outcome, which are useful for early diagnosis and monitoring of immunotoxicants in marine environment.

A. Project Objectives

1. To establish potential toxicity pathways for predicting immunotoxic effects of environmental contaminants, using the marine medaka as model organism;
2. To investigate dose-response, sensitive life stages and lifelong impact of short-term and long-term exposure to environmental estrogens on immune competence of marine medaka;
3. To develop biologically based dose-response models for predicting immunotoxic effects in fish population;
4. To identify novel and sensitive biomarkers for early diagnosis and monitoring of immunotoxic chemicals in the marine environment;

B. Research Activities

An increasing number of field and laboratory studies have shown that many environmental contaminants can modulate immune competence and increase the susceptibility of fish to pathogens. Fish immunotoxicology is an important field linking sub-organismal alterations to population effects, because of its high relevance to the health and fitness of individual. In teleost fish, the innate immune system serves as the first line of host defense to pathogens before the adaptive immune system is ready and potent enough to take over.

- A suppression subtractive hybridization (SSH) cDNA library was constructed for the marine medaka *O. melastigma* to identify differentially expressed immune genes in the liver of fish upon infection with pathogenic *Vibrio parahaemolyticus*. Among all the 396 genes identified, 38 (9.6%) were immune-related genes, in which 17 genes were involved in the complement system. The complement system is known to provide a bridge between innate and adaptive immunity in animals. Fish contain a highly developed complement system which plays a key role in phagocytosis, respiratory burst, chemotaxis, cell lysis and in host defense against pathogens. In vertebrates, the complex complement system can be activated through one or a combination of four complement activation pathways: viz. the classical, alternative, lectin and proteases (thrombin) pathways, and Complement C3 is the central protein of all four known activation pathways. Fifteen components of complement system sequences were obtained from our *O. melastigma* SSH library. All key complement system genes were found in marine medaka.

- Pathway analysis conducted with DAVID showed that the complement and coagulation cascades were more important than other analyzed pathways, such as, the ribosome pathway, galactose metabolism and systemic lupus erythematosus pathway. A clear temporal response is evident when comparing the pattern of transcription of genes involved in the four complement activation pathways, suggesting a tightly regulated complement system response in medaka after bacterial challenge.
- Hepcidin, also termed LEAP-1 (liver expressed antimicrobial peptide), is known to be an important antimicrobial peptide (AMP) in the innate immune system of mammals. Two hepcidin genes (OM-hep1 and OM-hep2) were identified, cloned and characterized in the *O. melastigma*, which were found to be highly conserved with other reported hepcidins. During embryogenesis, significant elevation of OM-hep1 and OM-hep2 transcripts were coincided with liver development in the embryos. In adult medaka, differential tissue expressions of both hepcidin transcripts were evident: high in liver, moderate in spleen and low in non-immune tissues. After exposure to pathogenic *V. parahaemolyticus*, the two hepcidin mRNAs were rapidly and remarkably induced in liver and spleen. The results suggest that the multiple forms of hepcidins are effective components and play a complementary role in response to invading microorganism during early developmental stages and adulthood.

C. Peer-reviewed Journal Publication Arising from this Research Project

IDENTIFICATION OF DIFFERENTIALLY EXPRESSED GENES AND QUANTITATIVE EXPRESSION OF COMPLEMENT GENES IN THE LIVER OF MARINE MEDAKA *ORYZIAS MELASTIGMA* CHALLENGED WITH *VIBRIO PARAHAEMOLYTICUS*

J. Bo, J. P. Giesy, R. Ye, K.J. Wang, J.S. Lee, D.W.T. Au

The innate immune system of fish is the primary defense against acute diseases. The marine medaka *Oryzias melastigma* has been shown to be a potential marine fish model for ecotoxicology, but little is known about the innate immune system of this small fish. In this study, suppression subtractive hybridization (SSH) was used to identify differentially expressed immune genes in the liver of *O. melastigma* infected with *Vibrio parahaemolyticus*. Among the 396 genes identified, based on NCBI BLAST search of the 1279 sequenced clones in the SSH libraries, 38 (9.6%) were involved in the immune process. Besides, genes involved in biological regulations (5.6%); cellular metabolism (24.7%); general response to stimuli (4.8%); cellular component organization (2.3%); signal transduction (2.5%) and transport process (2.8%) were

also obtained. Ten complement component genes involved in four activation pathways were quantified (using q-PCR) and exhibited different patterns of transcription between the control and challenged individuals. The results reported upon here support the feasibility of developing *O. melastigma* as a marine model fish to understand the basic biological processes related to immune function and for immunotoxicological research. Findings of this study established a genetic platform for studying immune function using *O. melastigma*.

Comp Biochem and Phys –Part D: Genomics and Proteomics, 2012, 7(2), 191-200.

D. Recognized international conference in which paper related to this research project was delivered

GENDER-SPECIFIC MODULATION OF INNATE IMMUNE RESPONSES IN FISH UNDER ENVIRONMENTAL STRESSES

R. Ye, J. Bo, K.M. Cheung, D.W.T. Au

An increasing number of field and laboratory studies have shown that many environmental contaminants can modulate immune competence and increase the susceptibility of fish to pathogens. Fish immunotoxicology is an important field linking sub-organismal alterations to population effects, because of its high relevance to the health and fitness of individual fish. Fish contain a highly developed innate immune complement system which plays a key role in phagocytosis, respiratory burst, chemotaxis, cell lysis and in host defense against pathogens. The Japanese medaka (*Oryzias latipes*) and the seawater counterpart *Oryzias melastigma* have been proven useful for studying innate immunity and molecular toxicology. Our recent studies have shown that immunomodulatory effects of ubiquitously occurred environmental contaminants (e.g. PBDE, estrogen-active chemicals) on complement gene transcription were very different between male and female medaka. The underlying molecular mechanisms should be unraveled and identified for both male and female fish. Gender dependent response is an important consideration when assessing the risk of immunosuppressive chemicals. Future direction for fish immunotoxicology must include parallel assessment for both genders. Development of biomarkers for fish immunotoxicology has been hindered by our inability to predict significant effects at individual/population levels from molecular and cellular responses. Future immunotoxicological studies should gear toward the development of sensitive and specific molecular and cellular biomarkers for early diagnosis of immunotoxic chemicals in aquatic environments. A battery of cost effective immune endpoints using small model fish is needed for assessing potential immunomodulatory effects of environmental contaminants.

QUANTIFICATION OF POLY- AND PERFLUORINATED COMPOUNDS (PFCs) AND SPECIES IDENTIFICATION OF SHARK FINS PURCHASED FROM HONG KONG SEAFOOD SHOPS

Margaret B. MURPHY

Shark populations are declining around the world, largely due to overfishing and the global shark fin trade. In contrast to many other marine predators, there is a lack of information on the contaminant status of sharks, particularly with regard to poly- and perfluorinated compounds (PFCs), which are used as surface protectors and surfactants both industrially and in manufactured products. Some PFCs have been shown to be persistent and toxic, and thus these chemicals are of global concern. Unlike many organic contaminants, PFCs tend to partition to protein-rich tissues. As such, they can be detected in bird feathers, which are composed primarily of keratin; in this project, a method will be developed based on the method for feathers to measure PFCs in shark fins, which are composed of the structural protein collagen, using acid digestion, solid-phase extraction and high performance liquid-chromatography tandem mass spectrometry (HPLC-MS-MS). Shark fins are readily available in Hong Kong, which is the largest market in the global fin trade; shark fin soup is also commonly consumed locally. In this project, shark fins will be purchased from seafood shops around the city in collaboration with the marine conservation group BLOOM and analyzed for concentrations of short and long-chain PFCs, and a hazard assessment will be carried out to determine the hazard of shark fin consumption to humans due to PFCs. Individual fins will also be identified at the species level using DNA extraction and sequencing in order to get more information on the species involved in the global shark fin trade, and to relate PFC exposure and accumulation to trophic level and species. By combining ecotoxicology and conservation biology, this project will provide data with relevance to both environmental and human health.

A. Project Objectives

1. To develop a method to quantify short- and long-chain PFCs in shark fins purchased from Hong Kong seafood shops;
2. To carry out a hazard assessment of PFC exposure in humans due to shark fin consumption;
3. To identify the species of the purchased fins in order to:
 - a. Determine the trophic levels represented by the sampled species and relate them to PFC levels and accumulation in the fins;
 - b. Further understand the conservation implications of the shark fin trade in Hong Kong.

B. Research Activities

Eight PFCs were quantified in fins from seven shark species using a newly developed method specific for shark fins in which the fins were acid-digested for 48 hours, then subjected to solid phase extraction followed by Envi-Carb cleanup and quantified using liquid chromatography-tandem mass spectrometry (LC-MS/MS). The target analytes were perfluorooctane sulfonate (PFOS), perfluorooctanoate (PFOA), perfluorohexanoate (PFHxA), perfluorohexane sulfonate (PFHxS), perfluorononanoate (PFNA), perfluorodecanoate (PFDA), perfluoroundecanoate (PFUnDA) and perfluorododecanoate (PFDoDA). These PFCs were measured in individual fins identified as tiger shark (*Galeocerdo cuvier*), scalloped hammerhead shark (*Sphyrna lewini*), great hammerhead shark (*S. mokarran*), oceanic whitetip shark (*Carcharhinus longimanus*), shortfin mako shark (*Isurus oxyrinchus*) and blue shark (*Prionace glauca*). Of the target compounds, only PFOS and PFNA were detectable (limits of quantification [LOQs] of PFOS, PFOA, PFHxA and PFHxS were 20.8-23.3 pg/g dry weight (dw); LOQs of PFNA, PFDA, PFUnDA and PFDoDA were 208-233 pg/g dw). PFOS levels ranged from <LOQ to 0.80 ng/g dw, while those of PFNA ranged from <LOQ to 12 ng/g dw.

Although PFOS is commonly found to be the most prevalent PFC in biological samples, PFNA levels in the fins were found to more than approximately 10- to 100-fold higher than those of PFOS, which is contrary to the findings in other protein-rich matrices such as bird feathers. These results may therefore indicate differential uptake and accumulation of PFNA in shark fins, although the potential mechanism for this accumulation is currently unknown. A review of the literature on PFCs in Hong Kong seafood showed that shark fins contained higher PFNA levels than other seafood such as fishes (range: 90-300 pg/g wet weight) and shrimp (not detected). As there is currently no reference dose for PFNA, a preliminary risk assessment of the measured PFNA levels was conducted using the proposed reference dose (RfD) for PFOS of 0.025 mg/kg body weight/day. PFOS is expected to be more toxic than PFNA, and therefore this is a very conservative assessment. A consumption calculation for the potential risk of PFNA exposure to the Hong Kong public was carried out using shark fin consumption data collected by project collaborators at BLOOM Association. The results of the assessment indicated that the risk posed to Hong Kong consumers was very low, even under worst-case (i.e., frequent consumption) conditions.

This study analyzed a relatively small number of samples, as the method development required more time than expected. It was predicted that a method for shark fins could be developed based on a previously published method for bird feathers, but the feather method was found to be unsuitable, and an entirely new method that included a clean-up step was required. The new method is suitable for a range of shark species, though ion suppression was observed for some of the target PFCs. Collaboration in this project occurred between SKLMP members, colleagues at BLOOM Association, and a colleague at the University of Toronto (Dr. Leo W.Y. Yeung), who carried out the LC-MS/MS sample quantification.

C. Recognized international conference in which paper related to this research project was delivered

MEASUREMENT OF TRACE ELEMENT AND PERFLUORINATED COMPOUND (PFC) CONCENTRATIONS AND SPECIES IDENTIFICATION OF SHARK FINS FROM THE HONG KONG MARKET

M.B. Murphy, D.L.H. Chu, E.I.H. Loi, P. Lam, Y.P. Mak

Shark populations are declining around the world, largely due to overfishing and the global shark fin trade. In contrast to many other marine predators, such as marine mammals, there is a lack of information on the contaminant status of sharks, likely because sharks are often more difficult to obtain. The larger pelagic species, in particular, are often available only for opportunistic sampling as bycatch, but are well represented in the shark fin market. The purpose of this study was to assess the levels of two groups of contaminants, trace elements and perfluorinated compounds (PFCs), in shark fins purchased from dried seafood shops in Hong Kong, one of the largest markets in the world for shark fins, and to identify the sampled species using DNA sequencing. Shark fins, composed primarily of collagen, can be used for monitoring protein-bound contaminants, and also can be used to assess the hazards of human exposure to these chemicals through the consumption of shark fin soup, a traditional Chinese dish. Shark fins (n=30) from several trade categories were purchased and analyzed for trace element (Ag, As, Cd, Co, Cr, Cu, Mn, Mo, Ni, Pb, Sb, Se, Ti, and Zn) levels using microwave digestion and inductively coupled plasma-mass spectrometry (ICP-MS). A new method was developed for the extraction of short- and long-chain PFCs from shark fins and PFC concentrations were quantified by liquid chromatography-tandem mass spectrometry (LC-MS/MS). DNA was extracted from the purchased fins using a commercial kit and species were identified using validated primer sequences. Trace element concentrations varied among individuals and species, and hazard assessment of the measured levels indicated that the overall risk to human consumers was low. The results of the species identification confirmed the presence of shark species listed as threatened or endangered by the International Union for the Conservation of Nature on the Hong Kong market, and will be discussed in the context of changes in shark populations over the past 10 years, as well as the occurrence and bioaccumulation of trace elements and PFCs in sharks.

D. Other Impact

The PI, along with colleagues from BLOOM Association, was interviewed and filmed in the SKLMP by RTHK and CCTV about this project.

SKLMP Funded Project Final Report

Jan 2010 - Dec 2011 (Completed)

DEVELOPMENT OF HIGHLY EFFICIENT ZnO TETRAPODS NANOPARTICLES FOR PHOTODEGRADATION OF ORGANIC POLLUTANTS IN WATER UNDER VISIBLE LIGHT

Tai Chu LAU

A. Project Objectives

To prepare ZnO tetrapods doped with various metals and non-metals for the photodegradation of various organic pollutants in water using visible light.

B. Research Activities

Various semiconductor nanoparticles have been prepared and their use as photocatalysts for degradation of organic pollutants in water and for generation of renewable hydrogen sources have been investigated.

C. Peer-reviewed Journal Publications Arising from this Research Project

A COBALT(II) QUATERPYRIDINE COMPLEX AS A VISIBLE LIGHT-DRIVEN CATALYST FOR BOTH WATER OXIDATION AND REDUCTION

C.F. Leung, S. M. Ng, C. C. Ko, W.L. Man, J. Wu, L. Chen and T.C. Lau

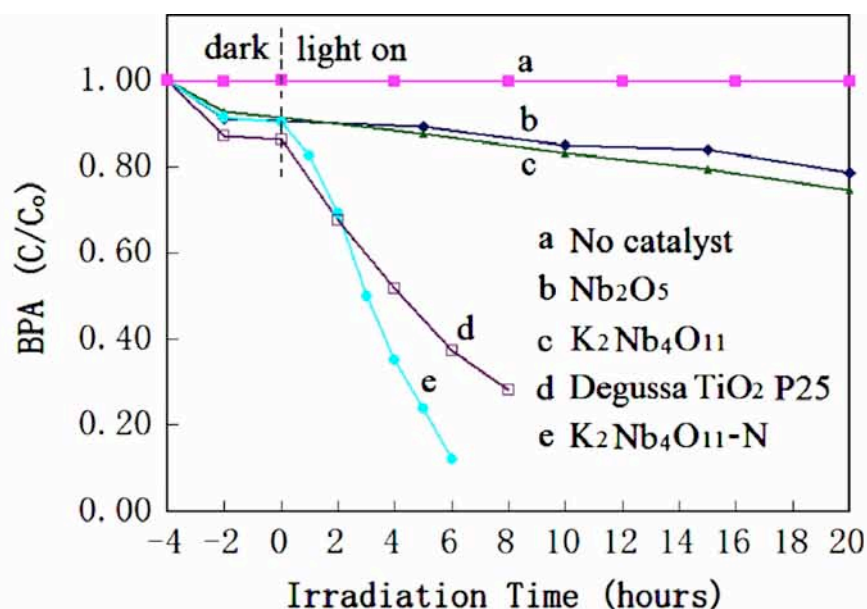
The complex $[\text{Co}^{\text{II}}(\text{qpy})(\text{OH}_2)_2]^{2+}$ (qpy = 2,2':6'2'':6''2'''-quaterpyridine) is an efficient visible light-driven catalyst for both water oxidation and reduction. It catalyses photochemical oxygen evolution from water at pH 8.0 with $[\text{Ru}^{\text{II}}(\text{bpy})_3]^{2+}/\text{S}_2\text{O}_8^{2-}$ ($\lambda = 457 \text{ nm}$, max TON = 335, bpy = 2,2'-bipyridine). It also catalyses photochemical hydrogen generation from $[\text{Ir}^{\text{III}}(\text{dF}(\text{CF}_3)\text{ppy})_2(\text{dtbbpy})]^{+}/\text{TEOA}$ (dF(CF₃)ppy = anion of 2-(2,4-difluorophenyl)-5-trifluoromethylpyridine, dtbbpy = 4,4'-di-*tert*-butyl-2,2'-bipyridine, TEOA = triethanolamine) in aqueous acetonitrile ($\lambda > 420 \text{ nm}$, max TON = 1730)

Energy Environ. Sci., 2012, 5, 7903

PREPARATION OF NITROGEN DOPED $K_2Nb_4O_{11}$ WITH HIGH PHOTOCATALYTIC ACTIVITY FOR DEGRADATION OF ORGANIC POLLUTANTS

Y. Qiu, L. Wang, C. Leung, G. Liu, S. Yang, and T.C. Lau

Nitrogen doped $K_2Nb_4O_{11}$ ($K_2Nb_4O_{11}$ -N) has been prepared by solid state reaction between $K_2Nb_4O_{11}$ and urea at 400 °C. $K_2Nb_4O_{11}$ -N has been characterized by XRD, SEM, XPS and UV/vis diffuse reflectance. The photodegradation of various organic pollutants in water by this material, including Orange G (OG), bisphenol A (BPA) and pentachlorophenol (PCP) have been studied at $\lambda > 330$ nm and > 399 nm. The results show that the photocatalytic activity of $K_2Nb_4O_{11}$ -N at > 399 nm is higher than those of $K_2Nb_4O_{11}$ and Degussa TiO_2 P25, indicating the activating effect of nitrogen doping. A mechanism for the photodegradation of organic substrates by $K_2Nb_4O_{11}$ -N is proposed.



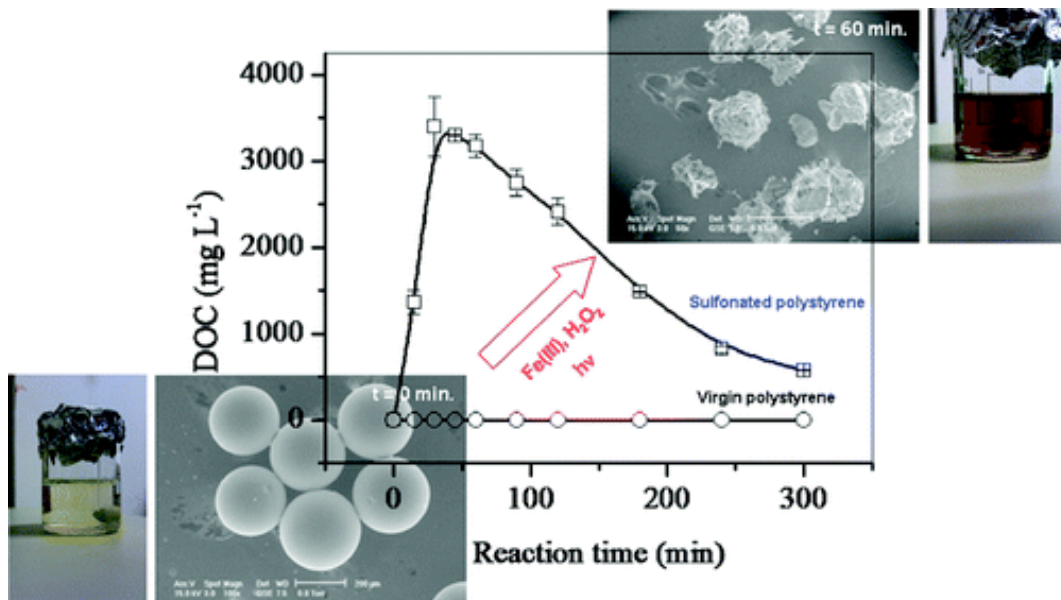
Applied Catalysis A: General 402 (2011) 23–30

PHOTOASSISTED FENTON DEGRADATION OF POLYSTYRENE

H.M. Feng, J.C. Zheng., N.Y. Lei, L. Yu, K.H.K. Kong, H.Q. Yu, T.C. Lau and M.H.W. Lam

Fenton and photoassisted Fenton degradation of ordinary hydrophobic cross-linked polystyrene microspheres and sulfonated polystyrene beads (DOWEX 50WX8) have been attempted. While the Fenton process was not able to degrade these polystyrene materials, photoassisted Fenton reaction (mediated by broad-band UV irradiation from a 250 W Hg(Xe) light source) was found to

be efficient in mineralizing cross-linked sulfonated polystyrene materials. The optimal loadings of the Fe(III) catalyst and the H₂O₂ oxidant for such a photoassisted Fenton degradation were found to be 42 μmol-Fe(III) and 14.1 mmol-H₂O₂ per gram of the sulfonated polystyrene material. The initial pH for the degradation was set at pH 2.0. This photoassisted Fenton degradation process was also able to mineralize commonly encountered polystyrene wastes. After a simple sulfonation pretreatment, a mineralization efficiency of >99% (by net polymer weight) was achieved within 250 min. The mechanism of this advanced oxidative degradation process was investigated. Sulfonate groups introduced to the surface of the treated polystyrene polymer chains were capable of rapidly binding the cationic Fe(III) catalyst, probably via a cation-exchange mechanism. Such a sorption of the photoassisted Fenton catalyst was crucial to the heterogeneous degradation process.



Environ. Sci. Technol. 2011, 45, 744–750

D. Other Impacts

Chinese patent: Preparation and photocatalytic activity of K₂Nb₄O₁₁-N, filed in January 2011 (H1000471H).

US patent: Nitrogen doped A₂Nb₄O₁₁, process for preparation thereof, and method for the degradation of organic pollutants (US 2012/0205321), filed in October 2012.

INTERNATIONAL COLLABORATIVE RESEARCH ON ENDOCRINE DISRUPTING COMPOUNDS (EDC) AND EMERGING PERSISTENT ORGANIC POLLUTANTS (POPS) IN SOUTH CHINA SEA

Shuk Han CHENG

A. Project Objectives:

1. To develop an innovative and mechanism-based in vivo test methods (using transgenic fish assay) for sensitive and rapid monitoring of algal toxins.
2. To investigate the effects of parental and embryonic exposure to environmentally relevant concentrations of propranolol on embryonic heart rate and cardiac morphology of two model fish species, *Danio rerio* and *Oryzias latipes*.
3. To characterize the differential proteomes of the toxic and nontoxic hepatic tissue of grouper, *Cephalopholis argus* and moray eel, *Gymnothorax undulates*.

B. Research Activities:

1. *Oryzias melastigma*, also called *O. dancena*, is becoming a very useful model for estuarine and marine ecotoxicity studies. With *O. melastigma* being adopted by ILSI Health and Environmental Science Institute (HESI) for embryo toxicity testing, improved knowledge of biomarker based embryonic development becomes especially important for mechanism-based toxicity evaluations. Using whole mount in situ hybridization and immunostaining techniques together with widely used molecular markers, this study describes the molecular development of marine medaka embryos, focusing on the brain, eye, heart, pectoral fin, pancreas, liver, muscle and neuron system. These organs are targets of environmental pollutants that disrupt normal embryonic development in medaka and other fish.
2. Propranolol is a beta-adrenergic receptor antagonist (beta-blocker) that is frequently used to treat hypertension and other cardiovascular conditions in humans. Detected in surface waters due to discharge of domestic wastewater, propranolol has demonstrated significant species differences in toxicity between fish. The aim of this study was to investigate the effects of propranolol on heart rate and development in embryos of two species of fish; Japanese medaka (JM) *Oryzias latipes* and zebrafish (ZF) *Danio rerio*. Parents and fertilized embryos of each species were exposed to nominal (measured) concentrations of 0.1 (0.09), 1 (1.1) and 10 (8.3) $\mu\text{g/L}$ of propranolol. Heart rate was monitored during subsequent exposure in embryos at incremental developmental periods (44, 54, 64 h post-fertilization (hpf) for ZF

and 68, 116, 164 hpf for JM). Heart development and morphology was examined using whole mount immunostaining with distance measurements between the sinus venosus (SV) and bulbus arteriosus (BA). Morphological measurements were made at 44 hpf for ZF and 164 hpf for JM. In ZF, a significant reduction in heart rate was observed at 0.08 µg/L propranolol, along with an increase in the SV–BA distance at 44 hpf. Significant reductions in heart rate were also observed in ZF at 54 and 64 hpf at all concentrations of propranolol. For JM, heart rates generally decreased at all developmental timepoints (68, 116 and 164 hpf) after propranolol treatment, with concentration dependent decreases observed at 164 hpf and a lowest observed effect concentration (LOEC) of 0.09 µg/L propranolol at each timepoint. However, significant alterations in cardiac morphology were not observed in JM at 164 hpf. In contrast, heart rates and morphology in ZF were affected with a non-monotonic concentration response in morphology and a LOEC of 0.09 µg/L propranolol for morphological alterations at 44 hpf and for heart rate at each timepoint. These data indicated unique developmental stages of susceptibility between species and that combined parental and embryo exposures may lead to greater impairment of cardiac development and function in offspring than separate exposures of adults and embryos.

3. Ciguatera fish poisoning is a global human food-borne illness caused by the consumption of coral reef fish contaminated with ciguatoxins (CTXs). Some reef fishes, such as groupers and moray eels, are more toxic than others. We hypothesized that fish containing CTXs could produce special proteins or detoxification proteins as a strategy for survival during the accumulation of CTXs. The objective of this study was to characterize the differential proteomes of the toxic and nontoxic hepatic tissue of grouper, *Cephalopholis argus* and moray eel, *Gymnothorax undulatus*, which had elevated levels of CTXs. A combination of two dimensional electrophoresis and mass spectrum approaches was employed for preliminary screening of the liver proteome of wild-caught individuals. In *C. argus* with elevated CTXs, the expression level of cytoskeleton proteins was increased, whereas those of ubiquitin enzymes, ATP related enzymes, and telomerase reverse transcriptase were greatly reduced. In CTX-containing *G. undulatus*, the proteins involved in Ca²⁺ binding, detoxification, antiapoptosis, immune defense, enhanced cell survival and proliferation were elevated. In both toxic fish species, the ATP synthase subunit beta and cytochrome c were down-regulated. However further study is needed to assess their potential roles in the resistance mechanism to contamination by CTXs. In conclusion, the comparative proteomic analysis revealed that CTXs induced influx/efflux of Na⁺ or Ca²⁺ changes in fish liver, with a concomitant interference with signal transduction, metabolomics processes, detoxification, antiapoptosis, immune defense, enhanced cell survival and proliferation etc.

C. Peer-reviewed Journal Publications Arising from this Research Project

MOLECULAR STAGING OF MARINE MEDAKA: A MODEL ORGANISM FOR MARINE ECOTOXICITY STUDY

X. Chen, L.L. J. Cheng, L.L. Chan, D.Z. Wang, K.J. Wang, M.E. Baker, G. Hardiman, D. Schlenk and S.H. Cheng

Oryzias melastigma, also called *O. dancena*, is becoming a very useful model for estuarine and marine ecotoxicity studies. With *O. melastigma* being adopted by ILSI Health and Environmental Science Institute (HESI) for embryo toxicity testing, improved knowledge of biomarker based embryonic development becomes especially important for mechanism-based toxicity evaluations. Using whole mount *in situ* hybridization and immunostaining techniques together with widely used molecular markers, this study describes the molecular development of marine medaka embryos, focusing on the brain, eye, heart, pectoral fin, pancreas, liver, muscle and neuron system. These organs are targets of environmental pollutants that disrupt normal embryonic development in medaka and other fish.

Marine Pollution Bulletin 63 (2011) 309–317

EFFECTS OF PROPRANOLOL ON HEART RATE AND DEVELOPMENT IN JAPANESE MEDAKA (*ORYZIAS LATIPES*) AND ZEBRAFISH (*DANIO RERIO*)

J. Finn, M. Hui, V. Li, V. Lorenzi, N. de la Paz, S.H. Cheng, L.L. Chan and D. Schlenk

Propranolol is a β -adrenergic receptor antagonist (β -blocker) that is frequently used to treat hypertension and other cardiovascular conditions in humans. Detected in surface waters due to discharge of domestic wastewater, propranolol has demonstrated significant species differences in toxicity between fish. The aim of this study was to investigate the effects of propranolol on heart rate and development in embryos of two species of fish; Japanese medaka (JM) *Oryzias latipes* and zebrafish (ZF) *Danio rerio*. Parents and fertilized embryos of each species were exposed to nominal (measured) concentrations of 0.1 (0.09), 1 (1.1) and 10 (8.3) μ g/L of propranolol. Heart rate was monitored during subsequent exposure in embryos at incremental developmental periods (44, 54, 64 h post-fertilization (hpf) for ZF and 68, 116, 164 hpf for JM). Heart development and morphology was examined using whole mount immunostaining with distance measurements between the sinus venosus (SV) and bulbus arteriosus (BA). Morphological measurements were made at 44 hpf for ZF and 164 hpf for JM. In ZF, a significant reduction in heart rate was observed at 0.08 μ g/L propranolol, along with an increase in the SV–BA distance at 44 hpf. Significant reductions in heart rate were also observed in ZF at 54 and 64 hpf at all concentrations of propranolol. For JM, heart rates generally decreased at all

developmental timepoints (68, 116 and 164 hpf) after propranolol treatment, with concentration dependent decreases observed at 164 hpf and a lowest observed effect concentration (LOEC) of 0.09 µg/L propranolol at each timepoint. However, significant alterations in cardiac morphology were not observed in JM at 164 hpf. In contrast, heart rates and morphology in ZF were affected with a non-monotonic concentration response in morphology and a LOEC of 0.09 µg/L propranolol for morphological alterations at 44 hpf and for heart rate at each timepoint. These data indicated unique developmental stages of susceptibility between species and that combined parental and embryo exposures may lead to greater impairment of cardiac development and function in offspring than separate exposures of adults and embryos.

Aquatic Toxicology 122–123 (2012) 214–221

PROTEOMIC ANALYSIS OF HEPATIC TISSUE OF CIGUATOXIN (CTX) CONTAMINATED CORAL REEF FISH CEPHALOPHOLIS ARGUS AND MORAY EEL GYMNOTHORAX UNDULATES

X.W. Jiang, X.M. Li, P.K.S. Lam, S.H. Cheng, D. Schlenk, Y. Sadovy de Mitcheson, Y. Li, J.D. Gu and L.L. Chan

Ciguatera fish poisoning is a global human food-borne illness caused by the consumption of coral reef fish contaminated with ciguatoxins (CTXs). Some reef fishes, such as groupers and moray eels, are more toxic than others. We hypothesized that fish containing CTXs could produce special proteins or detoxification proteins as a strategy for survival during the accumulation of CTXs. The objective of this study was to characterize the differential proteomes of the toxic and nontoxic hepatic tissue of grouper, *Cephalopholis argus* and moray eel, *Gymnothorax undulatus*, which had elevated levels of CTXs. A combination of two dimensional electrophoresis and mass spectrum approaches was employed for preliminary screening of the liver proteome of wild-caught individuals. In *C. argus* with elevated CTXs, the expression level of cytoskeleton proteins was increased, whereas those of ubiquitin enzymes, ATP related enzymes, and telomerase reverse transcriptase were greatly reduced. In CTX-containing *G. undulatus*, the proteins involved in Ca²⁺ binding, detoxification, antiapoptosis, immune defense, enhanced cell survival and proliferation were elevated. In both toxic fish species, the ATP synthase subunit beta and cytochrome c were down-regulated. However further study is needed to assess their potential roles in the resistance mechanism to contamination by CTXs. In conclusion, the comparative proteomic analysis revealed that CTXs induced influx/efflux of Na⁺ or Ca²⁺ changes in fish liver, with a concomitant interference with signal transduction, metabolomics processes, detoxification, antiapoptosis, immune defense, enhanced cell survival and proliferation etc.

Harmful Algae 13 (2012) 65–71

EFFECTS OF EXOGENOUS CARBON MONOXIDE ON RADIATION-INDUCED BYSTANDER EFFECT IN ZEBRAFISH EMBRYOS IN VIVO

V.W. Choi, M.Y. Wong, S.H. Cheng and P.K.N. Yu

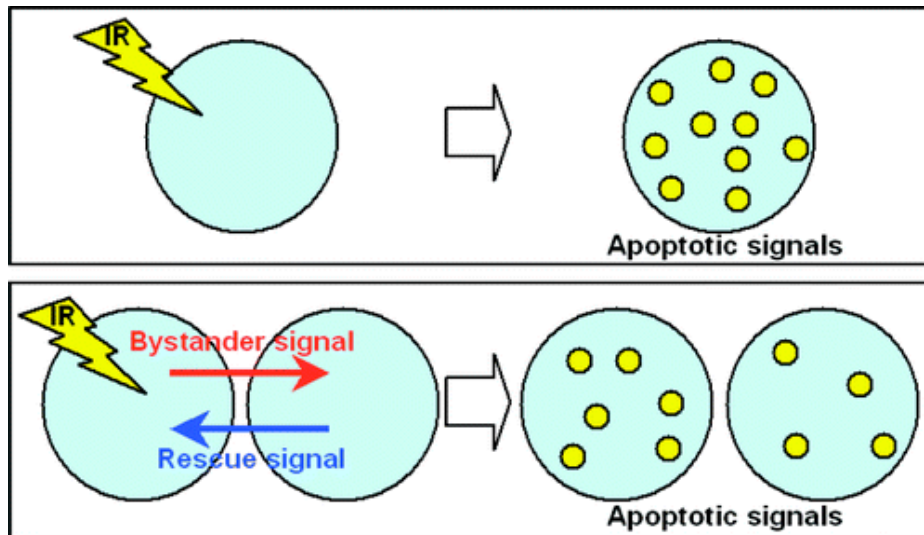
In the present work, the influence of a low concentration of exogenous carbon monoxide (CO) liberated from tricarbonylchloro(glycinato)ruthenium (II) (CORM-3) on the radiation induced bystander effect (RIBE) *in vivo* between embryos of the zebrafish was studied. RIBE was assessed through the number of apoptotic signals revealed on embryos at 25 h post fertilization (hpf). A significant attenuation of apoptosis on the bystander embryos induced by RIBE in a CO concentration dependent manner was observed.

Applied Radiation and Isotopes 70 (2012) 1075–1079

ALPHA-PARTICLE IRRADIATED ZEBRAFISH EMBRYOS RESCUED BY BYSTANDER UNIRRADIATED ZEBRAFISH EMBRYOS

V.W. Choi , C.Y. Ng , S.H. Cheng and P.K.N. Yu

We report data demonstrating that zebrafish embryos irradiated by α -particles can release a stress signal into the water, which can be communicated to the unirradiated zebrafish embryos sharing the same water medium, and then these unirradiated zebrafish embryos can release a feedback stress signal back to the irradiated embryos. The effects of radiation on the whole embryos were studied through quantification of apoptotic signals at 24 h post fertilization through staining with the vital dye acridine orange, followed by counting the stained cells under a microscope. We refer to this phenomenon as the “rescue effect”, where the unirradiated embryos successfully helped the irradiated embryos mitigate the radiation induced DNA damages. The results showed that the number of apoptotic signals in the irradiated embryos was smaller when they were partnered with bystander unirradiated embryos in the same medium. The results also showed significantly fewer apoptotic signals in the irradiated embryos when the population of bystander embryos increased from 10 to 30, while keeping the population of irradiated embryos at 10. These data suggest that the stress communicated between the unirradiated zebrafish embryos and the irradiated embryos sharing the same medium will help “rescue” the irradiated embryos, and that the strength of the rescue effect depends on the number of rescuing bystander unirradiated embryos.



Environ. Sci. Technol. 2012, 46, 226–231

TRIPHASIC LOW-DOSE RESPONSE IN ZEBRAFISH EMBRYOS IRRADIATED BY MICROBEAM PROTONS.

V.W. Choi, E.H. Yum, T. Konishi, M. Oikawa, S.H. Cheng and P.K. Yu

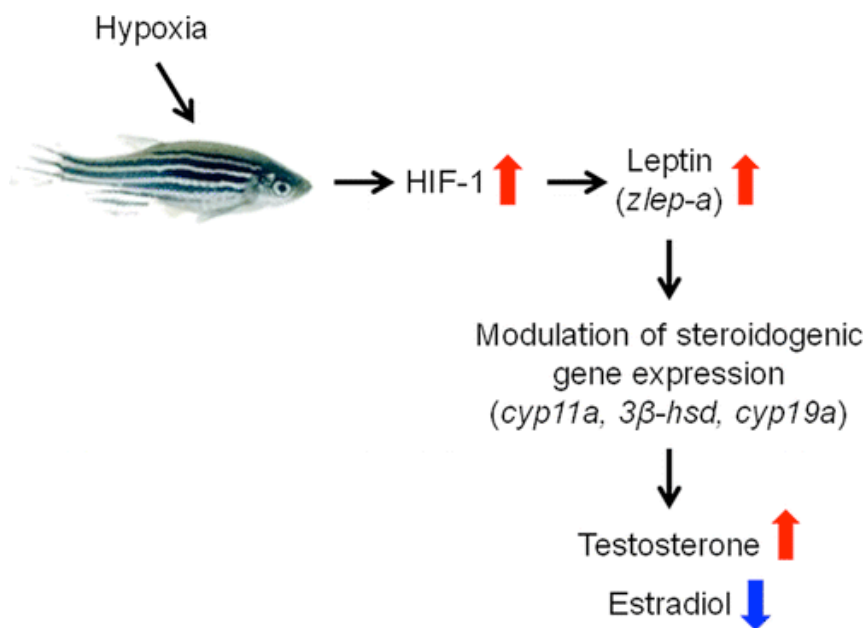
The microbeam irradiation system (Single-Particle Irradiation System to Cell, acronym as SPICE) at the National Institute of Radiological Sciences (NIRS), Japan, was employed to irradiate dechorionated zebrafish embryos at the 2-cell stage at 0.75 h post fertilization (hpf) by microbeam protons. Either one or both of the cells of the embryos were irradiated with 10, 20, 40, 50, 80, 100, 160, 200, 300 and 2000 protons each with an energy of 3.37 MeV. The embryos were then returned back to the incubator until 24 hpf for analyses. The levels of apoptosis in zebrafish embryos at 25 hpf were quantified through terminal dUTP transferase-mediated nick end-labeling (TUNEL) assay, with the apoptotic signals captured by a confocal microscope. The results revealed a triphasic dose-response for zebrafish embryos with both cells irradiated at the 2-cell stage, namely, (1) increase in apoptotic signals for <200 protons (<30 mGy), (2) hormesis to reduce the apoptotic signals below the spontaneous number for 200–400 protons (at doses of 30–60 mGy), and (3) increase in apoptotic signals again for >600 protons (at doses >90 mGy). The dose response for zebrafish embryos with only one cell irradiated at the 2-cell stage was also likely a triphasic one, but the apoptotic signals in the first zone (<200 protons or <30 mGy) did not have significant differences from those of the background. At the same time, the experimental data were in line with induction of radiation-induced bystander effect as well as rescue effect in the zebrafish embryos, particular in those embryos with unirradiated cells.

J. Radiat. Res., 53, 475–481 (2012)

LEPTIN-MEDIATED MODULATION OF STEROIDOGENIC GENE EXPRESSION IN HYPOXIC ZEBRAFISH EMBRYOS: IMPLICATIONS FOR THE DISRUPTION OF SEX STEROIDS.

R.M. Yu, D.L. Chu, T.F. Tan, V.W. Li, A.K. Chan, J.P. Giesy, S.H. Cheng, R.S.S. Wu and R.Y. Kong

Hypoxia can impair reproduction of fishes through the disruption of sex steroids. Here, using zebrafish (*Danio rerio*) embryos, we investigated (i) whether hypoxia can directly affect steroidogenesis independent of pituitary regulation via modulation of steroidogenic gene expression, and (ii) the role of leptin in hypoxia-induced disruption of steroidogenesis. Exposure of fertilized zebrafish embryos to hypoxia (1.0 mg O₂/L) from 0–72 h postfertilization (hpf), a developmental window when steroidogenesis is unregulated by pituitary influence, resulted in the up-regulation of *cyp11a*, *cyp17*, and 3β -*hsd* and the down-regulation of *cyp19a*. Similar gene expression patterns were observed for embryos exposed to 10 mM cobalt chloride (CoCl₂, a chemical inducer of hypoxia-inducible factor 1, HIF-1), suggesting a regulatory role of HIF-1 in steroidogenesis. Testosterone (T) and estradiol (E2) concentrations in hypoxic embryos were greater and lesser, respectively, relative to the normoxic control, thus leading to an increased T/E2 ratio. Expression of the leptin-a gene (*zlep-a*) was up-regulated upon both hypoxia and CoCl₂ treatments. Functional assays suggested that under hypoxia, elevated *zlep-a* expression might activate *cyp11a* and 3β -*hsd* and inhibit *cyp19a*. Overall, this study indicates that hypoxia, possibly via HIF-1-induced leptin expression, modulates sex steroid synthesis by acting directly on steroidogenic gene expression.



Environ. Sci. Technol., 2012, 46 (16), pp 9112–9119

ALPHA RADIATION EXPOSURE DECREASES APOPTOTIC CELLS IN ZEBRAFISH EMBRYOS SUBSEQUENTLY EXPOSED TO THE CHEMICAL STRESSOR, Cd.

P.K.N. Yu, M.M. Tung, V.W. Choi and S.H. Cheng

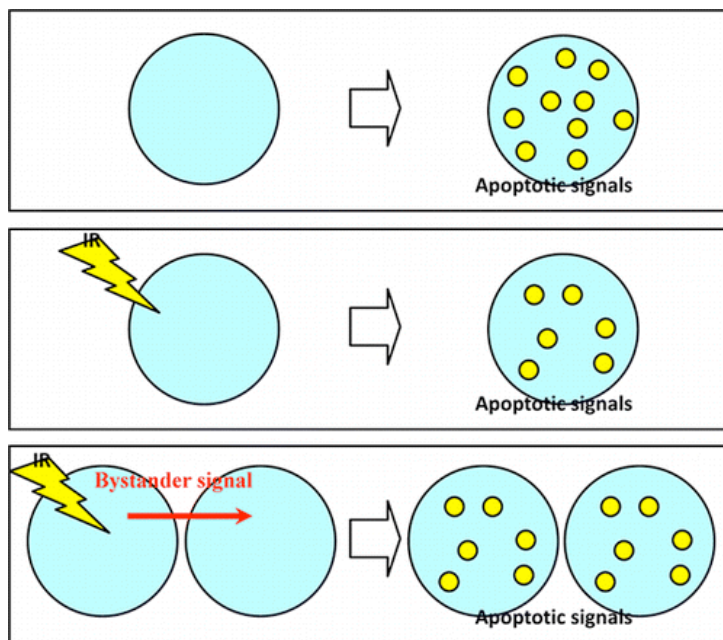
The aim of this study was to demonstrate that zebrafish embryos subjected to a priming exposure provided by one environmental stressor (low-dose alpha particles) can induce an adaptive response against a subsequent challenging exposure provided by another environmental stressor (heavy metal Cd). The effect thus identified would be an antagonistic multiple stressor effect. The effects of alpha particle radiation and/or Cd on whole embryos were studied through quantification of apoptotic signals at 24 h post-fertilization (hpf). Embryos were stained with the vital dye acridine orange, followed by counting the stained cells. For each set of experiments, 30 dechorionated embryos were divided into three groups, each having ten embryos. The three groups of embryos were referred to as (A) the control group, which received no more further treatments after dechorionation, (B) Cd-treated group, which did not receive any priming exposure and would receive a challenging exposure at 10 hpf and (C) (alpha + Cd)-treated group, which would receive both priming and challenging exposures. We defined the normalized net number of apoptotic signals in the (alpha + Cd)-treated group as $N_C^* = [(\text{apoptotic signals for (alpha + Cd)-treated group} - \text{average apoptotic signals for the corresponding control group}) / \text{average apoptotic signals for the corresponding control group}]$ and that in the Cd-treated group as $N_B^* = [(\text{apoptotic signals for Cd-treated group} - \text{average apoptotic signals for the corresponding control group}) / \text{average apoptotic signals for the corresponding control group}]$. By using the non-parametric Mann–Whitney U statistic, we were able to show that N_C^* was significantly smaller than N_B^* ($p = 0.006$). These demonstrated an antagonistic multiple stressor effect between ionizing radiation and Cd through the induction of an adaptive response by the ionizing radiation against subsequent exposures to Cd.

Environ. Sci. Pollut. Res. 2012, 19:3831–3839

HORMETIC EFFECT INDUCED BY ALPHA-PARTICLE-INDUCED STRESS COMMUNICATED IN VIVO BETWEEN ZEBRAFISH EMBRYOS

V.W. Choi, A.L. Cheung, S.H. Cheng and P.K.N. Yu

We report data showing that embryos of the zebrafish, *Danio rerio*, at 1.5 h post fertilization (hpf) subjected to a low-dose alpha-particle irradiation can release a stress signal into the water, which can be communicated to unirradiated bystander zebrafish embryos sharing the same water medium to induce a hormetic effect in the bystander embryos. Hormetic responses are characterized as biphasic dose–response relationships exhibiting a low-dose stimulation and a high-dose inhibition. The effects on the whole embryos were studied through quantification of apoptotic signals at 24 hpf through staining with the vital dye acridine orange, followed by counting the stained cells under a microscope. The results show that, for low alpha-particle dose, the number of apoptotic signals decreases in the irradiated embryos and also in the unirradiated bystander embryos having partnered with the irradiated embryos. These suggested that alpha-particle-irradiated zebrafish embryos could release a stress signal into the water, which could be communicated to unirradiated bystander zebrafish embryos sharing the same water medium to induce a hormetic effect in the bystander embryos.



Environ. Sci. Technol., 2012, 46:11678–11683

LONG-TERM MEASUREMENTS OF ULTRAVIOLET RADIATION IN MARINE ENVIRONMENTS IN HONG KONG

Peter Kwan Ngok YU

A. Project Objectives

1. Measurement of UV sensitivity of EBT films under water;
2. Studies on the sensitivities of EBT films to UVA and UVB spectrums;
3. UV measurements in marine environments using EBT films.

B. Research Activities

1. Measurement of UV sensitivity of EBT films under water

We measured the UV sensitivity of EBT films under water. There were negligible changes in the absorption spectra of irradiated EBT Gafchromic film. The films were also characterized for UV measurements in control conditions.

2. Studies on the sensitivities of EBT films to UVA and UVB spectrums

We used EBT films to measure the solar ultraviolet radiation, and optimized the methodology regarding the use of radiochromic films and the scanning techniques. The results were published in the Physics in Medicine and Biology journal in 2010.

3. UV measurements in marine environments using EBT films

We tested the performance of EBT films for UV measurements in marine environments and compared it with the UV transmission through clear/clean water and muddy/more polluted water, which showed large differences in UV penetration for these different scenarios. A journal paper is being prepared from these results.

C. Peer-reviewed Journal Publication Arising from this Research Project

MEASURING SOLAR UV RADIATION WITH EBT RADIOCHROMIC FILM

T. Ethan, B.T. Cheung, P.K.N. Yu and M.J. Butson

Ultraviolet radiation dosimetry has been performed with the use of a radiochromic film dosimeter called Gafchromic EBT for solar radiation exposure. The film changes from a clear colour to blue colour when exposed to ultraviolet radiation and results have shown that the colour change is reproducible within $\pm 10\%$ at 5 kJ m^{-2} UV exposure under various conditions of

solar radiation. Parameters tested included changes in season (summer versus winter exposure), time of day, as well as sky conditions such as cloudy skies versus clear skies. As the radiochromic films' permanent colour change occurs in the visible wavelengths the film can be analysed with a desktop scanner with the most sensitive channel for analysis being the red component of the signal. Results showed that an exposure of 5 kJ m^{-2} (approximately 1 h exposure in full sun during summer) produced an approximate 0.28 change in the net OD when analysed in reflection mode on the desktop scanner which is significant darkening. The main advantages of this film type, and thus the new EBT2 film which has replaced EBT for measurement of UV exposure, is the visible colour change and thus easy analysis using a desktop scanner, its uniformity in response and its robust physical strength for use in outside exposure situations.

Phys. Med. Biol., 2010, 55 N487

SORPTION AND DEGRADATION OF POLYBROMINATED DIPHENYL ETHERS (PBDES) BY GREEN MICROALGAE WITH AND WITHOUT THE EFFECT OF METALS

Nora Fung Yee TAM

A.Peer-reviewed Journal Publication Arising from this Research Project

REMOVAL AND BIODEGRADATION OF NONYLPHENOL BY IMMOBILIZED *CHLORELLA VULGARIS*

Q.T. Gao, Y.S. Wong and N.F.Y. Tam

The removal and biodegradation of nonylphenol (NP) by alginate-immobilized cells of *Chlorella vulgaris* were compared with their respective free cultures. The effects of four cell densities of 10^4 per algal bead were investigated, as were the four algal bead concentrations, with regard to the removal and biodegradation of NP. Although immobilization significantly decreased the growth rate and NP's biodegradation efficiency of *C. vulgaris*, NP removal over a short period was enhanced. The NP removal mechanism by immobilized cells was similar to that by free cells, including adsorption onto alginate matrix and algal cells, absorption within cells and cellular biodegradation. The optimal cell density and bead concentration for the removal and biodegradation of NP was $50\text{--}100 \times 10^4$ cells algal bead $^{-1}$ and $2\text{--}4$ beads ml $^{-1}$ of wastewater, respectively. These results demonstrated that immobilized *C. vulgaris* cells under optimal biomass and photoautotrophic conditions are effective in removing NP from contaminated water.

Bioresource Technology 102 (2011) 10230–10238

GROWTH, PHOTOSYNTHESIS AND ANTIOXIDANT RESPONSES OF TWO MICROALGAL SPECIES, *CHLORELLA VULGARIS* AND *SELENASTRUM CAPRICORNUTUM*, TO NONYLPHENOL STRESS

Q.T. Gao and N.F.Y. Tam

The effect of nonylphenol (NP) on growth, photochemistry and biochemistry of two green microalgae, *Chlorella vulgaris* and *Selenastrum capricornutum*, and their ability to degrade NP were compared. The 96 h EC $_{50}$ of *C. vulgaris* and *S. capricornutum* were greater than 4.0 and 1.0 mg L $^{-1}$ NP, respectively, suggesting that the former species was more tolerant to NP. Both microalgae acclimated to NP stress through down-regulating their photosynthetic activities, including antenna size (chlorophyll a content), maximal photochemistry (Fv/Fm) and the light absorbed by PSII (ABS/CS $_0$), but the dissipation of energy from reaction centres (DI $_0$ /RC)

increased with the increase of NP concentrations. In *C. vulgaris*, the changes of these parameters were more significant than in *S. capricornutum* and recovered completely after a 96 h exposure. The antioxidant responses, such as GSH content, CAT and POD activities in *C. vulgaris* increased with the increase of NP concentrations after a 24 h exposure, but these changes disappeared with exposure time and recovered to the control levels after 96 h. In *S. capricornutum*, although GSH content, CAT and POD activities also increased when exposed to low- to moderate-NP concentrations, these values were significantly reduced at a high concentration (4 mg L^{-1}) even after a 96 h exposure, indicating its antioxidant responses were significantly delayed. It is clear that the more NP-tolerant species, *C. vulgaris*, acclimated better with a faster recovery of its photosynthetic activity from the NP-induced damage, and exhibited more efficient and rapid responses to NP-induced oxidative stress. *C. vulgaris* also had a higher NP degradation ability than *S. capricornutum*.

Chemosphere 82 (2011) 346–354

UNRAVELING TISSUE-SPECIFIC MECHANISMS FOR IN VIVO REGULATION OF ESTROGEN TARGET GENES IN MEDAKA

Doris Wai Ting AU

A. Project Objectives

1. To study tissue expression patterns of different estrogen receptor (ER) isoforms in medaka;
2. To investigate how each form of medaka ER is involved in regulating the activity of each functional ERE present in the medaka TERT promoter.

B. Research Activities

- Gene expression of all known subtypes of ER and ERR in multiple organs and both sexes of the Japanese medaka (*Oryzias latipes*) was profiled and systematically analyzed. In contrast with the ubiquitous nature of ERs, the expressions of ERs and ERRs are proven organ- and sex-dependent. Moreover, expressions of specific ERR isoforms (ERR γ 1, ERR γ 2) are strongly correlated with that of all ERs (ER α , ER β 1 and ER β 2), suggesting the existence of potential interactions. Findings of this study shed light on the co-regulatory role of particular ERRs in estrogen-ERs signaling and highlight the potential importance of ERRs in determining organ- and sex-specific estrogen responses. Using medaka as an alternative vertebrate model, the present study provide new directions that call for collective efforts from the scientific community to unravel the mechanistic action of ER-ERR cross talks, and their intertwining functions, in a cell- and sex-specific manner *in vivo*.
- According to the deletion mutants of oITERT promoter, the Region D (195 bp) is the most important region of promoter, regarded as the core promoter. Point-mutants (ERE mutation) on the whole promoter did not show the predicted response influencing down-stream gene expression. But point mutants (AP-1 element mutation) in core promoter showed highly regulation effect to down-stream gene with ER α and E2, especially for AP-1 element mutants. There are five AP-1 elements and ER elements in the shorter promoter. According to the analysis of point mutants in this short promoter, the fourth Ap-1 element closely up-stream of transcription start site (TS) is the most important cis-element in regulating down-stream gene expression of oITERT promoter by E2 and ER α . The fifth AP-1 element closely down-stream of transcription start site maintains the normal induced-expression level of the gene regulated by oITERT promoter. If it is mutated, the down-stream gene regulated by oITERT promoter will be highly over-expressed with the regulation of E2 and ER α . When there are the AP-1 elements in the oITERT promoter, the regulation effect of EREs is not seemly important for inducing expression of TERT by E2 and ER α .

C. Peer-reviewed Journal Publications Arising from this Research Project

ISTA15: NEW DIRECTIONS IN ECOTOXICOLOGY AND MEETING THE CHALLENGE AHEAD.

D.W.T. Au

This special issue of Environmental Science and Pollution Research highlights selected papers presented at the 15th International Symposium on Toxicity Assessment (ISTA15) which was held in Hong Kong, from 3 to 8 July 2011, and officially hosted by the City University of Hong Kong.

In the Asia Pacific region in general, and Hong Kong and China in particular, governments now recognize that proper management of the environment is vital to protect public health and sustain socioeconomic development. In China, rapid population growth and industrial development have occurred in the Pearl River Delta, which is now known as “the world’s factory”. The emission and disposal of enormous quantities of industrial and domestic waste have exerted unprecedented pressures on our environment. More-over, many large infrastructural development schemes have caused great damage to our ecosystems and serious economic losses, and have posed significant threats to public health. The impact of these developments needs to be predicted, identified, assessed, monitored, and controlled in order to avoid costly or irreversible mistakes.

ISTA15 brought together academics, professionals, and policy makers to discuss new developments, approaches, and strategies in the fields of toxicity assessment, ecotoxicology, regulatory toxicity, and risk assessment for ensuring environmental safety and public health. A total of 300 registered participants from over 33 countries attended the ISTA15 which was characterized by these 10 major scientific themes:

- Biomarkers and bioindicators
- Biotoxins
- Chemical toxicities and mechanisms of action
- Chemicals of global and emerging concerns
- Environmental chemistry and green chemistry
- Environmental safety and public health
- “Omics” and bioinformatics in (eco)toxicology
- Regulatory toxicology
- Remediation and mitigation
- Risk assessment and environmental management

Environ. Sci. Pollut. Res., 2012, 19:2463–2464

GENDER SPECIFIC MODULATION OF IMMUNE SYSTEM COMPLEMENT GENES EXPRESSION IN MARINE MEDAKA *ORYZIAS MELASTIGMAS* FOLLOWING DIETARY EXPOSURE OF BDE-47.

R.R. Ye, E.N.Y. Lei, M.H.W. Lam, A.K.Y. Chan, J. Bo, J.P. van de Merwe, A.C.C. Fong, M.M.S. Yang, J.S. Lee, H.E. Segner, C.K.C. Wong, R.S.S. Wu and D.W.T. Au

BDE-47 is one of the most widely found congeners of PBDEs in marine environments. The potential immunomodulatory effects of BDE-47 on fish complement system were studied using the marine medaka *Oryzias melastigma* as a model fish. Three-month-old *O. melastigma* were subjected to short-term (5 days) and long-term (21 days) exposure to two concentrations of BDE-47 (low dose at 290 ± 172 ng/day; high dose at 580 ± 344 ng/day) via dietary uptake of BDE-47 encapsulated in *Artemia nauplii*. Body burdens of BDE-47 and other metabolic products were analyzed in the exposed and control fish. Only a small amount of debrominated product, BDE-28, was detected, while other metabolic products were all under detection limit. Transcriptional expression of six major complement system genes involved in complement activation: C1r/s (classical pathway), MBL-2 (lectin pathway), CFP (alternative pathway), F2 (coagulation pathway), C3 (the central component of complement system), and C9 (cell lysis) were quantified in the liver of marine medaka. Endogenous expression of all six complement system genes was found to be higher in males than in females ($p < 0.05$). Upon dietary exposure of marine medaka to BDE-47, expression of all six complement genes were downregulated in males at day 5 (or longer), whereas in females, MBL-2, CFP, and F2 mRNAs expression were upregulated, but C3 and C9 remained stable with exposure time and dose. A significant negative relationship was found between BDE-47 body burden and mRNA expression of C1r/s, CFP, and C3 in male fish ($r = -0.8576$ to -0.9447). The above findings on changes in complement gene expression patterns indicate the complement system may be compromised in male *O. melastigma* upon dietary exposure to BDE-47. Distinct gender difference in expression of six major complement system genes was evident in marine medaka under resting condition and dietary BDE-47 challenge. The immunomodulatory effects of BDE-47 on transcriptional expression of these complement components in marine medaka were likely induced by the parent compound instead of biotransformed products. Our results clearly demonstrate that future direction for fish immunotoxicology and risk assessment of immunosuppressive chemicals must include parallel evaluation for both genders.

Environ. Sci. Pollut. Res., 2012, 19:2477–2487

D. Recognized international conference in which papers related to this research project was delivered

ESTROGEN RECEPTORS (ERS) AND ESTROGEN RELATED RECEPTORS (ERRS) IN THE JAPANESE MEDAKA *ORYZIAS LATIPES*

A.C.K Cheung, W. Ge, J.P Giesy and D.W.T Au

In the past decade, there has been increasing concern about environmental pollution by various endocrine disrupting chemicals (EDCs), particularly those with estrogenic activities. Estrogen receptors (ERs) are involved in signalling pathways affected by some EDCs. To understand the potential adverse effects of EDCs and to screen for these effects additional information on the mechanisms of response would be useful since current understanding is incomplete. Estrogen-related receptors (ERRs) are involved in pathways without direct interaction with estrogen. Previous studies have focused on interactions between estrogen and either ERs or ERRs. However, since there can be dynamic interactions between these two classes of receptors, understanding interactions between ERs and ERRs will facilitate interpretation of screening results and the development of biomarkers or response.

The three classic ERs (ER α , ER β and ER γ) and five ERRs (ERR α , ERR β , ERR γ , ERR δ and ERR ϵ) reported in mammals are also present in Japanese medaka, *Oryzias latipes* which make medaka a useful non-mammalian animal model. The objective of this study was to determine the gender- and tissue-specific (brain, gill, gonad, heart, liver and spleen) distribution of ERs and ERRs in adult (8-M) *Oryzias latipes*, and to explore interaction between and among ERs and ERRs. Expression mRNA of oERs and oERRs were determined by qPCR and normalized to expression of RPL-7. Gender-specific distribution were observed in most of the organs studied. Tissue-specific distributions were observed for oERR β , γ and δ . These genes were expressed in some, but not all of the organs. The results of principal component analysis (PCA) were consistent with interactions among organs and genes. A description of basal distribution of ERs and ERRs in tissues of Japanese medaka is provided.

JAPANESE MEDAKA AS ALTERNATIVE VERTEBRATE MODEL FOR GENDER AND AGING STUDIES

D.W.T. Au

It has been well established in mammals (including humans) that females live longer than males. Our laboratory studies on Japanese medaka (*Oryzias latipes*) also show a similar longevity gender gap in aging population. *Oryzias latipes* confers significant advantages for gender and aging studies over other small laboratory fish because i) similar to humans, Japanese medaka undergo a gradual senescence and telomere shortening during aging; ii) the *Dmy* in *O. latipes* is the only sex-determining gene reported in non-mammalian vertebrates thus far, enabling the exact determination of gender in individual medaka as early as 7-day post fertilization, well before phenotypic sex is discernible. Our recent development of *Dmy* gender-typing on high throughput *O. latipes* embryo chip can unveil early life stage gender-specific responses of Japanese medaka to a variety of endogenous and exogenous stimuli. Notably, many diseases (e.g. osteoporosis, immune related diseases) in humans are known to be gender and age-related. Estrogen (E2) has been proposed as the key factor contributing to the observed gender difference in bone development, immune function, aging and life expectancy in mammals. Interestingly, our pilot studies showed that estrogen dynamics and telomere biology of female and male *O. latipes* are very similar to those of humans. The potential use of *O. latipes* as alternative non-mammalian vertebrate model to unravel the role of estrogen for gender difference in telomere attrition, aging and other age-related diseases will be discussed.

Summary of the Seed Collaborative Research Fund (SCRF) Projects SCRF項目概要

SCRF

SKLMP Funded Project Progress Report

Jan 2012 - Dec 2014 (Ongoing)

ASSESSING THE IMPACTS OF ORGANIC AND METAL POLLUTION ON SYMBIOTIC MICROBIAL COMMUNITIES IN MARINE CORALS AND SPONGES BY METAGENOMICS AND TRANSCRIPTOMICS APPROACHES

Peiyuan QIAN

Corals and sponges are two major groups of marine benthos around the world. Apart from providing shelter, protection from currents and predators, and areas for breeding, spawning, feeding and resting for fish and other species, they are of great conservation values and also important sources for antibiotics and drugs. Indeed, many potential pharmaceuticals are not directly produced by the corals and sponges, but the symbiotic micro-organisms therein. Despite their great ecological and economic values, the diversity and abundance of corals and sponges in the world experienced a tremendous decline over the last 30 years, mainly because of pollution, global climate changes, habitat destruction, and emerging diseases. In Hong Kong, coastal development and reclamation and pollution from sewage, heavy metal and other industrial wastes pose serious threats on corals and sponges. It has been estimated that about 90% of coral communities in Hong Kong is subject to a high degree of threats from pollution, yet only few works have been carried out to evaluate the impacts of pollution on corals and sponges. It has been well documented that the well being of corals and sponges rely on their symbiotic microorganisms. We hypothesize that that environmental stress may affect the structure and function of these symbiotic microbes, therefore affecting the well being of corals and sponges. In this project, experiments are designed to examine the impacts of selected organic and metal pollutants on the composition and function of these symbiotic microbes, and relate this to the health of corals and sponges. The result of this study will provide a thorough understanding on the relationships between pollution, symbiotic microbial communities and their coral and sponge hosts, which remained poorly known.

A. Project Objectives

1. To evaluate the impacts of organic and metal pollutants on the health of selected Hong Kong coral and sponge;
2. To study the structural and functional responses of symbiotic microbial communities to metal and organic pollutants using a metagenomic and transcriptomic approach; and
3. To relate the responses of the symbiotic microbial community to the health of corals and sponges.

B. Research Activities

1. Experimental design

After discussion with several experts in the field, paying site visits to local fish farms for checking coral and sponge populations and carrying out an extensive literature review on the subject areas, we chose PBDEs (BDE47 congener) as the target pollutant and the sponge *Haliclona cymaeformis* as the testing organism for this study. This study aimed at assessing the effect of PBD47 on the microbial community associated with the sponge. Preliminary experiments were performed with DGGE to decide the duration and dosage of BDE47 treatment. After that, lab-controlled experiments were setup in aquaria using healthy sponge colonies collected from the field to test the effect of 6 d and 12 d exposure ('short-term' and 'long-term' treatments, respectively) of the sponge to 10 ng/L and 1 µg/L ('low-dose' and 'high-dose' treatments, respectively) of BDE47 on the sponge-associated microbial communities. At each time point, treated sponge tissues and controls (both blank and DMSO controls) were collected and preserved in DNA extraction buffer and RNA later for metagenome and transcriptome studies, respectively. DNAs were extracted and amplified by PCR using the universal 16S rRNA gene primers, 341F and 907R. Microbial community compositions were profiled by DGGE and barcoded-tag prosequencing.

2. Results

16S rRNA gene amplicons from different samples (N0, D2, L2, H2, D6, L6 and H6) were mixed and pyrosequenced on the ROCHE 454 Titanium platform (Table 1). About 100,000 raw reads with an average read length of more than 400bp. After quality filtering, about 62,000 reads were qualified for subsequent analysis by QIIME pipeline. Microbial diversity in the DMSO control sponge was high and treatment of BDE47 decrease microbial diversity. The microbial communities in the sponges showed a dynamic change in composition along with treatment dosage and duration. All the associated microbial communities, except for the long-term high-dosage treatment (H6), were dominant by the genus *Nitrosococcus*, with the relative abundance ranging from 42 to 89%. The relative abundance of this genus dropped to 4.3% in H6 and its dominance was replaced by the genus *Clostridium* (43%), which is a potential pathogen. Other bacteria including *Muricauda* and *Caedibacter* were also reduced in H6. In the short-term high-dosage treatment (H2), the enrichment of *Spirochaeta*, the *Marinilabiaceae*,

Meridianimaribacter and *Arcobacter* and the reduction of an uncultured *Alphaproterium* were observed when compared with the control. However, the microbial communities of the low-dosage treatments (L2 and L6), regardless of the treatment duration, were similar when compared with their corresponding DMSO controls (D2 and D6). Jackknife-tree clustering supported a high similarity among microbial communities in L2 compared with D2 and N0, yet the high-dosage treatment seemed to exert an effect on the communities at day 2. The cluster formed by the samples from day 0 and 2 was distantly separated from the cluster formed by L6 and D6 where a relatively high similarity was observed. H6 was clearly distinct from the rest of the samples with a high bootstrap support, indicating its uniqueness in terms of microbial community composition. These results indicated that BDE47 shifted the microbial communities in the sponge in a concentration- and time-dependent manner. The sponge-associated microbial community was changed from an ammonium or sulfur-oxidizing-dominant community to a pathogen-dominant community.

By visual comparison of DGGE band patterns, the microbial communities in L2 and H2 were similar to those in N2 and N0, except that decreases in intensity of certain bands were noticed. In contrast, substantial differences were observed in H6 compared to D6 and L6, indicating a strong effect of high-dosage BDE47 treatment on the sponge-associated microbial communities. The two dominant bands were excised and sequenced. Sequencing data suggested that the bands belonged to the genus *Nitrosococcus*. The intensities of these bands were decreased in the L2, H2 and L6 treatments and became barely detectable in H6, suggesting a significant drop of this genus in the treatment groups, especially in the long-term high dosage treatment. These results were consistent with our observations based on barcoded-tag pyrosequencing and further confirmed the shift in the sponge-associated microbial community by BDE47.

3. Problems encountered

Due to the technical difficulty in sampling and culturing adult corals in the lab, we decided to focus on sponge as the testing organism for this study. PCR products for DGGE and pyrosequencing were difficult to prepare. We spent much time selecting right primers and conditions for PCR. For unknown reasons, mixing equal amount of PCR products for pyrosequencing did not result in an even distribution of reads among different samples. Some samples did not have enough data for subsequent analysis and thus more samples needed to be prepared for additional pyrosequencing.

C. Peer-reviewed Journal Publication Arising from this Research Project

SPATIAL AND SPECIES VARIATIONS IN BACTERIAL COMMUNITIES ASSOCIATED WITH CORALS FROM THE RED SEA AS REVEALED BY PYROSEQUENCING

O.O. Lee, J. Yang, S. Bougouffa, Y. Wang, Z. Batang, R. Tian, A. Al-Suwailem and P.Y. Qian

Microbial associations with corals are common and are most likely symbiotic, although their diversity and relationships with environmental factors and host species remain unclear. In this study, we adopted a 16S rRNA gene tag-pyrosequencing technique to investigate the bacterial communities associated with three stony *Scleractinea* and two soft *Octocorallia* corals from three locations in the Red Sea. Our results revealed highly diverse bacterial communities in the Red Sea corals, with more than 600 ribotypes detected and up to 1,000 species estimated from a single coral species. Altogether, 21 bacterial phyla were recovered from the corals, of which *Gammaproteobacteria* was the most dominant group, and *Chloroflexi*, *Chlamydiae*, and the candidate phylum *WS3* were reported in corals for the first time. The associated bacterial communities varied greatly with location, where environmental conditions differed significantly. Corals from disturbed areas appeared to share more similar bacterial communities, but larger variations in community structures were observed between different coral species from pristine waters. Ordination methods identified salinity and depth as the most influential parameters affecting the abundance of *Vibrio*, *Pseudoalteromonas*, *Serratia*, *Stenotrophomonas*, *Pseudomonas*, and *Achromobacter* in the corals. On the other hand, bacteria such as *Chloracidobacterium* and *Endozoicomonas* were more sensitive to the coral species, suggesting that the host species type may be influential in the associated bacterial community, as well. The combined influences of the coral host and environmental factors on the associated microbial communities are discussed. This study represents the first comparative study using tag-pyrosequencing technology to investigate the bacterial communities in Red Sea corals.

Appl. Environ. Microbiol., 2012, 78:7173-7184

HEALTH RISK ASSESSMENTS OF RESIDENTS IN THE PEARL RIVER DELTA EXPOSED TO BROMINATED FLAME RETARDANTS (BFRs)

Ming Hung WONG

The concentrations of polybrominated diphenyl ethers (PBDEs) and novel brominated flame retardants (nBFRs) such as methoxylated (MeO-) and hydroxylated (OH-) PBDEs in the environment and humans have been increasing in recent years all over the world. This is particularly the case in the Pearl River Delta (PRD), which is a global power house producing a large amount of electronic products. As important environmental endocrine disruptor chemicals, these organobrominated compounds have the potential to cause serious thyroid hormone disruption, neurotoxicity and adverse developmental effects in humans. However, our knowledge about the intakes, body accumulation and potential transgenerational exposures to these BFRs, especially nBFRs, is very limited. We hypothesize that (1) PBDEs and nBFRs could accumulate in human bodies via inhalation and food ingestion, especially fish consumption, and (2) The accumulated PBDEs and nBFRs in the maternal body can be biotransferred to the next generation via transplacental transport and breast feeding. The major objectives of the present proposal are to develop validated methods for the analysis of nBFRs, investigate their occurrences in foodstuffs, indoor dust and human tissues and to evaluate the associated health risks. This proposed project comprises 6 parts: (1) Determination of average daily intakes of BFRs for residents in the PRD, via consumption of fish and other food products available in the region; (2) Determination of average inhalation intakes of BFRs for residents in PRD, by measuring BFRs in indoor dust samples; (3) Measuring the transfer of BFRs through the intestinal barrier using the Caco-2 cell line model, the transport of BFRs across human cultured alveolar A549 cell monolayers and their tissue distribution in an animal model; (4) Assessing human health risks of organobromine compounds in human blood and breast milk samples, and the correlation of BFRs concentrations between daily intakes and human body burdens; (5) Determining transgenerational exposure of BFRs to newborns in the region via placental transfer and breast feeding; and (6) Reviewing the current issues and providing guidance concerning food consumption for local residents, especially for sensitive populations such as pregnant woman, women of childbearing age, and infants, with regards to BFRs.

A. Project Objectives

The major aim of this project is to determine the daily intake of organobromine compounds via food ingestion and dust inhalation and ingestion for residents in the Pearl River Delta (PRD), their

accumulation in the human body and transgenerational exposure via placental transfer and breast feeding. More specifically, the objectives of the proposed project are:

1. To investigate the daily intakes of a variety of organobromine compounds via the consumption of fish and other food products collected from the PRD;
2. To evaluate inhalation intakes of organobromine compounds for residents, based on their concentrations in indoor dust;
3. To study the transfer of BFRs through the intestinal barrier using the Caco-2 cell line model, the transport of BFRs across human cultured alveolar A549 cell monolayer and to study their tissue distribution in an animal model;
4. To evaluate the human health risks of organobromine compounds contained in different human tissues (blood and milk), and the correlation between daily BFR intakes and human body burdens;
5. To determine the transgenerational exposure to organobromine compounds to offspring via placental transfer and breast feeding;
6. To review the current issues and provide guidance on food consumption for local residents, especially for sensitive populations such as pregnant woman, women of childbearing age and infants, with regards to BFRs.

B. Research Activities

1. Food consumption survey

A food consumption survey involving 25 pregnant women was conducted (please see below for details) in The Third Affiliated Hospital of Guangzhou Medical University, in Guangzhou. These women came from 5 cities, namely Zhuhai (ZH), Dongguan (DG), Guangzhou (GZ), Foshan (FS), and Zhongshan (ZS), around the Pearl River Delta (PRD):

In order to study the consumption rate for representative food types in PRDs, international references were reviewed during the course of food selection. These included the List of Priority Food and Contaminants prepared by the WHO GEMS/Food, total diet studies by the US FDA (2004) and local references. Subsequently, 86 food items (FIs) were selected and divided into 11 groups, namely [i] freshwater fish (FIs = 4), [ii] marine fish (FIs = 3), [iii] meat (FIs = 2), [iv] poultry (FIs = 2), [v] animal viscera (FIs = 7), [vi] egg (FIs = 4), [vii] milk and dairy products (FIs = 3), [viii] cereal (FIs = 7), [ix] vegetable (FIs = 35), [x] fruit (FIs = 13), [xi], and beverages (FIs = 6) for their respective consumption rates. The questionnaire was the same as the one employed in our previous study (Chan et al., 2007, 2012a; 2012b). Dietary change over the course of pregnancy was ignored. There was 100% response rate.

2. Sampling of Food items

According to the food consumption survey results (i.e. local consumption pattern) and food items availability in the markets of the studied cities, a total of 40 food items (out of the original 86) were selected for further food basket analyses, focusing on PBDEs and novel brominated flame retardants (nBFRs) such as methoxylated (MeO-) and hydroxylated (OH-) PBDEs, contained in different food items. The number of food groups was modified from 11 to 9 major food groups, in which the two groups: [viii] milk and dairy products and [xii] beverages, were omitted from further chemical analyses. Consequently, the 40 food items were further regrouped into 9 major food groups in each of the 5 cities (ZH, DG, GZ, FS, and ZS): [i] freshwater fish (Fls = 4), [ii] marine fish (Fls = 2), [iii] meat (Fls = 3), [iv] poultry (Fls = 2), [v] egg (Fls = 1), [vi] animal viscera (Fls = 3), [vii] vegetable (Fls = 12), [viii] cereal (Fls = 7), and [ix] fruit (Fls = 3). Food sampling was subsequently conducted, with food items randomly selected.

3. Human samples collection and analysis

Twenty five sets of human samples [(1) human milk, (2) blood, (3) placenta] were also collected from the 25 pregnant women who were involved in the food consumption survey, from the same hospital, after going through the Ethics Committee. The human samples will be used to monitor the body loadings of the above mentioned PBDEs levels in PRDs.

4. Collaboration activities

The human samples will be analyzed in the Department of Microbial and Biochemical Pharmacy School of Pharmaceutical Sciences Sun Yat-sen University, Guangzhou, under the supervision of one of the team members, Dr HS Wang.

C. Peer-reviewed Journal Publications Arising from this Research Project

DIETARY EXPOSURE TO POLYCHLORINATED DIBENZO-*P*-DIOXINS AND DIBENZOFURANS VIA FISH CONSUMPTION AND DIOXIN-LIKE ACTIVITY IN FISH DETERMINED BY H4IIE-LUC BIOASSAY

J.K.Y. Chan, Y.B. Man, G.H. Xing, S.C. Wu, M.B. Murphy, Y. Xu and M.H. Wong

Dietary exposure to polychlorinated dibenzo-*p*-dioxins and dibenzofurans (PCDD/Fs) via fish consumption in two major electronic (e) waste sites: Guiyu (GY), Guangdong Province and Taizhou (TZ), Zhejiang Province, and dioxin-like activity in fish determined by H4IIE-luc bioassay. In the present study, all fish were below EU's maximum allowable concentration in muscle of fish (4 pg WHO-TEQ/g wet wt), except crucian (4.28 pg WHO-TEQ/g wet wt) and silver carps (7.49 pg WHO-TEQ/g wet wt) collected from GY rivers. Moreover, the residual concentration in bighead

carp collected from GY (2.15 pg WHO-TEQ/g wet wt) was close to the EU's action level (3 pg WHO-TEQ/g wet wt) which gives "early warning" to the competent authorities and operators to take measures to eliminate contamination. In addition, results indicated that the maximum human intake of PCDD/Fs via freshwater fish consumption in GY was 4.31 pg WHO-TEQ/kg bw/day, which exceeds the higher end of the tolerable daily intake recommended by the WHO, EC-SCF and JECFA (1–4, 2 and 2.3 pg WHO-TEQ/kg bw/day respectively). Furthermore, H4IIE-luc cell bioassay provides a very sensitive and cost-efficient screening tool for assessing the overall dioxin-like toxicity in the study, and is therefore valuable for high-throughput environmental monitoring studies.

Science of the Total Environment (In Press)

DIETARY INTAKE OF PBDES OF RESIDENTS AT TWO MAJOR ELECTRONIC WASTE RECYCLING SITES IN CHINA

J.K.Y. Chan, Y.B. Man, S.C. Wu and M.H. Wong

The dietary intake of polybrominated diphenyl ether (PBDE) of local residents from 2 major electronic waste (e-waste) processing sites (Guiyu, Guangdong Province and Taizhou, Zhejiang Province) in China was investigated. Seventy-four food items were collected from these sites, divided into 9 food groups (freshwater fish, marine fish, shellfish, pork, poultry, chicken offal, egg, vegetables and cereals), and examined for residual PBDE concentrations. Out of all food items examined, the freshwater bighead carp (*Aristichthys nobilis*) contained extremely high (11,400±254 ng/g wet wt.) concentrations of PBDE, the highest concentrations amongst published data concerning PBDE detected in freshwater fish. Food consumption data obtained through semi-quantitative food intake questionnaires showed that Guiyu residents had a PBDE dietary intake of 931±772 ng/kg bw/day, of which BDE-47 (584 ng/kg bw/day) exceeded the US EPA's reference dose (100 ng/kg/day). Taizhou (44.7 ± 26.3 ng/kg bw/day) and Lin'an (1.94 ± 0.86 ng/kg bw/day) residents exhibited lower readings. The main dietary source of PBDEs in Guiyu and Taizhou residents was seafood (88–98%) and pork (41%) in Lin'an. The present results indicated that health risks arising from PBDE dietary exposure are of significance in terms of public health and food safety to local residents of e-waste processing sites.

Science of the Total Environment (In Press)

HYDROXYLATED AND METHOXYLATED POLYBROMINATED
DIPHENYL ETHERS IN BLOOD SERUM OF HUMANS IN BLOOD
PLASMA OF HUMANS IN HONG KONG.

H.S. Wang, Z.J. Chen, K.L. Ho, L.C. Ge, J.D. Lam, M.H.W. Lam, J.P. Giesy, M.H. Wong and C.K.C. Wong

Hydroxylated (OH-) and methoxylated (MeO-) polybrominated diphenyl ethers (PBDE) are suspected endocrine disruptors. Little is known about the accumulation or sources of these chemicals in tissues of humans, particularly those residing in Hong Kong, which is one of the most densely populated cities in the world. Seven MeO-BDEs, fifteen OH-BDEs and three bromophenols (BRPs) were analyzed in blood plasma of 116 humans that had been collected by the Hong Kong Red Cross. Total concentrations of MeO-BDEs, OH-BDEs and BRPs ranged from 3.8×10^2 to 52×10^3 pg g⁻¹ lipid (median 4.5×10^3 pg g⁻¹), 5.3 to 4.9×10^2 pg g⁻¹ lipid (81 pg g⁻¹) and ND to 1.1×10^2 pg g⁻¹ lipid (3.7 pg g⁻¹), respectively. 3-MeO-BDE-47, 6-OH-BDE-47 and 2,4,5-TBP were the predominant MeO-BDEs, OH-BDEs and BRPs, respectively. These results are consistent with accumulation of MeO-BDEs, OH-BDEs and BRPs in human plasma being primarily from natural products and inter-conversion of natural products. Coefficients of determination for some pairs of congeners such as 3-OH-BDE-100 and 6-OH-BDE-47, 6-OH-BDE-85 and 5'-OH-BDE-99, and 2,4-DBP and 6-OH-BDE-85, were near 1.0, which is consistent with them having common sources. Patterns of relative concentrations of the target analytes were similar in the diet, particularly fish, as in blood plasma of humans, which suggests that the diet and particularly seafood might be a source of these compounds and PBDEs. Furthermore, biotransformation of natural chemicals such as OH-BDEs to BRPs might be the primary route of their elimination from humans.

Environment International 47 (2012) 66–72

ESTABLISHING THE GREEN LIPPED MUSSEL *PERNA VIRIDIS* AS A UNIVERSAL MARINE MODEL ORGANISM AND POLLUTION BIOMONITOR FOR ECOTOXICOLOGY AND ENVIRONMENTAL GENOMICS

Kenneth Mei Yee LEUNG

The green-lipped mussel *Perna viridis* is widely used as a sentinel species in marine pollution monitoring and ecotoxicological studies in Asia-Pacific region. This species is considered as a subtropical equivalent biomonitor of the temperate *Mytilus* species. In order to elucidate pollutant effects on this common biomonitor species and provide early warning signals of pollutant-mediated stresses, it is prerequisite and essential to understand the toxic mechanisms at molecular level and identify a suite of reliable exposure- and effect-related biomarkers for diagnostic purpose. Global analyses of the expression levels of genes and their products (i.e., RNA and proteins) are increasingly employed in marine model organisms (e.g., copepods, medaka fish, and mussels *Mytilus* species) to achieve such goals. Despite the important role of *P. viridis* in environmental and toxicological studies, its genomic resources are currently extremely limited when compared with their temperate counterparts, *Mytilus* species. Such an obstacle has significantly hindered the further development of *P. viridis* as a universal model species for ecotoxicological, genomic and proteomic studies. Therefore, this proposed study we will first establish a comprehensive and representative putative transcriptome database for *P. viridis* using the next generation sequencing technology. With the help of bioinformatics, the outcomes of this study will significantly improve our genomic knowledge of *P. viridis*. The analysis will cover three main organs, i.e., hepatopancreas, adductor muscle and gills, which are commonly used in biomonitoring. The established tissue-specific transcript databases will provide a novel and important “back-bone” resource for genome-wide association studies of *P. viridis* which will enable us to uncover the toxic mechanisms, establish concentration-dependent biomarker responses, and develop advanced pollution monitoring tools. Secondly this proposed study will further address the molecular toxic mechanisms of selected trace metals (e.g. cadmium and copper), persistent organic pollutants (e.g. triphenyltin and PBDE) and nano-particles (e.g. nano metal oxides and carbon nanotubes) in *P. viridis* upon waterborne and/or dietary exposure. The results of this study will also help us to underpin the toxic response pathways for various groups of pollutants, from which we will identify a suite of reliable biomarkers for further development of gene-based biosensors and ELISA-based protein-arrays. These advanced diagnostic tools can be used to provide rapid effect-based biomonitoring of marine pollution and screening for new chemicals in the near future. Given the popularity and increasing importance of *P. viridis*, this

work will make significant visible impact and contribution to the advancement of marine pollution research in the region.

A. Project Objectives

1. To establish transcriptome databases for three target tissues, i.e., hepatopancreas, gill and adductor muscle of *P. viridis* using the next generation sequencing technology;
2. To reveal the chemical-specific toxic mechanisms by quantifying the differentially expressed genes of the three target tissues of *P. viridis* upon exposure to selected pollutants including trace metals, persistent organic pollutants and engineered nano-materials; and
3. To identify a suite of reliable exposure- and effect-related biomarkers for pollution monitoring purposes and for the future development of gene-based biosensors and ELISA-based protein arrays as advanced biomonitoring tools.

B. Research Activities

1. Scope of investigation undertaken

Since there was some problem in setting up an account at HKU, this inevitably caused a delay in the commencement of the project. The project was actually started in summer 2012. Research activities conducted in the 1st year (summer 2011-Oct 2012) focused on Task 1 of the project - to develop a *de novo* transcriptome sequencing profile for each of the three target tissues, i.e., hepatopancreas (hp), adductor muscle (ad) and gills (g) of *P. viridis*.

2. Research conducted.

In order to obtain a wide spectrum of environmentally associated transcripts, the *P. viridis* subjected to RNA sequencing covered samples collected from the field (field-type) and from exposure experiments (exposed-type) with various challenges of physical and chemical stresses. Adults (4.0-5.0 cm shell length) mussels were collected from Hong Kong waters using SCUBA diving. The field-type samples were collected from western (Butterfly Beach), southern (Po Toi) and eastern waters (Tung Lung Island and Sam Mun Tsai), with different hydrography. Exposed-type samples were collected from a fish farm in Yung Shue O mariculture zone located at Sai Kung, Hong Kong and were acclimated under running seawater for 7 days and fed with the diatom, *Thalassiosira pseudonana*, prior to the exposure experiments. The mussels were then acutely exposed to different model chemicals (e.g., metals, endocrine disruptors, organic pollutant and engineered nano-particles) and to different physical conditions (e.g., thermal and salinity regimes, and scenarios for hypoxia and ocean acidification) so as to cover a wide range of chemical and physical conditions related transcription patterns which are essential for future environmental studies.

Sublethal concentrations were applied for chemical exposure, and the concentrations were chosen according to the corresponding EC50 concentration or 10% of LC50 concentration of the available data for *P. viridis* or marine bivalves from the USEPA's ECOTOX database (<http://cfpub.epa.gov/ecotox/>) and literature search engines such as the ISI Web of Science. In the exposure experiment, mussels were treated chemically with (1) metals: CdCl₂ (730 µg/L) and CuSO₄ (65 µg/L); (2) endocrine disruptors: triphenyltin chloride (TPTCl; 0.32 µg/L) and dichlorodiphenyltrichloroethane (DDT; 40 µg/L); (3) organic pollutants: polybrominated diphenyl ether 47 (PBDE 47; 5 µg/L) and Benzo[a]pyrene (75 µg/L); and (4) engineered nano particle: nZnO (13 mg/L). For nZnO treatment, the concentration of Zn ion was estimated from the calculated dissolution rate of nZnO. In addition, the mussels were physically exposed to a range of (1) temperatures: 10, 15, 20 and 30°C; (2) salinities: 10, 20 and 30‰; (3) dissolved oxygen (DO): DO 2.5 and 4.0 mg/L; and (4) pH: 7.5, 7.8 and 8.1. The mussels were kept in the testing solutions / exposed conditions at a density of one mussel per 250 mL of 0.2µm filtered artificial seawater (FASW, salinity: 30 ±0.5‰; temperature: 25 ±0.5°C; pH 8.0 ±0.1), except for those used in thermal and salinity exposures. Organic compounds including TPTCl, DDT, PBDE47 and Benzo[a]pyrene were delivered in the FASW using dimethylsulfoxide (DMSO) as solvent at a final concentration of 0.01% (v:v).

Four (2 males and 2 female) mussels from each field site and from each treatment were chosen for obtaining the three target tissues, i.e., hepatopancreas (hp), gill (g) and adductor muscle (ad). The total RNA was extracted using an RNeasy Mini Kit (QIAGEN). The RNA quantity was measured using a NanoDrop 2000 spectrophotometer (Thermo Scientific) while the quality was validated using a bioanalyser (Agilent Technologies). Each tissue of male and female mussels with the pooled total RNA samples from the field and treatments aforementioned were subjected to cDNA library construction using TruSeq RNA Sample Prep Kit (Illumina), and further validated by qPCR and the bioanalyser for their quantity and quality, respectively. The samples were then subjected for RNA-sequencing at the Centre for Genomic Services, The University of Hong Kong, where de novo transcriptome sequencing was employed using a Solexa GAIIx (Illumina).

3. Results achieved

In total, 6 cDNA libraries were constructed for the mRNA of the two sexes and the three target organs of *P. viridis*, i.e., male-hp; female-hp; male-g; female-g; male-ad and female-ad, which made up 6 individual lanes for Solexa sequencing. A PhiX control lane was also included in the run. In the meantime, the de novo transcriptome sequencing was undertaken, and an estimate of 40-60 Gb of data generated after the sequencing run. Sequence data was obtained in mid-December 2012, followed by one and a half month of bioinformatics analysis. After data analysis, we anticipate that a manuscript can be prepared on Task 1 by mid-2013.

Moreover, the PI and other SKLMP members (i.e., Dr. Michael Lam and Prof. John Giesy) have continued their joint research effort on studying the impacts of a common pollutant, triphenyltin to marine organisms. This resulted in two ISI publications appearing in *Chemosphere* 2012.

4. Problems encountered

Since the available amount of RNA extracted could be tissue specific, we encountered problems in extracting enough RNA from the adductor muscles when we followed the standard protocol from the extraction kit. Attempts were then made to optimize the RNA amount being extracted from the adductor muscles. After a number of trials, we discovered a way to obtain a better extraction result for this tissue by modifying the extraction step in which we added 10% more Dithiothreitol (DTT) and a double volume of lysis buffer.

We also faced a problem in obtaining the RNA integrity (RIN) values from the quality validation using the bioanalyser of which the default settings were originally based on RNA of mammals or vertebrates. It is found that the peak of 28S rRNA was relatively lower in *P. viridis*, which required additional adjustment for “the set-point of the anomaly threshold” to > 1 in order to obtain the RIN value. Detections of a low peak of/absence of 28S have also been commonly reported in other molluscs, and this could be associated with a fragment breaking point in the 28S rRNA structure that gives two fragments migrating at the same size as the 18S rRNA during the gel electrophoresis (Barcia et al. 1997, Muttray et al. 2008). We are currently trying to overcome this measurement problem.

5. Collaborative activities

We have consulted the Co-I Prof. Chris Wong and his colleague Dr. J.W. Qiu of the Hong Kong Baptist University regarding the experimental design and method used in this study. As expected, more joint efforts will need to be made in the forthcoming bioinformatics analyses. Also, more involvements of Prof. Paul Lam and Dr. Leo Chan are expected in the second phase of this project (i.e., Objectives 2 and 3).

6. Deviations from the original plan

In terms of the progress, according to the plan in the proposal, the 1st and 2nd year of the project will focus on Task 1, and the current schedule is generally good without much deviation from the original plan.

In terms of research activity, the sampling methods, experimental setup and sequencing protocols in general follow the original plan, except that the originally planned 96-hr exposure experiments have been changed to 24-hr exposure ones in order to obtain an earlier response of the mRNA of the tested conditions for the mussels. In addition, the service vendor for the NGS sequencing

equipment has been changed from BGI Hong Kong Company Limited to the Centre for Genomic Services at HKU so as to better facilitate HKU internal service-based collaboration.

C. Peer-reviewed Journal Publications Arising from this Research Project

ACUTE AND CHRONIC TOXICITIES OF IRGAROL ALONE AND IN COMBINATION WITH COPPER TO THE MARINE COPEPOD *TIGRIOPUS JAPONICUS*

V.W.W. Bao, K.M.Y. Leung, G.C.S. Lui and M.H.W. Lam

Irgarol 1051 has been widely used as a booster biocide in combination with copper (Cu) in antifouling paints. The combined toxicity of Irgarol with Cu on marine organisms, however, has not been fully investigated. This study investigated the acute and chronic toxicities of binary mixtures of Irgarol and CuSO₄ to the marine copepod *Tigriopus japonicus*. The acute combined toxicity of Irgarol and Cu was simple additive as revealed by two response surface models and their contours. However, based on chronic full life-cycle tests, when Irgarol was combined with Cu at an environmentally realistic concentration (10 µg/L), a slightly synergistic effect was observed at a high Irgarol concentration (940 µg/L), as shown by a significant increase in larval mortality. As Cu contamination is widespread in coastal environments, our results entail the importance of considering the combined toxic effect of the booster biocide and Cu for setting ecologically realistic water quality criteria.

Chemosphere 90 (2013) 1140–1148

REVIEW OF MEASURED CONCENTRATIONS OF TRIPHENYLTIN COMPOUNDS IN MARINE ECOSYSTEMS AND META-ANALYSIS OF THEIR RISKS TO HUMANS AND THE ENVIRONMENT

A.Xi. Yi, K.M.Y. Leung, M.H.W. Lam, J.S. Lee, and J.P. Giesy

The state of scientific knowledge regarding analytical methods, environmental fate, ecotoxicity and ecological risk of triphenyltin (TPT) compounds in marine ecosystems as well as their exposure and health hazard to humans was reviewed. Since the 1960s, TPT compounds have been commonly applied as biocides for diverse industrial and agricultural purposes. For instance, they are used as active ingredients in antifouling systems on marine vessels and mariculture facilities, and as fungicides in agriculture. Due to their intensive use, contamination of coastal waters by TPT and its products of transformation has become a worldwide problem. The proportion of quantified TPT to total phenyltin compounds in the marine environment provides evidence that TPT is photodegradable in water and sediment but resistant to

biotransformation. Concentrations of TPT in marine biota are consistently greater than concentrations in water and sediment, which implies potential of TPT to bioaccumulate. TPT is toxic to both marine plants and animals. The predicted no effect concentration (PNEC) for TPT, as determined by use of the species sensitivity distribution approach, is 0.64 ng/L. In some parts of the world, concentrations of TPT in seawater exceed the PNEC, indicating that TPT can pose risks to marine life. Although there is negligible risk of TPT to average human consumers, TPT has been detected in blood of Finnish people and the concentration was greater in fishermen who ate more seafood. It is, therefore, advocated to initiate regular monitoring of TPT in blood and breast milk of populations that consume greater amounts of seafood.

Chemosphere 89 (2012) 1015–1025

DEVELOPMENT OF THECA SPECIFIC ANTISERA FOR THE PROFILING OF CELL SURFACE PROTEINS IN THE MARINE TOXIC DINOFLAGELLATE GENUS ALEXANDRIUM HALIM.

L.L. Chan, X.M. Li, W.H. Sit, P.K.S. Lam and K.M.Y. Leung

Revealing the profile of dinoflagellate cell surface proteins (CSPs) is a crucial step for developing molecular probes for effective identification, separation and enumeration of toxic and non-toxic dinoflagellates. This study aimed to develop theca-specific antibodies against the dinoflagellates *Alexandrium affine* (non-toxic) and *A. tamarense* (toxic) to distinguish the two species, and verify if these antibodies would enable the analysis of multiple CSPs for probing phytoplankton's nutrient physiology, and facilitating rapid detection and enumeration of these harmful algal species. Using 2-DE immunoblots, we evaluated the specificity and effectiveness of the theca-specific polyclonal antisera against two types of antigens generated from fresh or fixed whole cell and insoluble cellular fractions, respectively. Our results showed that, of the four cell surface antigens, paraformaldehyde fixed whole cell antigen derived antiserum specifically recognized weakly bound theca-associated CSPs in toxic *Alexandrium* strain. Using the optimized theca-specific antisera, about 187 and 110 cell surface associated antigenic spots were identified on the 2-DE immunoblots of *A. affine* and *A. tamarense*, respectively. This immunoproteomic approach is proven to be very useful for phytoplankton CSP studies, permitting a more in-depth elucidation of the relationship among nutrient condition, bloom dynamic and toxin production of the harmful algae in the marine environment.

Harmful Algae 16 (2012) 58–62

D. Recognized international conference in which paper related to this research project was delivered

ISOFORM-SPECIFIC RESPONSES OF METALLOTHIONEINS IN THE MARINE POLLUTION BIOMONITOR *PERNA VIRIDIS* CORRESPONDING TO TWO DIFFERENT MODES OF TOXIC ACTION

P.T.Y. Leung, K.M.Y. Leung, T.J. Park and Yu Wang

Metallothioneins (MTs) are small metal-binding proteins that are involved in essential metals homeostasis, toxic metals detoxification and antioxidant defenses. The high metal affinity character of MTs makes them to be a popular biomarker in marine biomonitors (e.g. mussels) for assessing bioavailable metal pollution in the field. With aid of proteomic approach, this study identified and characterized various MT isoforms in hepatopancreas of the green-lipped mussel *Perna viridis* after waterborne exposure to cadmium (Cd) and hydrogen peroxide (H₂O₂), respectively. The mussels were exposed to the control (seawater only), cadmium (0.5 ppm; CdCl₂) and hydrogen peroxide (2.0 ppm; H₂O₂), respectively for 14 days followed by 3 days of depuration. A faster response was detected in Cd-treated mussels than H₂O₂-treat ones in terms of global MTs expression. Such MT expressions reached to a peak level at 14-day of exposure and returned to a basal level after 3-day of depuration for both treatments. Two-dimensional gel electrophoresis coupled with western blotting resolved at least 12 isoform spots of MT from all groups. Further complementary studies were conducted using genomic and de novo sequencing analyses. The combined genomic and proteomic results revealed that these MT isoforms can be grouped into two types: MT-I and MT-II, whereas MT-I is a previously described form and MT-II is a novel type. The two types differ in primary structural characteristics, with 73 amino acids in the deduced protein sequences for MT-I and 72 for MT-II isoforms. Both types of MT isoforms consist of 21 cysteines with 9 Cys-X-Cys motifs, except for one of the sub-types in MT-I which has 20 cysteines only. Temporal differential gene expression was also reported for these two MT types upon the two different chemical exposures. Based on their structures, MT-I is more readily to be oxidized than MT-II. Importantly, our findings indicated that both MT-I and MT-II could be induced by Cd treatment, while only MT-I responded to H₂O₂ exposure. These results suggest a potential application of the two types of MT isoforms in *P. viridis* for monitoring marine pollution and biological effects of chemical pollutants.

E. Other Impact

The PI, Dr. Kenneth Leung has been recently conferred with the VTC 30th Anniversary Outstanding Alumni Award, Vocational Training Council of Hong Kong as recognition for his professional achievements in environmental education and research as well as community services in both local and international communities.

INTERACTIVE EFFECTS OF CLIMATE CHANGE AND HYPOXIA ON FISH SEX DETERMINATION: ESTROGEN SYNTHESIS AND MASCULINISATION

Richard Yuen Chong KONG

Episodes of aquatic hypoxia ($< 2 \text{ mg O}_2 \text{ L}^{-1}$), along with elevated water temperatures are likely to be exacerbated as climate change progresses. Recent studies by our group have demonstrated for the first time that hypoxia disrupts fish sex differentiation, leading to a male-biased sex ratio in the zebrafish and Japanese medaka. Coincidentally, in many fish species exposure to elevated water temperature also leads to male-biased sex ratios. Under climate change, increased frequency of hypoxic episodes and warmer waters are likely to intensify such sex change events. Such shifts in the operational sex ratio are likely to have dire consequences for reproduction and recruitment of fish assemblages inhabiting small lentic waterbodies, potentially leading to losses in biodiversity and fisheries productivity. Despite this emerging threat, the question of how hypoxia and elevated temperature interactively alter fish sex differentiation and hence sex ratios remains unexplored.

One of the mechanisms known to disrupt sex differentiation is interference with sex hormone synthesis. Recent findings in our lab suggest that the inhibition of estrogen synthesis might be a crucial cause of hypoxia-induced masculinisation (the development of male sexual characteristics in a genotypic female). Curiously, inhibition of estrogen synthesis has also been widely observed in masculinised fish as a result of heat treatment, indicating both hypoxia and elevated temperature may share common mechanisms of action on sex differentiation.

Ovarian aromatase (cyp19a) is the steroidogenic enzyme that converts androgens to estrogens. Decreases in cyp19a gene expression can be a major cause of reduced estrogen synthesis under hypoxic and heat conditions, however, the mechanism underlying this gene suppression remains elusive. The transcription factor hypoxia inducible factor-1 (HIF-1) is the master regulator of a broad range of genes responsible for oxygen homeostasis. Intriguingly, HIF-1-mediated gene expression is upregulated not only by hypoxia but also high temperature, implying its critical role in adaptive responses to both stresses (including repression of energy-consuming reproductive processes). As suggested by a recent study using mammalian cancer cells, a possible mechanism of how HIF-1 suppresses cyp19a gene expression could be via depletion of the estrogen receptor (ER), a key transcriptional activator of cyp19a, although this possibility has not been tested in vivo or in fish thus far. Through understanding the responses and effects of hypoxia-and heat-induced HIF-1 on cyp19a gene expression and estrogen

synthesis, this study will establish a novel molecular link between climate change and altered sex ratio in fish populations.

A. Project Objectives

1. To evaluate the interactive effects of combined hypoxia and elevated temperature on altering estrogen synthesis and hence phenotypic sex ratios in fish; and
2. To investigate the functional role of HIF-1 in disrupting estrogen synthesis in relation to *cyp19a* gene regulation.

B. Research Activities

The research activities in this report are focused on preliminary experiments to investigate the general effects of hypoxia on the activation of the HIF-1 signalling pathway, *zlep-a* (zebrafish leptin gene) and seven steroidogenic enzyme genes in zebrafish embryos. Previous studies by other researchers have indicated that hypoxia affects expression of the leptin (Cascio et al., 2008) and ER α gene (Cho et al., 2005), both of which are reported to play important regulatory roles in reproductive functions. Importantly, there is recent evidence indicating that there is cellular and molecular crosstalk between leptin and ER α in the regulation of endocrine function (Fusco et al., 2010). In this first progress report, we provide details on the effects of hypoxia and *zlep-a* on steroidogenic genes and estrogen and testosterone levels in zebrafish.

1. Effects of hypoxia on expression of hypoxia marker genes and *zlep-a* in zebrafish

To confirm that the HIF-1 signaling pathway is activated in zebrafish under hypoxia, zebrafish embryos (at 24 hpf) were exposed to hypoxia (1.0 ± 0.2 mg O₂ L⁻¹) and CoCl₂ (10 mM) until 72 hpf, and real-time PCR measurement of the hypoxia marker genes, *igfb-1* and *vegf-a* was performed. The results showed that the expression of *igfbp-1* and *vegf-a* was increased by approximately 4- and 3-fold, respectively, in hypoxic and CoCl₂-treated embryos. The results confirmed that the HIF-1 pathway is activated in zebrafish embryos by both hypoxia and CoCl₂. Interestingly, the *zlep-a* gene which was previously identified as a HIF1-inducible gene in zebrafish (Chu et al., 2010), was found to be induced by 7- and 2-fold by hypoxia and CoCl₂, respectively.

2. Effects of hypoxia, CoCl₂ and *zlep-a* expression on steroidogenic enzyme genes

Following exposure of fertilized embryos (24 hpf) to hypoxia for 72 hpf, *cyp11a*, *cyp17*, and 3β -*hsd* and *cyp19a* expression was down-regulated by 2-fold; the expression of the other steroidogenic genes, *star*, 17β -*hsd* and *cyp19b*, was unaffected.

To determine the effect of *zlep-a* (zebrafish leptin) expression on steroidogenic gene expression, 1-2-cell stage zebrafish embryos were injected with *zlep-a* MO (knockdown) or *zlep-a* mRNA (overexpression) and reared under hypoxic or normoxic conditions, respectively, until 72 hpf. Embryos were injected with a standard control MO or Hanks buffer as injection controls for knockdown and overexpression, respectively. The relative expression levels of *star*, *cyp11a*, *cyp17*, *3 β -hsd*, *17 β -hsd*, *cyp19a* and *cyp19b* were quantified by real-time PCR followed by normalization to β -actin expression. Five replicates ($n = 5$) of 60 pooled embryos each were used. The data are presented as the mean relative fold change \pm SD with respect to the gene expression level in the normoxic control.

Knockdown and overexpression of *zlep-a* (by microinjection of anti-sense morpholinos and in vitro transcribed mRNA, respectively) selectively affected the expression of *cyp11a*, *3 β -hsd* and *cyp19a* mRNA but not the expression of the other steroidogenic genes. Upon *zlep-a* knockdown in hypoxic embryos, the expression of both *cyp11a* and *3 β -hsd* was decreased to normoxic levels, while *cyp19a* expression was restored to normoxic levels. In contrast, *zlep-a* overexpression in normoxic embryos resulted in a 1.7-fold up-regulation in the expression of both *cyp11a* and *3 β -hsd* and a 2-fold down-regulation of *cyp19a* expression. Altogether, the results suggest that elevated *zlep-a* expression stimulates *cyp11a* and *3 β -hsd* expression, but inhibits *cyp19a* expression in hypoxic zebrafish embryos, whereas the hypoxic up-regulation of *cyp17* seems to not be dependent on *zlep-a* expression.

CoCl₂-treated embryos exhibited a partially overlapping pattern of expression compared to that obtained under hypoxia; *cyp11a* and *3 β -hsd* expression was up-regulated by 1.8- and 1.6-fold, respectively, while *cyp19a* expression was down-regulated by 2.3-fold; *star* and *cyp19b* expression remained unaffected. In contrast to the hypoxic embryos (in which expression of *cyp17* and *17 β -hsd* were up-regulated and unaffected, respectively), the expression of both *cyp17* and *17 β -hsd* was down-regulated by 1.5-fold in CoCl₂-treated embryos.

3. Effects of hypoxia, CoCl₂ and *zlep-a* expression on E2 and T levels.

Exposure of whole embryos to hypoxia resulted in a 1.8-fold reduction and 1.4-fold increase in E2 and T concentrations when compared to their normoxic counterparts. This resulted in a 2.6-fold increase in the T/E2 ratio [from 0.7 ± 0.04 (normoxia) to 1.8 ± 0.37 (hypoxia)]. Interestingly, knockdown of *zlep-a* in hypoxic embryos restored both E2 and T levels and their ratio to that observed for the normoxic control. In contrast, *zlep-a* overexpression under normoxia resulted in concentrations of E2 and T that were 1.3-fold less, and 1.2-fold greater than the controls, respectively. However, the difference for T was not statistically significant ($p = 0.062$). These changes resulted in a 1.5-fold greater T/E2 ratio compared to the normoxic control. Overall, these findings suggest that greater expression of *zlep-a* under hypoxia results in less production of E2 but more production of T, which results in the greater T/E2 ratio observed.

CoCl₂-treated embryos displayed a slightly different pattern from that observed for hypoxic embryos; E2 and T concentrations were 1.7- and 1.4-fold, respectively, less than that of the controls and the T/E2 ratio was unaffected. These results might indicate that hypoxia and CoCl₂ modulate E2 and T synthesis in slightly different ways.

Overall, the results reported here support a causative relationship between elevated leptin gene expression and inhibition of E2 synthesis under hypoxic conditions. Since the changes in E2 concentrations occurred concomitantly with the changes in *cyp19a* expression, it is possible that the inhibition of E2 synthesis during hypoxia is associated with leptin-dependent inhibition of *cyp19a* expression. Likewise, the effects of leptin on T levels were comparable to those induced by hypoxia; leptin appeared to increase T levels, which were accompanied by the upregulation of *cyp11a* and *3β-hsd*.

C. Peer-reviewed Journal Publication Arising from this Research Project

PREVALENCE AND DIVERSITY OF NOROVIRUS GENOGROUPS I AND II IN HONG KONG MARINE WATERS AND DETECTION BY REAL-TIME PCR.

N. Yang, H. Qi, M.M.L. Wong, R.S.S. Wu and R.Y.C. Kong

Marine waters from six sites around Hong Kong with varying levels of sewage pollution were examined for noroviruses (NoVs) by PCR cloning and sequencing of a highly-variable N-terminal region of the VP1 capsid gene, at the ORF1-ORF2 junction of NoV. Phylogenetic analysis of genogroups GI- and GII-specific PCR clones obtained from different marine sites indicated that human NoV GI.1 and GII.4 strains are the most prevalent genotypes circulating in Hong Kong waters. GI- and GII-specific TaqMan-based real-time PCR assays targeting the ORF1-ORF2 junction of NoVs were used to quantify NoV particles in marine water samples in parallel with total *Escherichia coli* counts which were enumerated on TBX medium. No correlation of any significance between NoV and *E. coli* counts was observed which highlighted the inadequacy in using *E. coli* as a fecal indicator to predict the level of NoVs in marine waters to protect public health.

Marine Pollution Bulletin 64 (2012) 164–168

SOURCES AND BIOACCUMULATION OF METALS IN THE PEARL RIVER ESTUARY (PRE) AND HONG KONG COASTAL WATERS

Xiangdong Li

Toxic metals are among the major contaminants in marine ecosystems. Metal contamination has become a major environmental problem in many parts of the world. In the Pearl River Estuary (PRE) and Hong Kong coastal water, two metals are of major concern in the marine environments, namely mercury and cadmium. It is well known that mercury (especially its organic form: methylmercury) is biomagnified at the top of marine food chains (such as in marine fish). Recent evidence has also shown that cadmium can be biomagnified in marine benthic food chains (e.g., intertidal rocky shores). The biomagnification of these two metals in predatory marine fish and gastropods can present significant health risks to human through seafood consumption in Hong Kong and the South China coastal regions. Although metal pollution has been recognized in Hong Kong for several decades, the understanding of the biological and environmental behaviors of mercury and cadmium still remains very poor. The mechanisms on how different marine organisms handle metals and how such handling affects metal toxicity are extremely challenging research topics. Such complexity is further augmented by the very complicated but unique hydrographic conditions in the subtropical Pearl River Estuary and Hong Kong coastal waters. This proposed research project will aim to study (1) the sources and geochemical behavior of mercury and cadmium in the PRE and Hong Kong waters; (2) the biological fates and food web dynamics of mercury and cadmium in the subtropical region; and (3) the bioaccumulation of mercury and cadmium in the top predators of these food chains under different hydrographical conditions/exposure histories, and the assessment of seafood safety issues in the study area. The proposed research will increase our understanding of mercury and cadmium pollution in coastal environments, and will provide scientific advice to regulatory agencies and industries for better management of toxic metals in the marine environment.

A. Project Objectives

1. To study the major sources and geochemical cycling of metal pollutants in the Pearl River Estuary and Hong Kong coastal waters;
2. To investigate the bioaccumulation of metals in marine biota in the subtropical region; and
3. To assess the environmental exposure and potential toxicity of metal pollutants through seafood consumption in the local population.

B. Research Activities

One field sampling for water and sediment was conducted in the Pearl River Estuary in order to assess changes in the metal contamination status over the last 10-15 years and the current concentration of residual antibiotics and antibiotic resistance genes (ARGs) in the water environment. Method development of the Hg isotopic analysis using MC-ICP-MS at HKU are currently under way in order to plan further investigations on the Hg accumulation and geochemical cycling in subtropical marine systems.

Exposure to one metal might have significant effects on the bioaccumulation of other metals. In the present study, we therefore examined the possible effects of Zn exposure on the bioaccumulation of Cd and Cu in three populations of the oysters *Crassostrea hongkongensis*. We found that Zn exposure significantly enhanced the tissue concentrations of Cd and Cu in all populations, and the tissue concentrations of Cd and Cu were highly and positively related to the tissue Zn concentration. Furthermore, the enhanced bioaccumulation of Cd and Cu resulted mainly from their increasing accumulation and distribution in two subcellular fractions (i.e., metallothionein-like proteins and metal-rich granules). Tissue concentrations of Cd and Cu in the natural Zn-contaminated oysters also co-varied with tissue Zn concentration, and prediction analyses revealed that Zn exposure was a dominant contributor to Cd and Cu concentrations. Therefore, we concluded that the increased Zn bioavailability in ambient waters not only increased the tissue Zn concentration but also enhanced the overall bioaccumulation of Cd and Cu. This study strongly demonstrates that contamination of metals in oysters may result from concurrent exposure to other metals. Thus, environmental managers should consider the possible exposure to other metals such as Zn in order to reduce tissue concentrations of toxic metals in oysters.

C. Peer-reviewed Journal Publication Arising from this Research Project

THE CHANGES IN TRACE METAL CONTAMINATION OVER THE LAST DECADE IN SURFACE SEDIMENTS OF THE PEARL RIVER ESTUARY, SOUTH CHINA

B.W. Chen, X.M. Liang, W.H. Xu, X.P. Huang and X.D. Li

Surface sediments can provide useful information on the recent pollution status of an estuary. One recent field survey was carried out in the Pearl River Estuary (PRE), South China in 2011. The comparisons with previous surveys demonstrated that the concentrations of Ni and Pb in the PRE declined over the last decade, but the concentration of Cu increased in the same time frame. The significant decreases in the concentrations of Ni and Pb were probably due to a reduction of anthropogenic inputs, such as industrial wastewater, into the PRE environment, and the ban

imposed on leaded gasoline. Statistical analyses have consistently demonstrated that the process of the sedimentation of fine particles was the dominant factor in controlling the transport and distribution of trace metals in the PRE. The riverine trace metals generally displayed a pattern of diffusion from the northwest to the southeast in the estuary. However, the riparian industrial activities at the east bank of the inner PRE caused significant metal contamination in sediments. In general, effective pollution control measures in the PRD region have decreased the levels of some trace metals in the entire PRE over the last decade with the exception of Cu.

Science of the Total Environment 439 (2012) 141–149

D. Recognized international conferences in which papers related to this research project was delivered

DIFFERENTIATING ANTHROPOGENIC IMPACTS ON ARGs USING SUITABLE GENE INDICATORS IN THE PEARL RIVER ESTUARY, SOUTH CHINA

B.W. Chen, X.M. Liang, X.P. Huang, T. Zhang and X.D. Li

Antibiotic resistant genes (ARGs) are increasingly a focus of concern because they are a potential health risk. The Pearl River (PR) and Pearl River Estuary (PRE) show a distinct gradient in anthropogenic impacts from the river, to the estuary, and on to the coast. In this study, two surveys were conducted in the PR and PRE areas during the winter and summer of 2011, respectively. Seven tet genes consisting of efflux pumps (tetA, tetC, and tetH) and ribosomal protection proteins (tetB, tetM, tetO, and tetW) were analyzed using the polymerase chain reaction (PCR) technique. The tet genes, with the exception of tetA and tetH, were widely detected in the PRE environments. The tet genes exhibited a trend of an increase in total concentration and diversity with the degree of anthropogenic impacts from the river to the coast, indicating that riverine input was the main source of ARGs in the region. Significant correlations were observed between tet genes and antibiotic concentrations, as well as among different environmental compartments (water and sediments). The distribution patterns of tet genes were similar between the potential sources of pollution and the highly-impacted sites, but were significantly different between less-impacted sites and highly-impacted ones or pollution sources. The results suggest that ARGs and antibiotics may be released from identical sources, and transported in a similar manner in estuary/coastal environments close to sources of pollution.

THE CHANGES IN TRACE METAL CONTAMINATION OVER THE LAST DECADE IN SURFACE SEDIMENTS OF THE PEARL RIVER ESTUARY, SOUTH CHINA

W.H. Xu, X.D. Li, W. Yan, G. Zhang, J. Li, L. Miao and W.X. Huang

A total of 65 daily aerosol samples were collected in two open cruises covering the whole northern SCS in September 2005 and August 2007, respectively. The concentrations of Cr, Cu, Ni, Zn and Pb in particular matters (PM) of the northern SCS were comparable to the values measured in suburban and background sites of south China. Cu showed relatively high concentrations during the two sampling periods, suggesting the regional sources of Cu pollution in the Pearl River Delta (PRD) area, south China. The calculated enrichment factor (EF) values of Cu, Zn and Pb were generally greater than 10, indicating the strong influences of anthropogenic inputs. As shown by the backward air trajectory analysis, the high concentrations of trace metals in the air during the sampling periods were mainly related to the air mass passing over the neighboring cities or countries around the SCS. Elevated concentrations of Pb and Zn in the daytime of the 2007 summer cruise may indicate the effects of traffic emissions from the nearby cities around the SCS. The relatively high concentrations of trace metals during the northeastern monsoons in 2005 were probably attributed to local emissions and long range atmospheric transport of pollutants by the Asian monsoon.

Academic Meetings/Conferences

學術會議/講座

The State Key Laboratory in Marine Pollution (SKLMP) Second Academic Committee Meeting

海洋污染國家重點實驗室第一屆學術委員會第二次會議

The State Key Laboratory in Marine Pollution (SKLMP) Second Academic Committee Meeting was held on January 7, 2012 at the City University of Hong Kong. The meeting was mainly composed of three parts: (1) the SKLMP Internal Research Fund Program Presentation, (2) the Director's report and (3) the Academic Committee Meeting. More than 20 local and overseas experts and scholars carried out an assessment and evaluation of the work of the previous year, and gave planning and guidance for the next year's work. It was decided to convene the meeting to expand exchanges between and cooperation with members of the research team on a regular basis, and as much as possible to provide research funding for the application. In the meantime, the SKLMP should expand its own area of research to attract more outstanding scholars to join and to provide an opportunity for young scholars, so that we can jointly strive for research funding from Hong Kong and the Mainland China, and actively be involved in major research project applications.

海洋污染國家重點實驗室第一屆學術委員會第二次會議於2012年1月7日在香港城市大學舉行。會議主要由SKLMP項目工作報告，主任工作報告及學術委員會評議三部分組成。來自海內外的20多名專家學者對實驗室上年度的工作進行考核和評估，並對下一年度的工作進行規劃和指導。會議決定定期召開SKLMP會議擴大各成員在研究上的交流與合作，並盡可能多的提供研究經費以供申請。同時SKLMP也要拓展自己獨有的研究領域，吸引更多優秀學者加入，並為年輕學者提供機會，共同爭取香港以及大陸的研究經費，並積極參與重大研究項目申請。



The Xiangjiang Forum Series: Mini-symposium on Benthic Dinoflagellates and Coral Reef Ecology

香江海洋論壇系列之“底棲甲藻及珊瑚礁生態學研討會”

The workshop on Benthic Dinoflagellates and Coral Reef Ecology was held by the SKLMP and the Department of Biology and Chemistry of City University of Hong Kong from Feb 29 to Mar 3, 2012. The workshop comprised a one-day mini-symposium on benthic dinoflagellates and coral reef ecology, a one-day research cruise and a one-day hands-on workshop. The workshop invited scientists and experts in and out of Hong Kong to present research topics and reports and to develop potential collaboration. More than 60 participants from different parties, including government departments, corporations, universities and research institutes took part in this workshop. The workshop achieved the desired effect, had milestone significance and laid a foundation for the continuous organization of the Xiangjiang Marine Forum Series.

The goal of the forum is to create a relaxed environment for academic exchange both in the laboratory and through academic discussion. The forum aims to encourage the questioning of original theory and to promote a democratic academic culture through the expression of different opinions. By investigating the frontiers of scientific research, the forum will foster original ideas and insights and contribute to the development of interdisciplinary collaboration.

研討會由海洋污染國家重點實驗室與香港城市大學生物和化學繫於2012年2月29號至3月3號共同舉辦。內容主要包括底棲甲藻及珊瑚礁生態小型會議，開放式航次調查，及具體實驗操作交流。本研討會邀請了來自海外，台灣以及香港本地的專家學者做專題報告，分享他們在相關領域的研究成果，同時就可能的合作方向進行了深入探討。該研討會吸引了來自官產學研各個方面的六十多名代表參加，研討會取得了預期效果，具有里程碑意義，為今後繼續舉辦香江海洋系列論壇奠定了堅實的基礎。

Workshop on Benthic Dinoflagellates and Coral Reef Ecology
29 February - 3 March 2012

**AC2-1614, Academic Building 2
City University of Hong Kong
Tat Chee Avenue, Kowloon**

The objective of this workshop is to provide a forum for marine ecologists from USA, Taiwan and Hong Kong to exchange state-of-the-art information and techniques related to research on benthic dinoflagellates and coral reef ecology in order to evaluate possible impacts of human activities, including eutrophication and climate change, on local and regional coral communities, and to propose strategies for the conservation, restoration and management of corals in the South China Sea.

The workshop comprises a one-day mini-symposium, one-day research cruise and one-day hands-on workshop. The topics include:

1. Collection, isolation, and identification of benthic dinoflagellates;
2. Genotyping and mass culture of dinoflagellates;
3. Toxin extraction from dinoflagellate cultures and analysis by UPLC or UPLC-MS/MS;
4. Introduction to methods for studying coral ecology and culturing corals;
5. Current issues in coral ecology;
6. Future efforts on research on and conservation of corals.

Enquiries:
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Invited Speakers
Prof. Put ANG
The Chinese University of Hong Kong, Hong Kong
Dr. Tony Tung-Yung FAN
National Museum of Marine Biology and Aquarium, Taiwan
Mr. Shao-Liang HSU
Marine National Park Headquarters, Taiwan
Dr. Dickey LAU
Department of Forestry and Conservation Department
National Sun Yat-sen University, Republic of Chinese Medicine, Taiwan
Dr. Chung-Kuang LU
National Sun Yat-sen University, Republic of Chinese Medicine, Taiwan
Dr. Jian-Wen QIU
Hong Kong Baptist University, Hong Kong
Dr. Mindy RICHLEN
Rutgers University, USA
Dr. Tak-Cheung WAI
The University of Hong Kong, Hong Kong

Registration (Deadline: 24 Feb 2012)	
One-day symposium (29 Feb 2012)	Free
Conference lunch & dinner	HKS 600
Open research cruise (2 March 2012) (including transportation, cruise dining, boating, snorkeling and lunch)	HKS 500 (limited to 30 people)
Social dinner (2 March 2012)	HKS 300
Hands-on workshop (3 March 2012) (by invitation only)	Free (limited to 15 people)

Coorganized by SKLMP

Academic Meeting/Conference

The practical aspect of the forum will promote knowledge innovation and improved laboratory techniques and practices. Collaboration developed during the forum will be directed toward application for major funding opportunities, including projects supported by the SKLMP Seed Collaborative Research Fund and the SKLMP CityU Internal Research Fund.

香江海洋論壇的主旨及目的是鼓勵質疑原有理論，提倡發表不同意見，提出有獨創性的思路與見解，創造寬松的學術交流環境，弘揚學術民主風氣，促進學科交叉與融合，推進整體綜合性研究，提高實驗室學術水準，挖掘面向重大基金申請的科學問題，確定實驗室種子協作基金(SKLMP Seed Collaborative Research Fund)及城大內部基金(SKLMP CityU Internal Research Fund)的資助項目。



The Summer School on Emerging Environmental Problems and Risk Assessment 2012

2012年暑假學校“新興環境問題與風險評價”



The Summer School on Emerging Environmental Problems and Risk Assessment held on 2-13 July, 2012 was jointly co-organized by the State Key Laboratory in Marine Pollution, the Department of Biology and Chemistry, City University of Hong Kong, the State Key Laboratory of Marine Environmental Science, Xiamen University, the Centre for Marine Environmental Research and Innovative Technology, Area of Excellence (Hong Kong), the State Key Joint Laboratory of Environmental Simulation and Pollution Control, Peking University and San Diego State University.

The Summer School covered five topics: 1) Global Climate Change and Emerging Environmental Problems; 2) Toxicology and Human Health; 3) Atmospheric and Oceanic Sciences; 4) Impacts on the Ecosystem; and 5) Quantitative and Risk Assessment. The courses include lectures by experts and leading scientists, student presentations and group discussion, and a field trip. We expected that through diversification and an interactive teaching mode, students could understand more clearly and deeply the current international environmental hot spots.

香港城市大學海洋污染國家重點實驗室聯合了香港城市大學生物及化學系、廈門大學近海海洋環境科學國家重點實驗室、香港卓越領域-海洋環境研究與創新技術中心、北京大學環境模擬與污染控制國家重點聯合實驗室以及美國聖地亞哥州立大學於2012年7月2-13日在廈門大學舉辦“新興環境問題與風險評價暑期學校”。

暑假學校為一系列課程，今年課程圍繞五個主題：1) 全球氣候變化與新興環境問題；2) 毒理學與人類健康；3) 大氣與海洋科學；4) 人類活動與生態系統相互作用；5) 定量與風險評價進行講學。課程包括有專家學者學術講座、學生報告、學生小組討論、實地考察活動。我們期望透過多元化及互動的教學模式，理論與實際結合，學生更能深刻地理解課題，對當前的國際環境熱點有更深刻的體會。

Academic Exchange and Collaboration 學術交流與合作

Exchange Program Between the City University of Hong Kong and the School of Environmental Engineering, Peking University 北京大學環境科學與工程學院到香港城市大學海洋污染國家 重點實驗室作學生實踐交流活動

In early October, the Spin Kid Project continued with an exchange program between the City University of Hong Kong and the School of Environmental Engineering, Peking University under the theme of "Investigating the conservation of biodiversity through advanced environmental technology". Through this exchange program, teachers and students could share and learn about issues on environmental monitoring and control technologies, and discuss their concerns regarding damage to the marine environment, in order to increase their attention to the importance of environmental protection and conservation. They also experienced scuba diving, thus giving them an opportunity to reflect on the need to protect our marine resources.

學生實踐交流活動於10月3日-10月7日在香港城市大學進行。活動主題為“關注前沿環境科技 共話兩地生態安全”，旨在通過兩校之間教師、學生之間的交流，學習、分享兩地關於環境污染物監測及控制技術，生態系統監測及修復技術等方面的最新進展，討論區域間就生態環境破壞方面的不同關注點及其聯繫，起到增進兩校師生交流，加深兩地學生生態安全問題認識的目的。這次活動包括專家主題講座、小組討論、實驗室參觀、潛水體驗課程，形式多樣，內容豐富，對加深雙方同學於環境保護和生態安全等問題的認識有莫大裨益。



Development of Biosensors for Detecting Marine Pollutants

海洋污染物的生物傳感器研發

The City University of Hong Kong (CityU) and Jiangnan University (JNU) have signed an agreement regarding co-developing biosensors for detecting marine pollutants, in an attempt to accelerate collaboration in marine environmental monitoring technology development based on the strengths of the two institutions. In this project, a systematic ecological survey will be conducted in the estuary of the Pearl River Delta to collect water samples at different seasons. The various pollutants contained in the water samples will be tested using the biosensors, both qualitatively and semi-quantitatively, to validate the feasibility of deploying these biosensors in the marine environment for real-time monitoring of targeted pollutants. During the implementation of this collaborative project, CityU will be responsible for the planning of the whole project and sample collection as well as the analytical measurement of the various pollutant using the advanced equipment in the central laboratory of the SKLMP; while JNU will be in charge of the development and optimization of biosensors, ensuring their reliability and accuracy under laboratory conditions.

The expected outcomes include: i) a better understanding of the geochemistry of various pollutants in the Pearl River estuary via the systematic survey; ii) developing prototypes of the biosensors for different typical pollutants in the aquatic environment; and iii) 2 joint SCI papers will be published after the completion of this project.

江南大學與香港城市大學海洋污染國家重點實驗室本著共同發展、促進內地與香港海洋污染監測技術研發的宗旨，雙方就“海洋污染物的生物傳感器研發”合作項目達成協議，主要內容為針對珠江口和香港海域水體中污染物進行生態調查以及在實驗室條件下應用生物傳感器對污染物進行定性和半定量分析研究，為將來實現海洋污染物的實時原位監控創造條件。“城大”負責對整個項目的實施進行規劃，並承擔樣品採集和污染物的超痕量分析鑒定工作。“江大”負責和承擔生物傳感器的研發和優化；並在實驗室條件下，驗證其可靠性和準確性。

本項目合作內容的預期目標主要包括三個方面：①對珠江口和香港海洋目前普遍存在的污染物進行系統研究。②互派訪問學者2名，以完成環境樣品超痕量分析以及實驗室條件下生物傳感器的開發和性能優化研究；③共同發表論文2篇。

Marine Environmental Monitoring Programme for the Remediation and Development Project at the Estuary Area in the Eastern District of Shantou

汕頭市東部城市經濟帶河口治理及綜合開發項目施工期海洋環境跟蹤監測

Shantou University (STU) and the Research Centre for the Oceans and Human Health (H2O), Shenzhen Research Institute of City University of Hong Kong have jointly launched a project entitled 'Marine Environmental Monitoring Programme for the Remediation and Development Project at the Estuary Area in the Eastern District of Shantou', under a collaboration scheme between the two institutions. Prof Zhong HU from STU and his group will take responsibility for conducting in-situ monitoring and sample collection. As an essential part of this cooperation, Dr Bokun LIN will pay a one-year visit to the H2O Research Centre to conduct a collaborative study under the co-supervision of Dr James LAM and Prof Zhong Hu. It is hoped that the project will consolidate the foundation for future cooperation.

汕頭大學與香港城市大學深圳研究院海洋與人類健康研究中心達成協議，共同承擔“汕頭市東部城市經濟帶河口治理及綜合開發項目施工期海洋環境跟蹤監測”項目的實施工作。汕頭大學胡忠教授及其小組主要負責汕頭東部海域的實地監測和樣品採集工作。在此合作框架下，汕頭大學林伯坤博士將作為訪問學者來香港城市大學進行為期一年的合作研究工作，為雙方進一步聯合開展後續研究奠定基礎。



Background Levels of Typical Pollutants in the Aquatic Environments at Shenzhen and the Technology Development for Pollution Prevention and Control

深圳市環境科學研究院國家環境保護飲用水水源地管理技術重點實驗室與城大H2O合作

“深圳水環境樣品中抗生素檢測方法開發與含量檢測”及
“水環境樣品中抗生素檢測技術人員培訓”

As a milestone of the collaboration between the SKLMP and Shenzhen Academy of Environmental Sciences (SZAES), a cooperative project entitled 'Background levels of typical pollutants in the aquatic environments at Shenzhen and the technology development for pollution prevention and control' has been kicked off after long-term preparation and planning. As an essential part of the genuine collaboration, the SKLMP has begun to transfer state-of-the-arts analytical technology for detecting emerging pollutants to the SZAES. The major objective of the next stage aims to jointly apply for research grants related to the environmental issues arising at the Eastern River and the safety of the drinking water of Shenzhen and Hong Kong, on the basis of the complementary strength of the two institutions. The ultimate goal of the collaboration is to establish jointly within five years a State Key Laboratory through our common efforts. In addition, a smart-lab-management-system initiated by the SKLMP will be introduced and deployed at the new SZAES laboratory, as a demonstration of an excellent and advanced concept of modern laboratory management for universities and research institutes.

合作的第一步將是雙方以“深圳市水環境分布典型污染物水平及防控關鍵技術研究”項目合作為基礎，我方將轉移各類環境監測分析技術給深圳環科院；合作的第二步，雙方將圍繞東江面臨的各類環境問題，利用雙方的優勢聯合申請2013年的科研項目課題，並以改善粵港飲用水質為最終目標；合作的第三步，以深圳環科院為依託單位、5年內聯合申報科技部國家重點實驗室。通過深圳環科院提供實驗室、我方提供技術指導的形式推進實驗室管理智能化系統的實現。通過深圳環科院智能實驗室管理系統的建立可進一步將我們對實驗室管理的先進理念引進各大高校與科研機構，並形成廣泛的影響力，為提高中國的實驗室管理水平盡一份心力。

Review of Major Events

實驗室大事回顧

The Establishment of the Research Centre for the Oceans and Human Health (H2O)

於深圳虛擬大學校園設立“海洋與人類健康研究中心”

On July 19, 2012, the Research Centre for the Oceans and Human Health, referred to as the H2O was approved by the Innovation and Technology Commission of Shenzhen Municipal Government and the City University of Hong Kong to set up an office and laboratory at the City University of Hong Kong, Research Institute Building. The Research Centre for the Oceans and Human Health (H2O) is a satellite institution of the State Key Laboratory in Marine Pollution (SKLMP), City University of Hong Kong (CityU), which has many achievements spearheading the fields of marine environmental research in Hong Kong and Asia Pacific region.

海洋環境的科學研究作為香港城市大學重點扶持發展的優勢研究領域，其不少研究成果在香港乃至亞太地區處於重要地位。2012年7月19日，「海洋與人類健康研究中心（Research Centre for the Oceans and Human Health，簡稱H2O）」獲得了深圳市政府創新科技委員會及香港城市大學的批准正式成立，並於香港城市大學產學研大樓內設立辦公室及實驗室，成為香港城市大學海洋污染國家重點實驗室設立在內地的衛星機構。

研究中心將關注海洋環境的熱點問題，並為海洋污染國家重點實驗室優秀團隊提供與內地高校和科研院所開展學術和技術交流的平台。另外，研究中心也將為表現優異的年輕學者進行有關先進精密儀器分析、海洋管理先進理念和專業技能的培訓，增強其申請外源研究經費的競爭力。



The Research Centre will focus on the hot issues of the marine environment and provide a solid platform for a coordinated and long-term approach for tackling marine pollution problems, and also provide good opportunities for the SKLMP's outstanding research team to strengthen its collaboration with mainland institutions in academic and technological exchanges. In addition, the Centre will assemble and nurture young scientists and provide them with specialized training in the use of modern instrumentation and novel technology to manage the marine environment, and to enhance their competitiveness in attracting research funds.

2012年自研究中心成立以來，已爭取科研項目8項，獲得資金支持為人民幣688萬元。其中，國家自然科學基金項目3項，包括面上基金2項，青年基金1項；以及國家重大科技專項1項和省市級合作項目3項。以及在成立短短幾個月內已經與國內幾所大學（廈門大學（夥伴國家重點實驗室、中國科學院城市環境研究所、北京大學、清華大學、南京大學、汕頭大學、深圳市環境科學研究院）建立了夥伴關係。



The Kick-start Ceremony of the Shenzhen Marine Research and Technology Consortium (SMART)

深圳海洋研究與技術聯盟啓動儀式(SMART)

On 15th November, 2012, a Joint Conference was held in the Hotel Kapok by the Shenzhen Virtual University Park. There were about 140 people who were leaders of member universities, the Ministry of Education, the Ministry of Science and Technology, the Department of Science and Technology of Guangdong Province and the Shenzhen government. The Kick-start Ceremony of the Shenzhen Marine Research and Technology Consortium (SMART) was also held in this conference. Prof Paul K.S. Lam attended this conference as a representative of the City University of Hong Kong.

SMART comprises ten members which are the top research teams in the marine environment, marine minerals and marine biology. The aim of SMART is to achieve research resource sharing and complementary cooperation, to set up public technological service platforms among members, to work together on large research projects and to cultivate high level researchers for Shenzhen marine research.

2012年11月15日下午，深圳虛擬大學園2012聯席會議在深圳灣木棉花酒店隆重舉行。深圳虛擬大學園成員院校領導、首席（駐園）代表140多人出席會議。教育部、科技部、廣東省科技廳、深圳市領導及深圳市相關委辦負責同志和相關媒體代表也參加會議。會上舉行了 深圳海洋研究與技術聯盟 Shenzhen Marine Research and Technology Consortium (SMART) 啓動儀式併發布行動宣言。林群聲教授作為香港城市大學的代表參與了啓動儀式。

SMART集結十個成員院校在海洋環境、海洋礦產、海洋生物等方面的高水平研究機構和科研團隊，旨在通過協同創新，實現資源共享與互補式合作，聯合開展重大項目、課題研究，為深圳海洋經濟構築人才團隊，搭建公共技術平台。



Introduction to the Shenzhen Marine Research and Technology Consortium (SMART)

Targets of cooperation

1. To be a collaborative innovation team which is geared to the needs of the cutting edge of marine science, aiming to be world-class in marine science research. It is a platform which will represent the high level of South China marine research and personnel training by uniting the universities and research institutions and other international well-known academic institutions.
2. To be a South China marine research platform for technology development and transfer which will support the development of strategic emerging marine industries by uniting universities, research institutions and key enterprises of the marine industry. Marine engineering technology would point the direction of this collaborative innovation team.
3. To be an advanced platform which can promote the development of regional innovation by integrating universities, research institutions in a team together with pillar enterprises and the industrialization bases of the marine economy. This collaborative innovation team would be led by local government, and focus on servicing the regional marine economy and the people's livelihoods.

深圳海洋研究與技術聯盟(SMART)簡介

SMART 的合作目標:

1. 以海洋科學為主題，以世界一流為目標，通過團隊成員院校、科研機構以及國際知名學術機構強強聯合，成為代表南方海洋領域科學研究、人才培養的高水平平台。
2. 以海洋工程技術學科為主導，以培育海洋戰略新興產業，通過團隊成員院校、科研機構，與海洋領域相關骨幹企業強強聯合，成為支撐海洋行業產業發展的技術研發和技術轉移南方重要平台。
3. 以地方政府為主導，服務區域海洋經濟和民生領域為重點，通過團隊成員院校、科研機構與支柱重點企業或海洋經濟產業化基地的交叉融合，成為促進地方區域創新發展的前沿平台。



Spotlights 年度焦點

Brominated Flame Retardants Exposure of Residents in the Pearl River Delta

珠三角地區居民的阻燃劑暴露

The concentrations of polybrominated diphenyl ethers (PBDEs) and novel brominated flame retardants (nBFRs) such as methoxylated (MeO-) and hydroxylated (OH-) PBDEs in the environment and humans have been increasing in recent years all over the world. This is particularly the case in the Pearl River Delta (PRD), which is a global power house producing a large amount of electronic products. As important environmental endocrine disruptor chemicals, these organobrominated compounds have the potential to cause serious thyroid hormone disruption, neurotoxicity and adverse developmental effects in humans. However, our knowledge about the intakes, body accumulation and potential transgenerational exposures to these BFRs, especially nBFRs, is very limited.

The major aim of this project is to determine the daily intake of BFRs via food ingestion and dust inhalation and ingestion for residents in the Pearl River Delta, their accumulation in the human body and transgenerational exposure via placental transfer and breast feeding.

近年來環境及人體中的多溴聯苯醚和其他一些新型阻燃劑如甲氧基及羥基多溴聯苯醚引起了越來越多的關注。珠江三角洲地區作為重要的製造業基地生產了大量的電子產品,阻燃劑污染問題對本地區而言尤其重要。作為一類重要的環境內分泌干擾物質,此類有機溴化合物可能導致人類嚴重的甲狀腺激素紊亂,神經毒性以及生殖毒性。但迄今為止關於各類阻燃劑尤其是新型阻燃劑人體攝入,體內蓄積以及可能的代際傳遞的所知還相當有限。

本項目的主要目的在於研究珠三角地區居民通過食用食物及吸入粉塵對阻燃劑的日常攝取,各類阻燃劑在人體內的蓄積以及母嬰間通過臍帶和母乳餵養的代際傳遞行為。



Extraction 提取



Purification 純化



Analysis 分析

Main analytical processes of food and human sample

食物及人體樣品的主要分析過程

by Prof Ming Hung Wong

黃銘洪 教授

Sampling

- A food consumption survey involving 25 pregnant women coming from five PRD cities (Zhuhai, Dongguan, Guangzhou, Foshan, and Zhongshan) was conducted.
- According to the food consumption survey results and food items availability in the markets of the studied cities, a total of 40 food items were selected for further food basket analyses (as shown in the table)
- Twenty five sets of human samples [(1) human milk, (2) blood, (3) placenta, (4) cord, (5) meconium] were also collected from the 25 pregnant women who were involved in the food consumption survey to monitor the body loadings of the above mentioned PBDE levels in the PRDs.

樣品採集

- 對來自珠三角地區五個城市(包括珠海,東莞,廣州,佛山和中山)的25個孕婦進行日常食用食物種類的問卷調查。
- 根據食物食用種類的調查結果及研究城市超市中食物供給情況,採集了共40種食物樣品以供食物暴露調查之用(如下表所示)。
- 各類人體樣本[(1)母乳,(2)血液,(3)胎盤,(4)臍帶,(5)胎糞]採集自食物調查中的25個孕婦以調查各類多溴聯苯醚的珠三角地區人體蓄積水平。

Food Group	Food Item	Zhuhai	Dongguan	Guaungzhou	Foshan	Zhongshan	
Freshwater fish	Crucian carp (<i>Carassius auratus</i>)						
	Grass Carp (<i>Ctenopharyngodon idellus</i>)						
	Common Carp (<i>Cyprinus carpio</i>)	N.A.		N.A.			
	Bighead carp (<i>Hypophthalmichthys nobilis</i>)			N.A.	N.A.		
Marine fish	Sea perch (<i>Lateolabrax japonicus</i>)		N.A.		N.A.	N.A.	
	Large yellow croaker (<i>Pseudosciaena crocea</i>)			N.A.			
Meat	Pork						
	Beef						
	Sausage						
Poultry	Chicken						
	Duck						
Egg	Chicken						
Viscera	Kidney of pig						
	Heart of pig						
	Liver of pig						
Vegetables	Kidney bean (<i>Phaseolus vulgaris</i> L.)					N.A.	
	Eggplant (<i>Solanum melongena</i>)						
	Chilli pepper (<i>Capsicum annuum</i>)						
	Waxgourd (<i>Benincasa hispida</i>)						
	Cucumber (<i>Cucumis sativus</i>)						
	Soybean (<i>Glycine max</i>)	N.A.	N.A.		N.A.		
	Leafy vegetables	Celery (<i>Apium graveolens</i> Linn.)					
		Broccoli (<i>Brassica oleracea</i> var. <i>italica</i>)					
		Cabbage (<i>Brassica oleracea</i>)					
		Flowering Chinese cabbage (<i>Brassica parachinensis</i>)					
Leaf of lettuce (<i>Lactuca sativa</i> L.)							
Root and Stem Vegetables	Chinese kale (<i>Brassica alboglabra</i> L. H. Bailey)						
	Root of lotus (<i>Nelumbo nucifera</i>)						
	Carrot (<i>Daucus carota</i> L. var. <i>sativa</i> DC.)						
Cereal	White radish (<i>Raphanus sativus</i> L.)						
	Rice						
	Rice noodle						
	Noodle						
	Potato (<i>Solanum tuberosum</i>)						
	Sweet Potato (<i>Ipomoea batatas</i>)						
	Corn (<i>Zea mays</i>)						
Fruits	Peanut (<i>Arachis hypogasa</i>)						
	Apple (<i>Malus pumila</i> Mill.)						
	Pear (<i>Pyrus</i> spp.)			N.A.			
	Citrus fruit (<i>Citrus</i> spp.)			N.A.			

Ciguatera

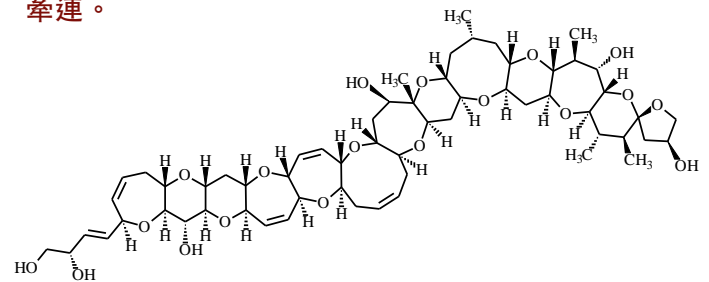
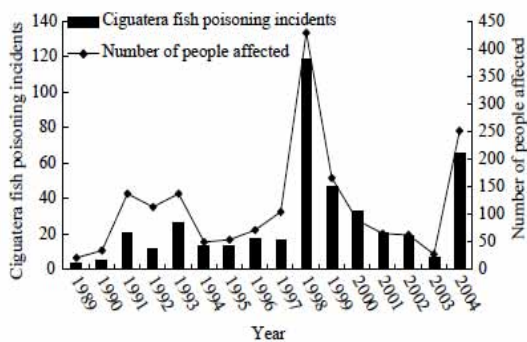
Ciguatera fish poisoning (CFP)

Ciguatera fish poisoning (CFP) is a form of non-bacterial food poisoning caused by consumption of tropical and subtropical fishes contaminated by ciguatoxins (CTXs). The high demand for coral reef fishes has led to the spread of ciguateric fishes over the globe (Dickey and Plakas 2010; Sadovy 1998). Annually, an estimated 10,000 to 50,000 people in the world are affected by CFP (Dickey and Plakas 2010; Lehane and Lewis 2000). In Hong Kong, an average of 26 CFP incidents, affecting more than 100 people, have been recorded since the 1990s (Wong et al. 2005, 2008). Due to the difficulties in diagnosis, the actual number of intoxications may, in fact, be under-reported and more people could be affected by CFP. CFP may also impose economic loss on endemic countries that rely mainly on fisheries, as well as on the seafood markets in non-endemic regions following an outbreak.

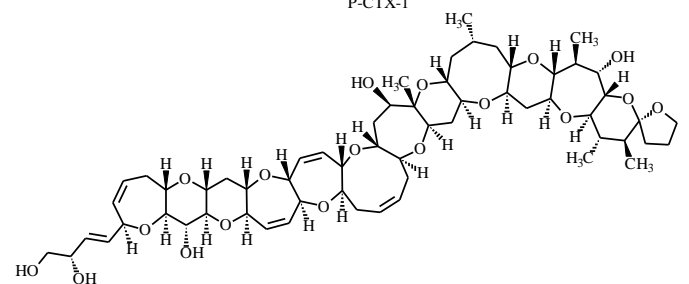
雪卡毒素

雪卡（西加）毒魚類中毒(CFP)引起的非細菌性食物中毒是通過食用含雪卡毒素(CTXs)的熱帶或亞熱帶魚類所導致。人類對食用珊瑚魚的大量需求使得含雪卡毒素的魚類在全球範圍內被廣泛消費(Dickey and Plakas 2010; Sadovy 1998)。世界上每年大約有10000至50000人中中了雪卡毒魚類中毒(Dickey and Plakas 2010; Lehane and Lewis 2000)。根據文獻資料，從1990年至今，香港每年約發生26次雪卡毒魚類中毒事件、中毒人數超100人(Wong et al. 2005, 2008)。由於難以根據病症準確診斷人們的中毒是否因雪卡魚毒素引起，因此實際中毒人數可能遠大於資料記載。另外，雪卡毒魚類中毒也會引起經濟損失，可能會導致以出口漁業為經濟支柱的國家損失慘重，同時也會導致地方漁業進口市場受到極大的牽連。

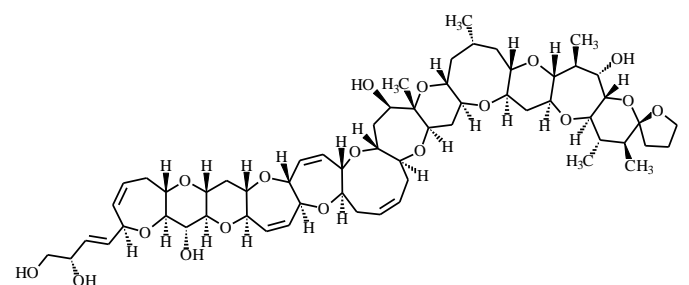
Two prominent outbreaks in 1998 and 2004
more than 100 cases, affecting more than 400 people
(Wong et al., 2005)



P-CTX-1



P-CTX-2



P-CTX-3

Our achievements in CFP research

The development of a ciguateric fish screening method is crucial to safeguard human health and to identify their distribution for better management of fishery resources. In the past three years, the State Key Laboratory in Marine Pollution (SKLMP) has carried out six main tasks in the CFP research, as follows:

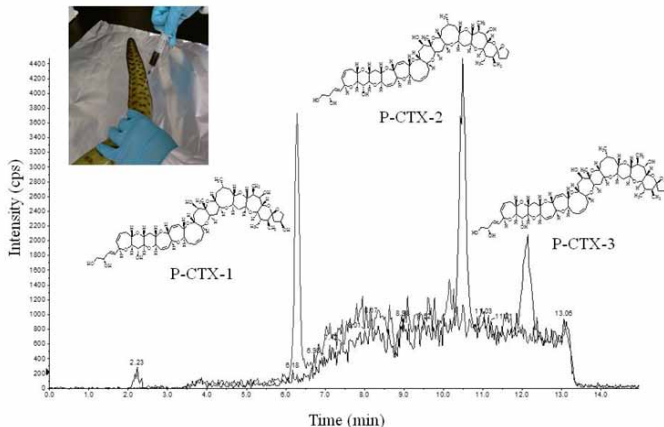
1. Isolation and purification of Pacific-CTX-1 (P-CTX-1) as an authentic standard for this researches;
2. Optimization and validation of analytical methods for quantification of three predominant P-CTXs (i.e. P-CTX-1, P-CTX-2 and P-CTX-3) detected in muscle and whole blood of marketed coral reef fishes collected from the Pacific Ocean;
3. Establishment of the relationship of total ciguatoxicity between whole blood and muscle of moray eel, proving that whole blood can be a representative matrix for the screening;
4. Examination of the spatial distribution of ciguatoxic fishes in one of the atolls of the Republic of Kiribati, where CFP is endemic;
5. Verification and confirmation of the proposed "food web hypothesis" that explains the origin of CTXs and the cause of CFP; and
6. Identification of biomarkers for wild-caught ciguateric coral reef fishes via proteomic analysis.

西加魚毒研究成果：

檢測與篩選含雪卡魚毒素魚類的方法對於保護人類健康以及管理漁業資源至關重要。在過去的三年里，海洋污染國家重點實驗室（SKLMP）承擔了6個主要研究項目，包括：

- (1) 通過對太平洋雪卡毒素的分離與純化，為研究者提供權威的 P-CTX-1 標準品；
- (2) 優化並驗證了三種主要 P-CTXs（如 P-CTX-1，P-CTX-2 和 P-CTX-3）在太平洋珊瑚魚肌肉、血液中的含量分析方法；
- (3) 建立了海鰻血液與其肌肉中雪卡魚毒素含量的關係；
- (4) 對雪卡毒魚類中毒嚴重的基里巴斯共和國，調查了其一個島嶼含雪卡魚毒素魚類的空間分布；
- (5) 證明瞭先前提出的“食物鏈假說”，解釋了 CTXs 的起源和 CFP 的原因；
- (6) 通過蛋白質組學的分析，得到了野外含雪卡魚毒素珊瑚魚的生物標記。

Chromatograph of P-CTXs in whole blood of wild-caught moray eel



Publications (发表的论文)

- Yim Ling Mak, Jia Jun Wu, James C. W. Lam, Wing Hei Chan, Margaret B. Murphy, Leo L. Chan, Paul K. S. Lam, submitted. Simultaneous quantification of three P-CTXs in blood of coral reef fishes using liquid chromatography tandem mass spectrometry. *Analytical and Bioanalytical Chemistry*. (Under Revision)
- Xi-Wen Jiang, Xiaomin Li, Paul Kwan Sing Lam, Shuk Han Cheng, Daniel Schlenk, Yvonne Sadovy de Mitcheson, Ying Li, Ji-Dong Gu, Leo Lai Chan, 2012. Proteomic analysis of hepatic tissue of ciguatoxin (CTX) contaminated coral reef fish *Cephalopholis argus* and moray eel *Gymnothorax undulates*. *Harmful Algae*, 13, 65 – 71.
- Jia Jun Wu, Yim Ling Mak, Margaret B. Murphy, James C. W. Lam, Wing Hei Chan, Mingfu Wang, Leo L. Chan, Paul K. S. Lam, 2011. Validation of an accelerated solvent extraction liquid chromatography-tandem mass spectrometry method for Pacific ciguatoxin-1 in fish flesh and comparison with the mouse neuroblastoma assay. *Analytical Bioanalytical Chemistry*, 400, 3165 – 3175.
- Wing Hei Chan, Yim Ling Mak, Jia Jun Wu, Ling Jin, Wai Hung Sit, James C. W. Lam, Yvonne Sadovy de Mitcheson, Leo Lai Chan, Paul K. S. Lam, Margaret B. Murphy, 2011. Spatial distribution of ciguateric fish in the Republic of Kiribati. *Chemosphere*, 84, 117 – 123.

Contributions to academia/community:

CFP research in the SKLMP has also contributed to academia and the community by providing

1. A purified P-CTX-1 authentic standard to scientists in CityU and Mainland China for in-depth toxicological study;
2. Collaboration with experts in Mainland China for examination of CTX-producing mechanisms of species of the dinoflagellates *Gambierdiscus* spp.;
3. Training of the analytical methods for researchers from the Chinese Government, such as the Xiamen Center for Disease Control and Prevention; and a service to screen suspected ciguatoxic fishes from Tuvalu, the South Pacific Ocean, after a CFP outbreak.

對學術界與社會的貢獻：

SKLMP對雪卡魚毒素的研究對學術界和社會有一定的貢獻。

- (1) 得到的 P-CTX-1 權威純品為香港城市大學甚至是全中國研究雪卡魚毒素的科學家們對該種毒素的深入研究提供了堅實的基礎；
- (2) 與中國研究有毒網比價藻 CTX 產生機制的專家們建立了合作關係；
- (3) 向中國政府相關部門如廈門疾病預防控制中心提供雪卡魚毒素檢測的培訓；
- (4) 為南太平洋島國圖瓦盧在 CFP 爆發後提供檢測疑似含雪卡魚毒素魚類的服務。

by **Prof. Paul Kwan Sing LAM**
林群聲 教授

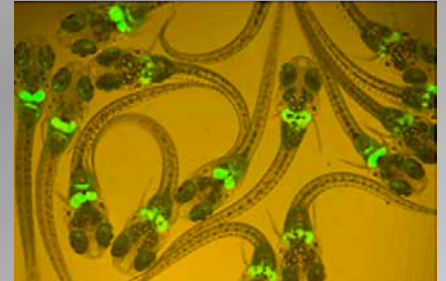
The Long and Winding Road of Patent Application

Transforming knowledge into practice

by Prof. Shuk Han CHENG

Research

Jan 2005 **Project started**
The generation of a transgenic medaka to detect endocrine disruptor started in January 2005 as a PhD project, and built on the previous work on choriogenin gene expression by other members (Yu et al., 2006. Aquatic toxicology).



Jan 2008 **Validation**
The validation of the applicability of this transgenic medaka was performed from January 2008.

Dec 2009 **1st draft of patent (scientists' view)**
The first draft of the patent application was generated by the scientists, with the title "transgenic fish for use in detecting estrogenic compounds".
Invention disclosure submitted to KTO for patent funding supports.

IP protection

Mar 2010 **2nd draft (lawyers' view) and filed**
The core patentable inventions in the first draft were identified and refined with lawyers, and the second draft was filed to with the U.S. Patent and Trademark Office (USPTO).



US Patent No. 12/730,956

Patent examination
A series of "obviousness rejection" notes came along in early 2012, and arguments had to be drafted based on scientific evidence.
The amendment and response to non-final Office Action was drafted in June, 2012.



Nov 2012 **Patent to be granted**
"Notice of Allowance" was received on November 12, 2012 from the U.S. Patent and Trademark Office USPTO.



Commercialization

Beginning of a new story.....

The technology is successfully transferred to Vitargent (International) Biotechnology Ltd.
An SKLMP post doc (Dr Xueping Chen) is the CTO.
Now, to culture CityU graduate entrepreneurs and create jobs.



Vitargent (International) Biotechnology Co. Ltd.

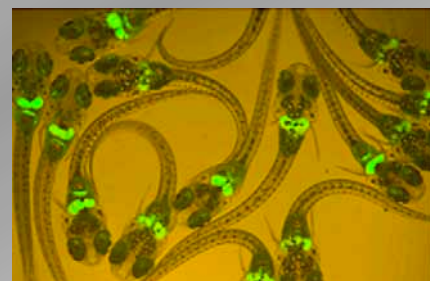
漫長而曲折的專利申請之路 知識向實踐的轉化



2005年
1月

項目啓動

轉基因青鱗魚檢測內分泌干擾物作為博士課題啓動於2005年1月，該項目建立於課題組對魚卵殼蛋白基因表達的研究基礎之上(Yu et al., 2006. Aquatic toxicology)。



2008年
1月

驗證

對該項目應用性的驗證始於2008年1月。

2009年
12月

專利第一稿(研究人員角度)

該項目專利申請第一稿由研究人員撰寫，標題為“轉基因魚應用於雌激素物質檢測”
遞交至知識轉移處申請專利基金。

2010年
3月

專利第二稿(專利代理人角度)及建檔

摘取專利第一稿中可用於專利申請的發明部分，由專利代理人修改完善形成專利第二稿，並遞交至美國專利商標局建檔。



**US Patent No.
12/730,956**

專利審批

2012年初，一系列“顯而易見性駁回”不期而至，基於科學依據起草了答辯材料。
於2012年6月起草了對非終審意見的回復。



2012年
11月

待批准專利

2012年11月12日收到美國專利商標局發出的“核准通知”。



一個新故事的開始.....

該項目技術成功轉移至水中銀(國際)生物技術有限公司。海洋污染國家重點實驗室博士後(陳雪平博士)任首席技術官。培養城大畢業生企業家並創造就業機會。



Vitargent (International)
Biotechnology Co. Ltd.



EQSPA E-2011

by Dr. Kenneth Mei Yee LEUNG
梁美儀 博士

The SKLMP co-sponsored and supported Dr. Kenneth Leung in organizing the First International Conference on Deriving Environmental Quality Standards for the Protection of Aquatic Ecosystems (EQSPA E-2011) which was successfully held at The University of Hong Kong during 3-7 December 2011. With an Environment and Conservation Fund's Conference Grant, Dr. Leung brought leading experts in the field of environmental quality standard (EQS) derivation and application from North America, Europe, Australia, New Zealand, ASEAN countries, Korea and Mainland China to share their expertise and experience at this conference. The conference provided an essential platform for international, regional and local experts from government, academia and the environmental sector to exchange their knowledge and experience in EQS derivation and water quality management, build a professional network and establish research collaboration. Two training workshops were also provided on 3 December 2011 for local environmental practitioners and postgraduate students to acquire the knowledge and skills in derivation and application of water or sediment quality guidelines. There were 84 and >150 delegates from over 14 countries participating in one of the two workshops and the conference, respectively. The outcomes of the syndicate discussions and selected papers will be published in a special issue of Environmental Science and Pollution Research in 2013.

2011年12月3號至7號，海洋污染國家重點實驗室聯合贊助並協助Dr.Leung在香港大學成功舉辦了第一屆“水環境生態系統保護的環境質量標準制定”國際會議(EQSPA E-2011)。在環境及自然保育基金會項目的支持下，Dr.Leung通過此次會議將來自於全球各地(北美、歐洲、澳大利亞、新西蘭、東盟、韓國、中國大陸等地)的專家聚集起來，匯集了他們在環境質量標準的制定及應用領域的專業知識和經驗。該會議為來自於全球各地的政府、學術機構以及環保部門的專家提供了一個重要的學術交流平台，並在此次會議建立了專業的學術網絡及相關的研究合作。2011年12月3日，本實驗室還舉辦了兩次培訓班，為本地環境工作者以及碩士研究生提供了關於水與底泥質量准則的運用與技能培訓。兩次活動分別有84個和150個來自14個國家的代表參加。會議的討論成果和優秀文章將在環境科學與污染研究特刊上發表。

International Conference on Deriving Environmental Quality Standards for the Protection of Aquatic Ecosystems (EQSPA E - 2011)

推導保護水生生態環境質量標準國際研討會(EQSPA E-2011)

Conference Period: 3-7 December 2011



Summer School on Emerging Environmental Problems and Risk Assessment

2012年暑假學校“新興環境問題與風險評價”

Dr. Doris Wai Ting AU
區慧婷 博士

The Summer School series on Environmental Science was jointly organized by Xiamen University, City University of Hong Kong, San Diego State University and Peking University. The Summer School was designed not only for university postgraduate students, but it was also relevant and applicable for professionals working in the area of marine and environmental science.

The State Key Laboratory of Marine Environmental Science (MEL) has organized several summer schools with its partner institutions in previous years which were well attended by over 300 participants and very encouraging feedback was received. The themes of previous summer schools were “EIA and Ecotoxicology” in 2008, “EIA and Eutrophication (HAB)” in 2009 and “Multidisciplinary Research of Geo-Bio-Chemical Interactions in the Ocean and at the Seafloor” in 2011. This year we continue our mission to provide participants with quality learning, key concepts and current updates in various aspects of environmental science.

The course has trained a total of 64 participants selected from 6 countries representing 36 institutions.

環境科學夏季課程系列由廈門大學，香港大學，聖地亞哥州立大學和北京大學聯合舉辦，此課程不僅僅只面向於碩士研究生，而且也面向從事環境科學研究的其他科研工作者。

近海海洋環境科學國家重點實驗室在過去的幾年聯合其合作機構舉辦了多次夏季課程，共有300多人參與，獲得了一致好評。過去幾年的夏季課程主題分別是2008年舉辦的“酶免疫分析和生態毒理學”，2009年舉辦的“酶免疫分析和富營養化”以及2011年的“大洋及其海底的地球生物化學交互作用多學科研究”。本年度，我們繼續在環境科學的不同領域為課程參與者提供高質量的培訓以及前沿動態。

此課程已培訓了64位科研工作者，他們分別來自於六個不同國家，三十六個不同科研機構。



Summer School on Emerging environmental problems and risk assessment

July 2-13, 2012 Xiamen, China

The SKLMP Dive Team

「中華潛龍」

The SKLMP Dive Team was established on 21st Sept 2012. Under the Spin Kid Project scheme, 10 researchers were trained and became certified divers under the supervision of Dr WAI Tak Cheung. Their first research mission was to conduct the "Reef Check 2012" of the SKLMP in Sharp Island (South). Reef Check has been organized by the Hong Kong government for more than 15 years, and the data collected from different reef check teams are important for developing policies to protect the coral reef system. It is an activity that we should be proud of. Furthermore, Dr CHAN Lai, Leo and Dr WAI Tak Cheung led two SKLMP dive team members on a 14-day long sampling visit to Kiribati focused on coral reef research.

我室「中華潛龍」隊於九月二十一日正式成立，通過其「哪吒」計畫，十名研究人員及研究生成功考獲潛水執照，成為「中華潛龍」的一份子。香港珊瑚普查是「中華潛龍」隊的首項任務，採集的數據為香港政府提供了客觀依據，有助制定保育珊瑚政策，是一項香港人引以為傲的活動。

再者，陳荔博士及韋德祥博士於十月份帶領兩位首屆「中華潛龍」隊員遠赴沉沒的國度「基里巴斯共和國」，開展為期十四天的珊瑚礁研究。



SKLMP DIVE



**受訓潛水員
Certified Diver**

**資歷
Qualification**

**專業領域
Expertise**

CHAN Wing Hei
陳泳禧

City University of Hong Kong (MPhil)
香港城市大學 (哲學碩士)

Ecotoxicology,
Proteomics
環境毒理學, 蛋白學

KWOK Ying, Karen
郭盈

City University of Hong Kong (PhD)
香港城市大學 (哲學博士)

Analytical chemistry,
Environmental monitoring
分析化學, 環境檢測

LOI I Ha, Eva
呂綺霞

City University of Hong Kong (PhD)
香港城市大學 (哲學博士)

Analytical chemistry,
Environmental monitoring
分析化學, 環境檢測



MA Chui Ying, Teresa
馬翠盈

The University of Hong Kong (MPhil)
香港大學 (哲學碩士)

Ecology & Biodiversity
生態學及生物多樣性

MAK Yim Ling
麥艷玲

City University of Hong Kong (PhD)
香港城市大學 (哲學博士)

Analytical chemistry,
Food safety
分析化學, 食品安全

MAK Yiu Pan, Ben
麥耀彬

City University of Hong Kong (BSc)
香港城市大學 (科學學士)

Ecotoxicology, Risk Assessment
環境毒理學, 風險評估

PENG Xiao Ling
彭小玲

Baptist University of Hong Kong (PhD)
香港浸會大學 (哲學博士)

Statistics
統計學



WU Chun Yin
胡俊賢

Aquarium Specialist
水族專家

Aquarium architecture
水族建構

WU Qian
吳茜

China Pharmaceutical University (MPhil)
中國藥科大學 (哲學碩士)

Analytical chemistry,
Environmental monitoring
分析化學, 環境檢測

ZHU Bing Qing
朱冰清

City University of Hong Kong (PhD)
香港城市大學 (哲學博士)

Analytical chemistry,
Environmental monitoring
分析化學, 環境檢測



Exchanges and Visits

交流與訪問

3 OCT 2012



On 3rd October, 2012, Dr. Leo Chan and Dr. Maggie Mak visited the Research Centre for Harmful Algae and Aquatic Environment. In the meeting, the division of work was determined related to studying ciguatoxin as well as establishing a standard process for algal cultivation in cooperating laboratories through the training of researchers. The cooperative intention to apply for a 973 Program launched by Director Lui, to include Tsinghua University, City University of Hong Kong and the National Research Institute of Chinese Medicine was also proposed.

實驗室副主任陳荔博士及麥艷玲博士訪問了暨南大學赤潮與水環境研究中心，各方經過深入的學術交流，確定了未來在雪卡毒素研究上的分工，以及明確通過對研究人員的培訓，建立合作實驗室間藻類培養的標準化程序。會議上還提出了以呂頌輝主任牽頭，聯合清華大學、香港城市大學、台灣國立中國醫藥研究所共同申請973國家重點科研項目的合作意向。

18 SEP 2012



We invited Dr. Stephen Jones from Resolution Optics Inc. to the City University of Hong Kong, College of Life Science of Shenzhen University and Graduate School at Shenzhen, Tsinghua University for an academic exchange and to give us lectures. Dr. Stephen Jones gave a presentation about the application of digital in-line holographic microscopy (DIHM) in oceans and lakes and we discussed possible future cooperation. Through this visit, we hope that we could strengthen the academic exchanges between our

Laboratory and the research institutions in Shenzhen, leading to more cooperation in the future.

邀請Resolution Optics Inc.副總裁Dr. Stephen Jones到香港城市大學及深圳大學生命科學學院、清華大學深圳研究生院作探訪交流。Dr. Stephen Jones在講座中向我們介紹了可潛式數碼同軸全息顯微鏡的應用和技術優勢，又與實驗室副主任陳荔博士等探討了未來可能的合作方向與領域。此次訪深交流會加強了實驗室與深圳海洋科學研究相關科研機構之間的學術交流，通過相互瞭解、互通有無為未來的進一步合作打下了基礎。

7 MAY 2012



On 7th May, Prof Lam with five other researchers visited Peking University Shenzhen Graduate School. They were heartily welcomed by Deputy Dean Prof Luan, Prof Geng who is Dean of School of Environment and Energy, Prof Wang and Prof He. They exchanged ideas about their research work and the possibility of cooperation. At the end of this academic visit, it was agreed to cooperate with each other in this research field in order to create a better chance for development.

實驗室主任林群聲教授等一行六人訪問北京大學深圳研究生院，與汪光義教授和何凌燕教授分別在瞭解環境與健康和新能源開發利用等領域進行深入的交流，雙方都希望進一步加強在科研領域的合作，實現優勢互補，共謀發展。

In addition, the laboratory also received delegations from different local and overseas universities, academic institutions, governments and enterprises as outlined below. Through these visits, the community can recognize the kinds of marine environmental pollution problems we are facing and their awareness of environmental protection enhanced.

另外，實驗室也接待了國內外不同大學學術機構、政府、企業單位的訪問代表團，向他們介紹實驗室的組成、主要研究方向、及未來的發展規劃。透過探訪活動，讓更多的人識認到目前社會面對的各種海洋環境污染問題，及提升公民環境保護意識。

部份曾接待過的大學學術機構、政府、企業單位代表團

2012

15 Dec	Vice president and regional manager of PADI Asia Pacific 國際專業潛水教練協會的副總裁和亞洲太平洋區域經理
04 Dec	Prof Luis Echegoyen, Chair Professor, Department of Chemistry, University of Texas 德克薩斯州立大學化學系講座教授
28 Nov	Distinguished Chinese Scientists Lecture Series 當代傑出華人科學家公開講座講者
27 Nov	Beijing Institute of Technology 北京理工大學胡海岩校長
21 Nov	Research Centre of Environmental Engineering and Management, Graduate School at Shenzhen, Tsinghua University 清華大學深圳研究生環境工程與管理研究中心
18 Oct	ASU team of Global Institute of Sustainability 亞利桑那州立大學隊的全球可持續發展研究所
25 Sep	National Natural Science Fund of China 國家自然科學基金
17 Sep	Changchun District United Front Work Department 長春市委統戰部
30 Jul	Josai University Education Publisher 城西大學教育集團
20 Jul	The Affiliated High School of South China Normal University & Guangdong Experimental High School 華南師範大學附屬中學及廣東省實驗中學
09 Jul	National Tsing Hua University & Peking University 國立清華大學及北京清華大學學生交流團
20 Jun	Office of Naval Research Global (ORNG) 全球海軍研究辦公室
26-27 Aug	Exchange program of College of Ocean & Earth Science, Xiamen University 廈門大學海洋與地球學院學生交流團
25 Aug	Wuxi Municipal Government 無錫市政府
10 Aug	Kunshan Municipal Government 崑山市政府
23-25 Feb	National Museum of Marine Biology & Aquarium 國立海洋生物博物館
23 Feb	Shenzhen Water Group 深圳水務集團

List of Young Visiting Scholars

Visiting Scholars 訪問學者	Research Institution 研究單位
Mr. Yu Nan Yang 于南洋	Nanjing University 南京大學
Dr. Shi Wei 史薇	Nanjing University 南京大學
Dr. Li Yongyu 李永玉	Xiamen University 廈門大學
Dr. Liu Min 劉敏	Xiamen University 廈門大學
Dr. Kuang Hua 匡華	Jiangnan University 江南大學
Dr. Lin Bo Kun 林伯坤	Shantou University 汕頭大學
Dr. Peng Xiao Ling 彭小玲	BNU-HKBU United International College (UIC) 北京師範大學-香港浸會大學聯合國際學院
Mr. Song Zhan Feng 宋戰峰	Shenzhen Academy of Environmental Science 深圳市環境科學研究院
Dr. Wang Tai 黃泰	Shenzhen Academy of Environmental Science 深圳市環境科學研究院

Summary

The SKLMP has fulfilled several large collaboration projects in 2012, including (1) collaboration with Resolution Optics Inc. to explore the development of the Submersible Digital In-line Holographic Microscope (S-DIHM); (2) collaboration with the Research Center for Harmful Algae and Aquatic Environment, Jinan University on the isolation of benthic dinoflagellates in the coral reef system of the Republic of Kiribati; (3) collaboration with Shenzhen Academy of Environmental Sciences on the agreements "Determination of antibiotic compounds in Shenzhen source water" and "Training Program for Quantification of Antibiotics in Water Samples"; collaboration with Shantou University on "Environmental Marine Monitoring on the Control of the Economic Belt of the Estuary of the Eastern City and the Construction Period of Comprehensive Development Project in Shantou"; (4) became the leading institution for the establishment of the Shenzhen Marine Research & Technology Consortium (SMART); collaboration with Hong Kong Baptist University and the Graduate School at Shenzhen, Tsinghua University on the development of a 3D structural image observation system for coral reefs; (6) collaboration with the Graduate School at Shenzhen, Tsinghua University on the study of the trophic transfer of ciguatoxins; (7) collaboration with Shenzhen University on the development of a ciguatoxin rapid screening kit, etc. The above achievements, academic exchanges and visits mark the importance of the development of the SKLMP.

總結

2012年實現了多項合作項目，包括與Resolution Optics Inc.簽訂了開發潛水型同軸全息顯微鏡的協議；與暨南大學赤潮與水環境研究中心共同分離基里巴斯的藻種樣品；與深圳環境科學研究院簽訂《深圳水環境樣品中抗生素檢測方法開發與含量檢測》及《水環境樣品中抗生素檢測技術人員培訓》合作合同；與汕頭大學簽訂《汕頭市東部城市經濟帶河口治理及綜合開發項目施工期海洋環境跟蹤監察》合同；作為主要成員單位協助成立深圳海洋應用研究與技術聯盟 Shenzhen Marine Research & Technology Consortium (SMART)；與香港浸會大學、清華大學深圳研究生院共同開發珊瑚礁3D結構影像觀測系統；與清華大學深圳研究生院正在開展對西加毒素在海洋食物鏈傳遞的研究；與深圳大學簽署關於研制西加毒素快速檢測試劑盒的合作協議等等。對外交流獲取了多項實質性成果，顯示了學術交流對SKLMP發展的重要意義。

Social Education & Community Service

公眾教育與社會服務

The Spin Kid 哪吒計劃

The Spin Kid

哪吒計劃



“To ignite passion for the protection of the marine environment and pursue interest in marine science research”

傳播保護海洋環境資源火種
點燃追求海洋科學的興趣

The State Key Laboratory in Marine Pollution (SKLMP), City University of Hong Kong, besides working actively at the frontier of marine pollution research has also discovered the popularity of scuba diving during recent years, and it would be one of the most appropriate media to promote the key concepts in marine conservation. We hope that through scuba diving, the general public can experience the greatness of the ocean, and see how they could contribute to the protection and sustainable development of the marine environment.

In Chinese mythology, the Spin Kid is a well known character. When he was young, he was just like many teenagers nowadays, spoilt and rebellious. However, after he found some cool and powerful weapons, he decided to change. He started to help the underprivileged groups, fight against evil and protect justice. Finally, he became a hero in the eyes of Chinese people.

Through the Spin Kid project, scuba diving could be one of the platforms for the teenagers to find their strengths and inner-self to face the continuous challenges ahead. Furthermore, these ocean lovers would create an excellent army for marine pollution research and marine conservation.

香港城市大學海洋污染國家重點實驗室 (SKLMP) 除了開展與海洋污染有關的前沿研究外，有見近年來潛水運動日趨普及，而潛水業界及潛水愛好者又是海洋科普教育的最佳媒介，因此期望通過這種市民喜聞樂見的形式，共同為海洋環境保護和社會經濟可持續發展做出更大貢獻。

「哪吒」是中國家喻戶曉的神話人物，他像很多時下年輕人一樣，受寵壞及叛逆。在找到又酷又炫的風火輪、乾坤圈和混天綾後，洗心革面，為人間正義打拼，與邪惡力量抗衡，抑強扶弱，推翻不懂苦民所苦、欺壓人民的公害禍首，守護眾生，直到業盡情空。

「哪吒」計劃是希望透過潛水運動，為這些青少年哪吒提供一個揮灑青春精力的舞臺，幫助他們不停找尋挑戰，釋放潛藏而驚人的力量，從而為海洋研究培育潛在的優秀生力軍。



Clean Up Plastic Beads 膠珠清理

During July, Hong Kong was lashed by the strong typhoon Vicente. A cargo containing plastic raw material was damaged during the typhoon resulting in 150 tons of plastic pellets being spread out in the sea and consequently flushed to the shore. In addition to the help of the public to clean-up the beaches, the SKLMP sought another way to contribute in this issue. We collaborated with the diving team of the AFCD, Dr CHEUNG K. C. (IVE) and Mr AU Siu Kin, Stephen (Diving Adventure) and organized a voluntary team to investigate and record the contamination by plastic pellets in those affected waters which have significant conservation value, in order to relieve the doubts of the public. With help from the AFCD, IVE, SKLMP and Diving Adventure divers, 1.5 km of coastline in Sham Wan and Tung O Wan was successfully checked. Two of the shallow areas (<5m) and deep water areas were found to have no plastic pellets. This result showed that there was no deposition of plastic pellets in the affected waters.

在2012年7月期間，香港受到颱風吹襲，有貨櫃於颱風



期間被吹倒海中，導致150噸膠珠散落海面再沖上沙灘，引發市民廣泛關注。除了依賴社會大眾在沙灘上清理膠珠外，SKLMP希望從不同途徑為這件事出一分力。因此，我們馬上聯絡漁農自然護理署周永權先生、香港專業教育學院張桂宗博士及潛水歷險會區紹堅先生組織起隊伍前往保護價值高並最有可能受膠粒污染的主要沿海區域，例如在珊瑚區進行水底調查及記錄。在漁護署、香港專業教育學院、SKLMP和潛水歷險會義務潛水員的幫助下，我們在深灣及東澳灣已經檢查了約1.5公里長的海岸線。在兩個較淺（<5m）和較深的水域，我們並沒有發現任何膠珠，這表明沒有大量的膠珠沉積於水底。



Chinachem Charity Walk 「華懋慈善行2012」

The SKLMP would like to make more contributions to environmental protection, and so, besides working on research concerning environmental monitoring, we decided to raise public awareness on conservation and sustainable development of our society by supporting different green activities. The Chinachem Charity Walk 2012 was one of them. On 4th November, 2012, we participated in Hong Kong's largest-ever eco-tour Chinachem Charity Walk 2012 held at Tai Tam Reservoir, Hong Kong Island, organized by the ChinaChem Group. The funds raised from this event were used for full support of the beneficiary of the Sai Kung District Community Center "Green Life Education" project, which supported the organization of a series of seminars, workshops and eco-tours on environmental protection, to provide opportunities for the general public to understand the ecosystem and stay close to the natural environment.

我們希望為環境保護做出更多的貢獻，除了透過實驗研究結果引起公眾對環境的關注外，我們相信透過參與一些綠色活動，可以向大眾傳揚保育及可持續發展訊息，提高公眾保護自然環境的意識，所以我們支持「華懋慈善行2012」。2012年11月4日，我們參與了華懋集團於港島大潭水塘段舉行之全港最大型單日生態遊「華懋慈善行2012」，是次活動籌得款項全數支持受惠機構西貢區社區中心「拉闊綠色力量」計劃，用以舉辦一系列的環保講座、工作坊及導賞活動，為公眾提供接觸和了解生態保育的機會。



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